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Study Progress in Riverine Phytoplankton and its Use as Bio-Indicator – a Review

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Abstract

The value of algae as bio-indicators has already been recognized in the mid of 19th century, however, little attention has been paid to the application of phytoplankton in ecological evaluation of rivers. In this review, we found that studies of phytoplankton showed a long-term increasing trend from 1961 to 2014. However, most of these studies were carried out in oceans, coastal areas, gulfs, lakes and reservoirs, while very few of them (14%) focused on riverine phytoplankton. As well as modeling studies, the utilizations of riverine phytoplankton as bio-indicator are still poorly investigated and the available few studies were those mainly published after the year of 2000. Therefore, we describe 28 algal indices of riverine phytoplankton potentially used for bioassessment, which belong to community index, growth form, diversity index and biotic index. We also elucidate the calculation and classification methods of 5 common indices proposed in 1950s (Shannon's diversity index, saprobity index) and nowadays (trophic diatom index, Q index and phytoplankton index). Finally, four future directions and applications of riverine phytoplankton research were discussed and proposed: 1) standardization of sampling methods, 2) relations with environmental factors, 3) bio-indication and 4) modeling and predicting dynamics of riverine phytoplankton.

Keywords: Algal metrics; Bio-assessment; Environmental variables; Modeling; Riverine phytoplankton

Introduction

Phytoplankton (mainly planktonic algae), together with benthic algae and macrophytes, constitute the autochthonous primary producers in aquatic ecosystems and form part of the basis of the food web in terms of energy and material input [1]. Due to their short life cycle, planktonic algae respond quickly to environmental changes and are thus a valuable indicator of water quality [2-5] with the aim of effective water resources management and water pollution control. The value of algae as bio-monitor and bio-indicator for human disturbances (e.g. point and diffuse pressures, etc.) has already been recognized in the mid 19th century: the first concept which has been developed was the system of saprobity, which was mainly designed for organic pollution of streams and rivers [6,7]. Moreover, unlike fish and macroinvertebrates, algal communities are usually present before disturbance and generally persist in some form after disturbances. Therefore, application of algal indicators to assess rivers is increasing [8-11]. Recently, diatoms were used as a tracer of water source and hydrological connectivity in the mountainous Attert catchment [12]. The preliminary result of [13] showed that diatoms can help to detect the onset/cessation of surface runoff. Suggested for a meso-scale catchment [14,15] suggested that diatoms could reflect the geographic origin of stream water at the catchment outlet. However, compared to the numerous investigations in lentic water bodies (e.g. oceans, gulfs, lakes and reservoirs) little attention has been paid to the application of the phytoplankton in ecological evaluation of rivers [8].

In this study, by reviewing international scientific literatures, we described the long-term trends of phytoplankton research from past to

2014, with emphasis on riverine phytoplankton. We then summarized the algal indices widely used now for riverine bio-assessment. Based on our reviewed literatures, we finally proposed four possible future directions and applications of riverine phytoplankton research.

Methods and Summary of Literature Reviewed

We searched original papers about phytoplankton by means of Science Direct: http://www.sciencedirect.com/ and Springer link: http://www.springerlink.com/ to inspect the long-term publication trends from 1961 (very few publications before 1960) to 2014 (access on 15th May, 2014). Publications with an article title of "phytoplankton" or "potamoplankton" were searched. The results showed that most of these studies were widely carried out in oceans, coastal areas, gulfs, lakes and reservoirs, and demonstrated an increasing publication trend by the two databases (Figure 1).

Based on the previous searching results, we conducted an additional search to estimate the proportion of riverine phytoplankton studies. We took three journals for in depth analysis, which were "J. Plankt. Res. (JPR)", "Ecol. Indic. (EI)" and "Ecol. Model. (EM)", respectively. For the period reviewed, we examined a total of 771 publications (with an article title of "phytoplankton" or "potamoplankton"), and only 14.1% (109 studies) of them focused on riverine phytoplankton (Figure 2). The proportions of riverine phytoplankton studies compared to other surveys of phytoplankton from JPR, EI and EM were 15.0%, 9.6% and 13.5%, respectively (Figure 2).

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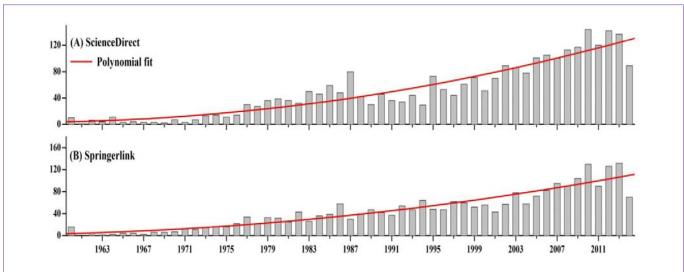
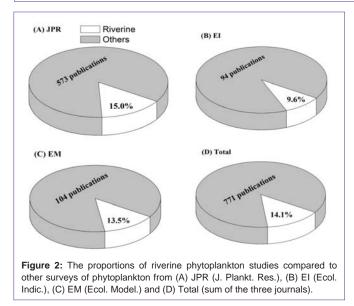


Figure 1: Long-term publication trends of phytoplankton studies searched by (A) ScienceDirect and (B) Springerlink (with article title of "phytoplankton" or "potamoplankton"). Red bold lines are study trends made by polynomial fit.



Furthermore, we classified these 109 studies into four major categories: I) primary studies (including taxonomic composition, temporal and spatial distribution, bio-volume, sampling methods, etc.), II) relations with abiotic factors, III) bio-indication and IV) modeling. We examined studies in five year increments (Figure 3). Overall, the number of riverine phytoplankton publications increased from 5 (first 5-y, 1981-1985) to 34 (the last 5-y, 2006-2010), except for a small number of 1991-1995 and 2011-2014. Most studies of them so far, however, were primary studies with a percentage of 68.8% (75 out of 109). There were only 17 publications (15.6%) studying "relations with abiotic factors", but an increasing trend was found from 1986-1990 (1 publication) to 2006-2010 (6 publications). As well as modeling studies, the utilization of riverine phytoplankton as bio-indicator was still poorly investigated and available studies were those mainly published after the year of 2000 (Figure 3).

Review of Algal Indices

Monitoring of the naturally occurring algal communities in

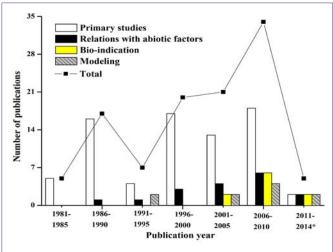


Figure 3: Long-term publication trends of riverine phytoplankton studies in J. Plankt. Res. (JPR), Ecol. Indic. (EI) and Ecol. Model. (EM) was grouped in 5-y increments. Studies were classified into primary studies, relations with abiotic factors, bio-indication and modeling (see text). The total number of publications was also indicated for each period. * access on 15th May 2014.

rivers provide data on species composition, number, diversity or quantitative occurrence of the phytoplankton. However, experts of administrative institutions who are responsible for water quality management need simple numerical values rather than species lists or scientific evaluation of the assemblages [8]. In this section, therefore, we summarized 28 algal indices of riverine phytoplankton with potential to be used for bio-assessment (Table 1), which belong to community index, growth form, diversity index and biotic index. We emphasized the importance of algal bio-volume, and discussed calculation and classification methods of five common indices proposed in 1950s (Shannon's diversity index (H'), saprobity index (SaI)) and recent years (trophic diatom index (TDI), Q index (QI) and phytoplankton index (PhI)).

Algal bio-volume and biomass

Algal bio-volume was commonly calculated to assess the relative abundance (as biomass or carbon) of co-occurring algae varying in

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Index (R)	Abbreviation	Description	Reference			
Community indices						
Total algal biomass (V)	TAB	Measures total algal biomass per liter, and is estimated based on multiplication of density data with volume (closest geometric form) supposing specific gravity of 1.00 g cm ⁻³	[16]			
Total algal density (V)	TAD	Measures algal numbers per liter	-			
Chlorophyll a Content (V)	Chl a	Measures total algal biomass	[103]			
Ash-free dry-weight (V)	AFDW		[104]			
Autotrophic index (+)	AI	Measures trophic status (autotrophic vs. heterotrophic) in rivers	[105]			
Growth form indices						
%benthic taxa (V)	BeT					
%mobile taxa (V)	MoT		Algal Data Analysis System (ADAS) using an attribute file of published			
%unattached taxa (V)	UnT		values [23,27,28]			
Diversity indices						
Shannon's diversity index (-)	H'		[21]			
Menhinick index (-)	Mel		[105]			
Pielou's evenness index1 (-)	J1		[107]			
Sheldon's evenness index2 (-)	J2		[108]			
Evenness index3 (-)	J3					
Evenness index4 (-)	J4	Measures ecological diversity in the community	[109]			
Evenness index5 (-)	J5					
Camargo's evenness index6 (-)	J6		[110]			
Simpson's dominance index (V)	D		[109]			
Margalef's diversity index (V)	М		[111]			
Species richness (V)	S	Number of specific or sub-specific taxa	-			
Biotic indices						
Trophic diatom index (+)	TDI	Designed to detect eutrophication	[33]			
Saprobity index (+)	Sal	Measures saprobic status of the water	[26]			
Q index (-)	QI	A new evaluation technique of potamoplankon for the assessment of the ecological status of rivers	[8]			
Phytoplankton index (+)	PhI					
Assessment value of total pigment (+)	A-value					
Pennales-Index (-)	Pel					
Chlorophyte-Index (+)	Chl	New German approach to assess running waters by phytoplankton community [39]				
Cyanobacteria-Index (+)	Cyl					
Trophic index of potamoplankton taxa (+)	TIP	ommunity index) were colculated based on both coll density and his volume data				

Table 1: Algal indices, their descriptions, and their expected response (R) to deterioration of water quality. + = indices expected to increase with deterioration, - = indices expected to decrease with deterioration, V = variable response.

Note: all the algal indices (except for Community index) were calculated based on both cell density and bio-volume data.

shape and / or size [16]. We highlighted algal bio-volume because it was basis for calculations of many other indices, such as growth form index, diversity index, biotic index (Table 1). To describe the whole community, as many algal indices as possible should be calculated based on not only cell density, relative cell density, entity density (numbers of colonies, filaments or free-living cells), but also bio-volume or relative bio-volume [11].

In general, the calculation of bio-volume is based on geometric approximations. The geometric shapes used for bio-volume determination should be as close to the real shape of the organism but at the same time easily discernible and conveniently measurable during routine analysis [17]. [16] recommended a standard set of 20 geometric shapes for over 850 genera and provided equations to be used for accurate estimates of cell volume for phytoplankton and micro benthic algae from linear dimensions measured microscopically. Based on this earlier work, [18] proposed a set of 31 geometric shapes and equations of 284 phytoplankton genera for routine analysis in China Sea waters. Furthermore, a set of geometric models was suggested by [19] for calculating the cell bio-volumes of 201 phytoplankton genera found in transitional water ecosystems of the Mediterranean Ecoregion. The equations were designed to minimize the effort of microscopic measurements. [11] provided geometric shapes for 303 taxa in three mid-continent US great rivers (the Upper Mississippi, Missouri and Ohio). After calculating the algal bio-volume, biomass of each algal taxon was estimated based on multiplication of cell density data with volume (closest geometric form) supposing specific gravity of 1.00 g cm⁻³ [16,20]. Total algal biomass (TAB) is calculated by summing up the biomass of each tax on.

Shannon's diversity index (H') [21]

Shannon's diversity index (H'), based on information theory, is one of several diversity indices used to measure diversity in categorical data. The advantage of this index is that it takes into account the number of species and the distribution of the species. The index is increased either by having additional unique species, or by having a greater species distribution. Its equation is:

$$H' = -\sum_{i=1}^{S} (P_i * \ln P_i)$$
 (Equation 1)

where:

H' = the Shannon's diversity index

P_i = proportion of all individuals in sample that belong to species i

S = total number of species in a sample

 $\Sigma =$ sum from species 1 to species S

H' was the most popular diversity index among ecologists [22], so values would more readily be interpreted and compared with other literature values. H' was expected to decrease with deterioration of water quality [23]. High values of H' would be representative of more diverse communities (namely good water quality). A community with only one species would have an H' value of 0, and if the species were evenly distributed among the S species then the H' value would be at a maximum. So the H' value allowed us to know not only the number of species but how the abundance of the species was distributed among all the species in the community. [24] suggested a relationship between H' and the pollution status of aquatic ecosystems and classified H' as:

>3.0 = "very good status (clean water)";

1.0-3.0 = "moderate status (moderately polluted)";

<1.0 = "bad status (heavily polluted)".

[25] modified the above mentioned classification by dividing '1.0-3.0' into two scales as:

2.0-3.0 = "good status (lightly polluted)";

1.0-2.0 = "moderate status (moderately polluted)".

Saprobity index (Sal) [26]

Saprobity index (SaI) is a weighted mean of the individual

saprobic value of each species "s" multiplied by their abundance "h" and divided by the total abundance:

$$SaI = \sum (h*s) / \sum (h)$$
 (Equation 2)

where "s" can take values between 1 for oligosaprobic, 2 for β -mesosaprobic, 3 for α -mesosaprobic, 4 for α -meso/polysaprobic and 5 for polysaprobic species according to [27,28]. SaI ranked from 0 to 5 and was characterized as:

<1.8 = "very good status (oligosaprobity)";

1.8-2.3 = "good status (β-mesosaprobity)";

2.3-2.8 = "moderate status (β - α -mesosaprobity)";

2.8-3.3 = "poor status (α-mesosaprobity)";

>3.3 = "bad status (polysaprobity)".

SaI was mainly designed for organic pollution of streams and rivers [7], and has been widely used as an indicator of water quality in reservoirs and rivers to date e.g. [10,29-32].

Trophic diatom index (TDI) [33]

The initial version of the TDI was derived empirically from graphs summarizing percent count vs. dissolved phosphorus concentrations for 86 taxa (genera plus key indicator species [33]). It produced values from 1 (low nutrient concentration) to 5 (very high nutrient concentration). However, to express a clear preference for an index that produces integer values over an extended numerical range, the TDI was therefore modified to 0 (low nutrient concentration) – 100 (very high nutrient concentration). This was achieved as follows:

$$TDI = (WMS * 25) - 25$$
 (Equation 3)

where:

TDI = trophic diatom index

WMS = weighted mean sensitivity, calculated using sensitivity and indicator values according to [34]:

$$WMS = \sum_{i=1}^{n} (a_i * s_i * v_i) / \sum_{i=1}^{n} (a_i * v_i)$$
 (Equation 4)

where:

WMS = weighted mean sensitivity

 $\boldsymbol{a}_i = proportion$ of all individuals in a sample that belong to species i

n = total number of species in a sample

 $s_i = pollution sensitivity (1-5) of species i$

 v_i = indicator values (1-3) of species i

TDI was originally designed for benthic algae, but has been employed for phytoplankton e.g. [4,10,35]. TDI was expected to increase with increasing eutrophication, ranked from 0 to 100, and was classified according to [27] as:

0-25 = "very good status (oligo-eutrophic)";

25-50 = "good status (meso-eutrophic)";

50-75 = "moderate status (eutrophic)";

75-100 = "poor status (hyper-eutrophic)".

Nevertheless, where there was heavy organic pollution, it was difficult to separate the effects of eutrophication from other effects. For this reason, the values of TDI are supplemented by an indication of the percentage pollution tolerant values (%PTV), which is calculated as the sum of values belonging to taxa generally regarded as particularly tolerant to organic pollution. According to [33], %PTV means different organic pollution state:

< 20% = "free of significant organic pollution";

21-40% = "some evidence of organic pollution";

41-60% = "organic pollution likely to contribute significantly to eutrophication of site";

>61% = "site is heavily contaminated with organic pollution".

Q index (QI) [8]

Based on the phytoplankton associations described for lakes [36,37], a new evaluation technique of potamoplankon for the assessment of the ecological status of rivers was proposed by [8]. To achieve an index, each species in the sample must be assigned to the appropriate functional group. Then the relative shares of each functional group are calculated. Relative shares are then multiplied by the factor number and the sum of these scores is the Q index (follow [37]).

$$QI = \sum_{i=1}^{n} (p_i F_i)$$
 (Equation 5)

where:

QI = Q index

 p_i = the relative share of the i-th functional group equal to n_i/N

 $n_i =$ the biomass of the i-th group

N = the total biomass

 F_i = the factor number (between 0-5).

The method is based on the functional group of algae represented in the potamoplankton and provides a single index number (Q), which has been tested on phytoplankton data of different rivers and proved to be more sensitive than the earlier used index (SaI) [8]. Thereby, the classifications of different functional groups were of great importance for such index and should be gathered from historical studies. Based on the list of the functional groups [36] and its updated version, the evaluation of the 37 functional groups of algae and factor number of each group were provided by [8]. Furthermore, [38] wrote a critical review with updates of the phytoplankton functional classification.

Theoretically, the maximum of QI is 5, while the minimum is 0, and is expected to decrease with decline of the ecological status of rivers. QI values for different water quality classes of proposed river types were summarized in Table 2.

Phytoplankton index (Phl) [39]

Phytoplankton index (PhI) is also a new approach to assess

Table 2: Proposed river types and Q index (QI) values for different water quality classes (modified from [8]).

Diversitives	Stream	Residence	QI				
River type	order*	time (day)	Excellent	Good	Moderate	Poor	Bad
Brooks							
and small	1-5	<2	5.00	4.95	4.85	4.75	<4.75
streams							
Streams	3-6	2-4	4.95	4.85	4.75	4.50	<4.50
Small rivers							
(lowland	4-7	4-8	4.75	4.50	4.00	3.50	<3.50
streams)							
Rivers	6-9	8-12	4.50	4.00	3.50	3.00	<3.00
Large rivers	7-10	12-16	4.00	3.50	3.00	2.50	<2.50
Very large rivers	>10	>16	3.50	3.00	2.50	2.00	<2.00

* Depending on local conditions.

running waters by phytoplankton introduced by [40] on behalf of the German Working Group on water issues of the Federal States and Federal Government (LAWA) to implement the European Water Framework Directive (WFD, EC 2000). It includes five sub-indices: Assessment value of total pigment (A-value_{totalpigment}), Pennales-Index (PeI), Chlorophyte-Index (ChI), Cyanobacteria-Index (CyI) and Trophic index of potamoplankton taxa (TIP). PhI is the mean of single results evaluated by the five sub-indices:

 $PhI = (A-value_{totalpigment} + PeI + ChI + CyI + TIP) / No. of used indices$

(Equation 6)

The scale of the PhI is in the range of 0.5-5.5:

<1.51 = "very good status";

- 1.51-2.50 = "good status";
- 2.51-3.50 = "moderate status";
- 3.51-4.50 = "low status";
- >4.50 = "bad status".

A-value_{totalpigment}:

Total pigment is estimated by phytoplankton Chl *a* values. A-value_{totalpigment} is calculated by specific formulas for different catchment types (Table 3).

PeI, ChI and CyI:

PeI, ChI and CyI are categorized into 5 scales (1-5) based on the percentages of pennales, chlorophyte and cyanobacteria, respectively. The scaling systems have been shown by [39]. For example, a percentage of pennales between 15 and 20 in catchment type 20.1 (large streams with high specific run-off > 10 ls⁻¹km⁻²) will lead to a PeI value of 2, which means 'good' state.

TIP:

The formula used to calculate TIP is:

$$TIP = -\sum_{i=1}^{n} (TI_i * GW_i * DW_i) / \sum_{i=1}^{n} (GW_i * DW_i)$$
(Equation 7)

where:

TIP = Trophic index of potamoplankton taxa

n = total number of species in a sample

Catchment type	Formulas from Chl a to A-value _{totalpigment}
10.1+20.1	1.8527*Ln(Chl a)-2.7981
15.1+17.1	1.9907*Ln(Chl a)-4.4749
15.2+17.2	1.9907*Ln(Chl a)-4.4749
9.2	1.9907*Ln(Chl a)-4.4749
10.2+20.2	1.8168*Ln(Chl a)-4.6772

Table 3: Formulas used for transformation between Chl a and A-value
A-value
Image: transformation of the transformation

10.1+20.1: large streams with high specific run-off (> 10 ls⁻¹km⁻²);

10.2+20.2: large streams with low specific run-off (< 10 ls⁻¹km⁻²);

15.1+17.1: lowland sandy streams with a catchment area of 1000-5000 km²; 15.2+17.2: lowland sandy streams with a catchment area of 1000-10000 km²; 9.2: Large high land streams with catchment area >5000-10000km²; A-value_{totalpigment} less than 0.5 is set equal to 0.5 and A-value_{totalpigment} larger than 5.5 is set equal to 5.5.

 $TI_i = type$ -specific index of species i

GW_i = weight factor of species i

 $\mathsf{DW}_i = \mathsf{proportion}$ of all individuals in sample that belong to species i

GW and TI values of indicator taxa in different catchment types were shown by [39].

Future Directions and Applications

Standardization of sampling methods

The investigation of the phytoplankton community has become an important part of the overall water quality monitoring [41], since reliable quantitative data on species composition are of primary importance for bio-assessment development. The precision obtained in the field may vary greatly due to the differences in sampling methods. There are two main methods for riverine phytoplankton sampling: 1) plankton nets 2) sedimentation protocols [1,42]. The two sampling protocols of phytoplankton were both widely used and had their advantages respectively. For example, the "plankton nets" protocol is labor saving, fast, easy to handle and can capture more rare species, but allows real nannoplankton to pass through its meshes [42,43]. It is thus a preferred method for clean water with low phytoplankton density. In contrast, sedimentation protocol is usually used in water bodies with high phytoplankton density (e.g. [45-49]). The above mentioned methods were both applied in the stream systems (e.g. [4,10,50-54]). Nevertheless, to our knowledge, except for [42], the influence of two sampling protocols on the outcome of bio-assessment in streams has not been investigated systematically yet. By comparison and unification between different sampling methods, phytoplankton data from various areas with different sampling protocols over multiple years could be merged to get a more comprehensive understanding of the ecological status by regional or country-wide assessment.

Relations with Environmental Factors

The response of phytoplankton to surrounding environmental factors has drawn particular attentions of present researches [55] and identification of the main factors controlling phytoplankton in a particular water body is essential for choosing an appropriate management strategy for the maintenance of a desired ecosystem state [56]. Distribution patterns of phytoplankton are strongly correlated with environmental factors [57]. Possible factors may

be physical, chemical, hydrological and biotic factors [5,45,54,58-63]. Unfortunately, there is no general consensus as to which factors regulate phytoplankton community in lotic habitats [64], and contributions of main environmental factors to phytoplankton variations are also unclear [54]. Understanding organism dynamics and resilience of river ecosystems in changing environmental factors (e.g. global changes) will greatly benefit the phytoplankton based bioassessment.

Bio-indication

The assessment of the ecological status of freshwater ecosystems is a key issue for many international laws such as the European Water Framework Directive (WFD) [65]. Many efforts have been devoted to the development of efficient tools to measure the ecological status of freshwater systems based on fish, macroinvertebrates, macrophytes and diatoms. Generally, all assessment methods can be sorted into three approaches. The first approach is based on the indicator species concept, the second one is based on the diversity of organisms, while third one applies multimetric approaches which are composed of several indices that can reduce information from individuals, population, community and ecosystem. More and more authors prefer the third approach because it integrates, condenses and summarizes biological data, and thus can reflect ecological status in a comprehensive manner [9,66-68]. One example is the multimetric Index of Biotic Integrity (IBI), originally developed by [69] which is the most common indicator of stream condition in use today. Many assessment methods based on IBI have been developed and used to date in several countries and regions (e.g. [4,23,65,68,70-Recently, a composite index at regional level named RIEI 76]). (Regional Index of Ecological Integrity) for sustainable management of natural resources was proposed by [77], and it is composed by not only "Physical Integrity", "Chemical Integrity", "Biological Integrity" but also by "Beauty", "Biodiversity" and "Ecosystem Health" indices. However, the riverine phytoplankton index of biotic integrity (PIBI) is rarely considered for river 'health' assessment [4]. This is in part because of the former understanding of riverine phytoplankton that algae found in rivers are believed to come from other sources than the rivers themselves - either from lentic waterbodies or the benthos [78]. With the confirmation of riverine phytoplankton, they should be combined with former assessing systems for rivers [54]. The selection of indicator depends on the stressor-type being assessed and the monitoring type. For example, according to [79], diatoms should be considered when the study focus is on nutrient enrichment and at small stream with relatively species-poor fish and macrophyte assemblages. However, in the case of hydromorphological degradation, fish, benthic macroinvertebrates and macrophytes should be considered instead of algae community.

Modeling and predicting riverine phytoplankton

Predicting freshwater phytoplankton dynamics is regarded as one of the important issues in the domain of river ecology and management [80]. The successful prediction by multi-variate processes either for short or long intervals of monitoring could drive the underlying mechanisms between phytoplankton and their environments. From the management decision-making point of view, [80] thought that if an accurate model for phytoplankton dynamics was reliable, then forecasting would be possible with only

phytoplankton data instead of monitoring a wide range of limnological changes, which usually has exorbitant costs. Many models, therefore, have been developed and used to simulate freshwater phytoplankton dynamics as well as aquatic insects in lakes and reservoirs, such as ANN (artificial neural networks) based models [5,81-84], PROTECH (Phytoplankton Responses To Environmental Change) [22,85-88], RIVERSTRAHLER [89,90], NPZ (nutrient/phytoplankton/ zooplankton) [91-93], process-based models [94,95]. However, only very few of them have been used for riverine phytoplankton simulation [5,80,96-98]. Therefore, the future studies should address to the followings: 1) comparing the performances of different models; 2) developing and testing of new comprehensive models that examine the impacts of multiple stressors on riverine phytoplankton. Besides, declining water quality worldwide and increasing progress in predictive potential of ecology and limnology greatly promote the development of the 'Ecohydrology' approach [99-101], which can provide means of integrating landscape hydrology with freshwater biology [102,112] and create an interdisciplinary background (ecological and hydrological) for the assessment and sustainable management of freshwater resources.

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