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“One Out–All Out” Principle in the Water Framework Directive 2000—A New Approach with Fuzzy Method on an Example of Greek Lakes

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Abstract: The “One Out–All Out” (OOAO) principle imposed by the WFD selects the worst ecological status assessed by different biological quality elements (BQEs). Since it is a precautionary rule that can lead to problems of underestimation of the overall status, its amendment has been a matter of debate for WFD 20⁺. The use of fuzzy methods that express the functional relationships between variables in ecology and management has been gaining more ground recently. Here is attempted the inclusion of a fuzzy regression among the frequently monitored BQE (phytoplankton) and the outcome of OOA application in six Greek lakes. The latter was determined by the comparison of four BQE indices in order to assess the extent to which BQEs might underpin the optimal/actual qualitative classification of a waterbody. This approach encompasses the uncertainty and the possibility to broaden the acceptable final EQR based on the character and status of each lake. We concluded that the fuzzy OOA is an approach that seems to allow a better understanding of the WFD implementation and case-specific evaluation, including the uncertainty in classification as an asset. Moreover, it offers a deeper understanding through self-learning processes based on the existing datasets.

Keywords: ecological quality assessment; OOA; fuzzy regression; biological quality elements; water framework directive fitness check; monitoring



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1. Introduction

The Water Framework Directive (WFD) [1], which was formally adopted in 2000, requires that all European Member States (MSs) have to assess the ecological status or potential of their inland, transitional, and coastal waterbodies. A key feature of the WFD is the use of the catchment-scale as the management unit, thereby allowing for the integrated effects of all pressures on waterbodies to be considered [2]. In particular, it introduces an innovative approach to manage and protect aquatic ecosystems in a holistic way rather than focusing only on specific aspects of water quality. Classification of ecological status or potential is based on different biological quality elements (BQEs) representing main ecosystem components, such as phytoplankton, benthic macroinvertebrates, other aquatic flora (macrophytes, phytobenthos), and fish [1].

Up to now, WFD complemented by its daughter and sister directives (as the Environmental Quality Standards Directive [3], the Groundwater Directive [4], the Flood Directive [5]), has achieved the set-up of a management framework across all European Union (EU) waterbodies, including lakes, mitigating their deterioration. However, at the same time, around 57% of rivers and 44% of lakes in EU have failed to achieve good ecological status or potential [6], while according to the WFD Fitness check [7], no substantial

progress in waterbodies' overall status has been made between the first and the second river basin management cycles. This means that waterbodies classified at less than good ecological status or potential must be restored, and further management responses should be applied to improve the overall system's health. These responses are presented under the term "Program of Measures, PoMs" in the River Basin Management Plans (RBMPs) and target the pressures and their drivers contributing to ecosystem dysfunction [8]. The overall system's classification is based on the "One Out–All Out" (OOAO) principle, meaning that the worst quality of any of the BQEs used in the assessment regulates the overall ecological status of a waterbody.

The majority of PoMs require significant cost while, often, the lack of financial resources for implementing a long list of measures can be a factor for not achieving better results. Thus, the diagnosis of the efficacy of each BQE in reflecting the actual status, which in turn guides the classification of each waterbody, is of high importance. Misclassification could lead either to over-precautionary results and imposition of restoration costs disproportionate or neglect of the need for strict measures to target pressures. It is acknowledged that it is more difficult to make progress visible in the WFD goals due to the OOAO principle [7]. Indeed, the OOAO principle was originally set up as a precautionary principle. The ecological basis for the debate on the usefulness of the OOAO principle has been clearly demonstrated by several researchers [9–11]. They state that the OOAO principle is well suited when different stressors are responsible for the degradation of the individual BQEs and further, that the tendency for misclassification increases with the BQEs uncertainty. Waterbodies are either affected by predominant pressures (e.g., organic pollution or eutrophication) or by multiple pressures, which can interact in additive, synergistic, or antagonistic ways (e.g., nutrient enrichment, hydromorphological degradation, toxic substances, overfishing) [12]. At the same time, the ecological knowledge of the response of different organism groups to pressures varies across Europe [13]. In addition, new pressures are appearing (e.g., microplastic pollution, pharmaceuticals, light and noise, freshwater salinization) [14].

The uncertainty of the BQEs and their significance for the ecological status classification have been addressed in many studies [15–20]. Currently, in relevant EU working groups, there are discussions on the use of a new set of indicators, which will be based on BQEs, and can serve as a supplementary guide to overall status or potential. For instance, in assessing the progress of ecological status, since the beginning of monitoring and the implementation of PoMs, the comparison of OOAO with other integrative methods in coastal and transitional waterbodies showed a low (18%) disagreement for the coastal and a much higher (58%) for the transitional [21]. This can be explained by the fact that one or more BQEs appear to have a low or a moderate correlation with various stressors, thus often describing similar inconsistencies [22]. Thus, the establishment of alternative combination rules is suggested [23], taking also into consideration the type I (i.e., a waterbody is below good status even if the waterbody in reality has good status) and type II (i.e., reducing the likelihood that a waterbody is classified as good status when, in reality, it is below good status) errors, as highlighted by Hering et al. [13].

Based on the above, Borja and Rodriguez [21] have suggested that MSs should avoid adopting this simplistic principle, although it may present an effective indication in the ecological status assessment. Thus, amendments on the application of OOAO or alternative approaches should be further considered. In a recent paper reporting the results of a survey on the suitable options of how the OOAO principle should be implemented, Carvalho et al. [11] show that the most popular responses opted for the boosting of selected BQEs and the emphasis on progress reporting. Therefore, the linkage of pressures with ecological status sets the basis for establishing management targets and restoration measures [24]. According to Carvalho et al. [11], the only state adopting management decisions dependent on the strongest evidence of BQE is the United Kingdom.

So far, the fundamental target of WFD implementation has been the development of assessment methods for all BQEs required in the WFD. For EU lakes, a wide variety

of assessment methods have been developed based on different BQEs that respond to specific pressures [15,25,26] or multiple pressures [27,28]. In Greece, until recently, there were gaps in the assessment of the ecological quality of lakes with WFD-compatible indices. Five national indices [29–33], applicable in the broader Mediterranean area, were developed for all BQEs reflecting the local peculiarities. It cannot be generally stated which BQEs underestimate or overestimate the actual status. It is empirically observed that the qualitative assessment depends more on the lake type, its special traits, and the type and intensity of pressures that is subject to.

Fuzzy logic in ecology has been gaining ground during the last two decades. The natural complexity can be reflected in fuzzy ecological modeling and scenarios simulation [34], while such “unconventional” data analyses can be applied for sustainability evaluation [35]. These approaches allow for overcoming the usual obstacle of precise quantification processes [36]. Moreover, fuzzy logic can assist in management planning and decision making by enhancing objectivity [37]. The fuzzy regression tool is useful in expressing functional relationships between variables, especially when the available dataset is insufficient [38]. Kitsikoudis et al. [39] employed fuzzy regression and set a fuzzy band to produce the lower and upper acceptable limits (left and right boundaries) of critical Shields stress. Thus, the ambiguity of selecting a threshold can be avoided, and a smoother transition to the actual state can be provided [40,41].

In this study, we first assessed the strictness of the OOA application based on different ecological assessment approaches and BQEs (phytoplankton, macrophytes, and benthic macroinvertebrates). Then, we examined a method utilizing fuzzy logic to express the functional relationships between two variables. Fuzzy regression was applied for phytoplankton (as the main BQE for lakes and more frequently monitored) and OOA that derived from all available BQE indices. This approach took place to assess the extent to which different BQEs can underpin the optimal/actual qualitative classification of a waterbody.

2. Materials and Methods

2.1. Study Area

As a case study, we used the BQEs from six natural lakes in northern and central Greece (Figure 1). These lakes are included in the National Water Monitoring Network, while one of them is transboundary (Doirani), shared with the Republic of North Macedonia. According to the Hellenic typology [29], these lakes belong to two types: (a) warm monomictic, deep, natural lakes with mean depth >9 m (type GR-DNL, 3 lakes) and (b) polymictic, shallow lakes with mean depth 3–9 m (type GR-SNL, 3 lakes). The studied lakes are characterized by a variability in their limnological and trophic state attributes (Table 1).

Table 1. Limnological characteristics of the studied lakes. OL, oligotrophic; MT, mesotrophic; ET, eutrophic.

Lake	Altitude (m.a.s.l.)	Mean Depth (m) ^a	Maximum Depth (m) ^a	Lake Area (km ²) ^a	Trophic Status
Doirani	142	4.5	5.5	32.4 *	ET
Lysimachia	16	3.5	7.7	13.0	ET
Ozeros	22	3.8	6.1	10.4	ET
Vegoritisi	510	26.1	52.4	47.4	MT-ET
Volvi	37	12.5	27.3	72.9	ET
Yliki	80	20.9	38.5	21.6	OL-MT

^a Data available from the national monitoring program implemented by the Greek Biotope-Wetland Centre (EKBY), * approximately 44% of the lake area being within the territory of Greece.

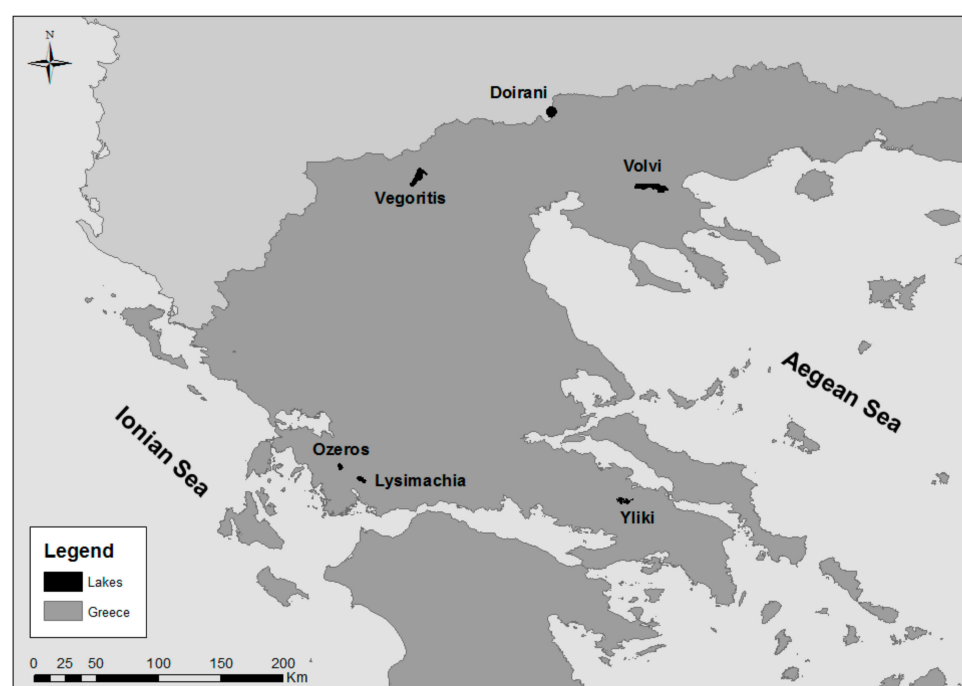


Figure 1. Map of the studied lakes.

2.2. Biological Assessment Methods

In the present study, each BQE was assessed by one index, except for benthic macroinvertebrates, as follows:

Phytoplankton: Hellenic Phytoplankton Assessment System (HeLPhy), which is composed of four metrics, i.e., chlorophyll-a, total biovolume, modified Nygaard index, and biovolume of cyanobacteria and responds to eutrophication [29].

Macrophytes: Hellenic Lake Macrophytes (HeLM), which consists of two metrics, i.e., trophic index and maximum depth of colonization and responds to eutrophication [30].

Benthic macroinvertebrates: Greek Lake Benthic Invertebrate Index (GLBiI), which is composed of three metrics, i.e., the number of taxa, the Simpson's diversity index in the profundal and sublittoral zones, and the relative contribution of Chironomidae (%) family in the profundal zone [32]; and Hellenic Lake Littoral Benthic Invertebrate Assessment System (HeLLBI), which is composed of three metrics, i.e., relative abundance of Odonata (% of abundance classes), average score per taxon, and Simpson Diversity Index [33]. The GLBiI responds to eutrophication, while HeLLBI to hydromorphological alterations and eutrophication.

Specifically, phytoplankton samplings took place during the warm period, in the pelagic zone, covering the euphotic zone water column ($2.5 \times$ Secchi disk depth) using a Nansen type sampler. Benthic macroinvertebrates from the profundal/sublittoral zones were sampled in spring and autumn, with the use of a grab [32]. Littoral benthic macroinvertebrates were sampled in spring using the three-minute kick/sweep method with standard hand net (500 μ m mesh size) [33]. Regarding macrophytes, sampling took place during the vegetative period, and the belt transect-mapping method was applied [30]. Finally, the dataset used comprised 46 lake years with phytoplankton EQRs, 11 with macrophyte, 6 with profundal/sublittoral zoobenthos, and 7 with littoral zoobenthos EQRs.

For each assessment method, as requested by the WFD, the Ecological Quality Ratio (EQR) was calculated as each index value was divided by the reference condition values, ranging from 0 (bad) to 1 (high). The reference values for GLBiI were estimated by the hindcasting procedure for each lake and do not correspond to actual reference sites' values, while for the rest of the indices, the reference values are presented in Appendix A (Table A1). The class boundaries were evenly spaced for all BQEs (i.e., high > 0.8 , good: (0.8–0.6), moderate: (0.6–0.4), poor: (0.4–0.2), bad ≤ 0.2), and thus, the comparison among

them was feasible. EQR values for each BQE were available at different time intervals (Table 2). The values for the application of the OOA principle were estimated for the two river basin management cycles (2012–2015 and 2016–2019). Analyses were applied using different combinations for integrating multiple BQEs at waterbody level. These combinations were:

- (a) *OOA*: the lowest EQR of BQEs was attributed for the whole waterbody;
- (b) *average*: the arithmetic average of the EQRs for all BQEs was calculated and rounded to the nearest class; and
- (c) *median*: the median of EQRs for all BQEs was calculated and rounded to the nearest class.

Table 2. Frequency of monitoring for each biological quality element. P, phytoplankton; M, macrophytes; B-L, littoral zoobenthos; B-SP, sublittoral/profundal zoobenthos.

Lake	1st Monitoring Period				2nd Monitoring Period			
	2012	2013	2014	2015	2016	2017	2018	2019
Doirani	P	P, M	P, B-SP	P	P, M	P		P
Lysimachia	P	P	P, M, B-SP	P, B-L	P	P, M	P, B-L	P
Ozeros	P	P	P, M, B-SP	P, B-L	P	P, B-L	P	P
Vegorit	P	P, M	P, B-SP	P	P, M	P	P, B-L	P
Volvi	P	P, M	P, B-SP	P	P, M	P, B-L		P
Yliki	P	P	P, M, B-SP	P, B-L	P	P	P	P

It is worth noting that we performed all the statistical analyses based on the above-mentioned BQEs since in Greece, for these monitoring periods, data for lakes are limited only to those. Moreover, we did not use diatoms since the national index based on diatoms is under development and not available yet.

2.3. The Proposed Fuzzy Regression Model

The data of the fuzzy regression can be either fuzzy or crisp. Usually, the data are rather crisp numbers, and thus, the uncertainty arises from the adopted fuzzy model, that is, from the fuzzy coefficients. In this work, we deal only with crisp data. Fuzzy regression analysis gives a fuzzy functional relationship between the dependent and independent variables [42]. In contrast to the statistical regression, fuzzy regression analysis has no error term, while the uncertainty is incorporated in the model by using fuzzy numbers [39,43].

From a computational point of view, according to the Tanaka [44] approach, the problem of fuzzy linear regression is finally formulated as a constrained optimization problem. In the case that symmetrical fuzzy triangular numbers are used, the problem is transformed into a linear programming problem [45,46]. The main fuzzy concept that is used in fuzzy linear regression is the fuzzy number. The general description of fuzzy numbers can be found in Klir and Yuan [47]. Usually, fuzzy triangular symmetrical numbers are used. Since in the examined problems there is a lack of data, the authors selected the simplest membership function, which is the fuzzy symmetrical triangular number, to avoid any overtraining. Hence, here, $\tilde{A}$ means a fuzzy triangular symmetrical number (Figure 1), which has the membership function (μ_A) presented below:

$$\mu_A(a) = \begin{cases} 1 - \frac{|a-a_i|}{c_i}, & \text{if } a_i - c_i \leq a \leq a_i + c_i \\ 0 & \text{otherwise} \end{cases} \quad (1)$$

Many times, the fuzzy triangular symmetrical numbers are denoted as $\tilde{A} = (a, c)$. The membership function expresses the degree according to which a member of the real axis (in general, the general set) belongs to the examined fuzzy number. The fuzzy numbers can be described as “approximately (or almost) a_i ” (e.g., almost one). The term a_i represents the central value and the term c the semi-width (Figure 2).

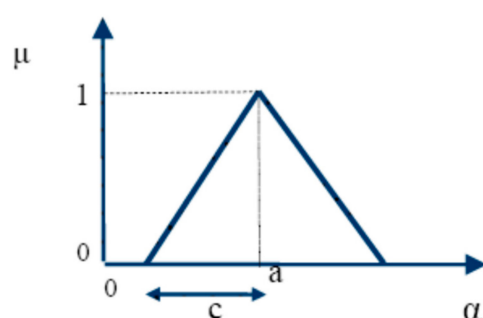


Figure 2. Fuzzy triangular symmetrical number “almost a”.

The fuzzy linear regression model proposed by Tanaka [44] and Tanaka et al. [48] has the following form in this study:

$$\tilde{Y}_j = \tilde{A}_0 + \tilde{A}_1 x_j \text{ with } j = 1, \dots, m \quad (2)$$

where x_j is the independent variable (here, the phytoplankton index), m is the number of data, and $\tilde{Y}_j$ is the fuzzy predicted value of the dependent variable (OOAO principle) considering the j th data. In other words, the fuzzy linear regression uses fuzzy numbers as coefficients instead of crisp numbers.

In addition, as it will be explained below, the fuzzy estimation of the dependent variable $\tilde{Y}_j$ must contain the observed data y_j (Figure 3). The observed data are crisp numbers (OOAO principle), whilst the fuzzy regression provides a fuzzy estimation of the OOAO.

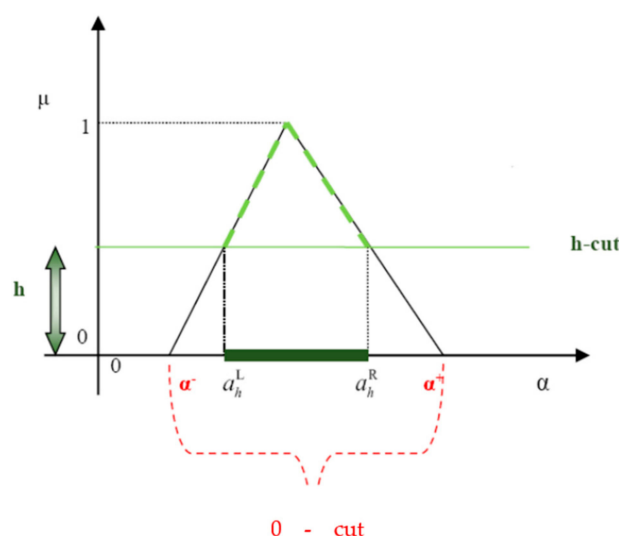


Figure 3. Triangular fuzzy number, its h-cut (is a crisp set), and zero-cut (α^- , α^+).

For instance, if the regression concludes to

$$\tilde{Y}_j = (-0.0457, 0.0457) + (1, 0)x_j, \quad (3)$$

this means that the main parameter that influences the OOAO is the phytoplankton index (x_j). However, an uncertainty arises from the constant term $(-0.0457, 0.0457)$. In contrast, if the model concludes to:

$$\tilde{Y}_j = (0.4237, 0.0137) + (0, 0)x_j \quad (4)$$

this means that the phytoplankton index has no influence in the evaluation of the OOAO. Then the outcome, that is, the OOAO (fuzzy) estimation, will be a fuzzy tube parallel to

xx' axis, and the estimation will be between $[a_0 - c_0, a_0 + c_0] = [0.41, 0.4374]$. The fuzzy constant term expresses the determinant influence of the other indices.

The h -cut set of the fuzzy number A (with $0 < h \leq 1$) is defined as follows (Figure 3):

$$[A]_h = \{\alpha | \mu_A(\alpha) \geq h, \alpha \in \mathbb{R}\} \quad (5)$$

where $\mathbb{R}$ is the real number axis.

Notice that the h -cut set is a crisp set determined from the fuzzy set according to a selected value of the membership function and, alternatively, a fuzzy set can be practically derived from a significant number of h -cut sets. In case of $h = 0$, the above definition of Equation (5) can be modified without the equality in order to describe the zero-cut [39].

The constraints express the concept of inclusion of the observed data (which are crisp numbers) within the produced fuzzy band. In general, the inclusion of a fuzzy set A into the fuzzy set B with the associated degree is defined as follows:

$$[A]_h \subseteq [B]_h \quad (6)$$

In our case, the set A is a crisp set whilst the set B is a fuzzy set. Therefore, for each point of data, the observed dependent variable y_j (OOAO) must be included into the produced h -cut of the fuzzy number $\tilde{Y}_j$ (blue line in Figure 4).

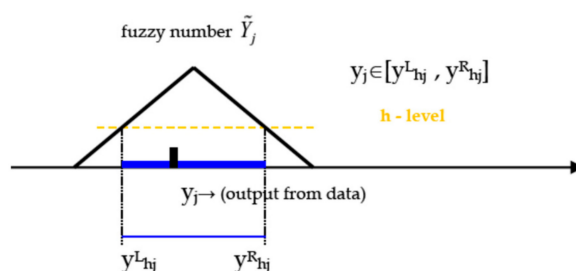


Figure 4. The concept of inclusion in the case of fuzzy linear regression.

According to the presented methodology, the selection of the used h -level is dependent on the quality of data. Another interesting point is that even if the initial definition of the inclusion property incorporates all the h -cuts, the definition of inclusion that is used in Equation (6) is based on a unique (pre-selected) level h instead of all the values.

In this study, since the data are crisp (for each individual data), the set A is only a crisp value (a point of data that must be included in the produced fuzzy band), and the fuzzy set B is a fuzzy triangular number. Hence, the inclusion constraints in our problem become [40,49]:

$$(a_1 x_j + a_0) - (1 - h)(c_1 |x_j| + c_0) \leq y_j \leq (a_1 x_j + a_0) + (1 - h)(c_1 |x_j| + c_0), j = 1, \dots, m \quad (7)$$

where a_1, a_0 are the centers of the coefficients that correspond to the independent variable (phytoplankton index) and the constant term correspondingly. The terms c_1, c_0 indicate the semi-widths of the coefficients that correspond to the independent variable and the constant term correspondingly. More analytically, the term $(a_1 x_j + a_0) - (1 - h)(c_1 |x_j| + c_0)$ expresses the left boundary of the h -cut of the fuzzy number $\tilde{Y}_j$, whilst the term $(a_1 x_j + a_0) + (1 - h)(c_1 |x_j| + c_0)$ expresses the right boundary of the h -cut of the fuzzy number $\tilde{Y}_j$.

In case that there is not enough data, as in our case, the minimization of the achieved fuzzy band is proposed. Indeed, it is obvious that if the produced fuzzy band has a large magnitude, then it will contain all data. However, no functional fuzzy relation would be produced.

Even if, initially, Tanaka [45] proposed the minimization of the semi-width of the fuzzy coefficients, finally, he proposed the sum of the semi-widths (fuzzy spreads) for the produced dependent variable for all the data:

$$\sum_{j=1}^m (y^R - y^L) = 2 \left(\sum_{j=1}^m (c_1 |x_j| + c_0) \right) = 2 \left(mc_0 + \sum_{j=1}^m c_1 |x_j| \right) \quad (8)$$

where y^R , y^L are the right-hand boundary and the left-hand boundary, respectively, of the fuzzy set, which are the boundaries of the zero-cut.

Therefore, in condition of fuzzy triangular numbers as coefficients and by using the mentioned objective function, the problem of fuzzy linear regression is concluded to a linear programming problem [39,43]. The linear programming can be solved by using a plenty of commercial packages. In addition, the linear programming has very good mathematical properties, for instance, a local minimum solution is global and hence, in contrast with the no linear optimization, in this case, the LP provides a global optimum solution.

In the examined case (by using fuzzy triangular symmetrical numbers), the equation concludes to:

$$J = \min \left\{ mc_0 + \sum_{j=1}^m c_1 |x_{ij}| \right\} \quad (9)$$

subject to restrictions:

$$y_j \geq (a_1 x_j + a_0) - (1 - h)(c_1 |x_j| + c_0) \quad (9a)$$

$$y_j \leq (a_1 x_j + a_0) + (1 - h)(c_1 |x_j| + c_0) \quad (9b)$$

$$c_1, c_0 \geq 0, \text{ where } j = 0, 1, \dots, m \quad (9c)$$

The above mathematical functions, $(a_1 x_j + a_0) - (1 - h)(c_1 |x_j| + c_0)$, $(a_1 x_j + a_0) + (1 - h)(c_1 |x_j| + c_0)$ present the lower and the upper boundaries, respectively, of the corresponding h -cut of $\tilde{Y}_j$. As y_j , the j th observed data is meant, considering the dependent variable, which in this application is a crisp number.

It is well known that the solution for any other cut-level, h' can be obtained from the optimal h -cut-level solution as follows [50,51]:

$$\mathbf{A}^{h'} = \left(\mathbf{a}^h, \frac{1 - h}{1 - h'} \mathbf{c}^h \right) \quad (10)$$

the term h means the initial h -cut and the term h' means the new level h' . From Equation (10), it is obvious that the new level h' affects only the semi-widths. Hence, in case that there is not enough data, after the solution that is achieved from $h = 0$, the optimal solution for any other level h' can be achieved. The total fuzzy band will be:

$$J(\mathbf{c}_{h'}) = \frac{1}{1 - h'} J(\mathbf{c}_0) \quad (11)$$

The term $\mathbf{c}_{h'}$, $\mathbf{c}_0$ are the matrices which represent the semi-width of the solution with h' and for $h = 0$, respectively. For instance, the new matrix of the semi-width for $h = 0.5$ will be equal to:

$$\mathbf{c}_{h=0.5} = 2\mathbf{c}_{h=0} \quad (12)$$

2.4. Tested Scenarios and Basic Interpretation

The testing that we included in our research was split in two distinct scenarios:

Scenario a: the fuzzy regression was applied between phytoplankton, as the most sensitive and more frequently monitored BQE, and the OOAQ values. The values for OOAQ were generated using the unique yearly EQR values of phytoplankton, but the same values were kept for the indices of macrophytes and benthic macroinvertebrates for

all years within each monitoring period (2012–2015 and 2016–2019). That is, the EQR values of these indices were extended beyond their actual monitored year (Table 2). According to the WFD, the monitoring frequency of macrophytes and benthic macroinvertebrates for operational monitoring is once every three years.

Scenario b: the fuzzy regression took place between phytoplankton yearly EQR values and the OOA values generated using the yearly EQR values of phytoplankton and only the values of the indices of macrophytes and benthic invertebrates corresponding to the year they were actually monitored (Table 2).

The presented fuzzy regression model is applied between the yearly values of the independent (phytoplankton index) variable and the quality expressed by the dependent variable (OOA). Therefore, when the coefficients of the phytoplankton index have low values, that is, small central value (a_i) and semi-width (c_i), then it is obvious that the other quality indices affect the OOA. On the other hand, if the coefficients of the phytoplankton index have a high value, which is large central value and significant semi-width, then the phytoplankton index significantly affects the OOA. To work properly, there has to be originally a wide range of EQR values from all indices included in the regression. When a lake has quality values only within an EQR class range, it “cripples” the lake, not allowing it to go beyond these values. When both EQR values in both axes are identical, the Phytoplankton index “commands” the OOA, and in that case, the observation value is placed closest to the central fuzzy value. The boundaries designate the maximum acceptable value for the OOA principle acknowledging every value in the dataset. That means that the final acceptable assessment value can be within the range set by the central value plus or minus the semi-width (c_i , the second coefficient). This coefficient is the uncertainty created by the existing dataset at hand each time. When a boundary value falls over an existing observed value, this means that another BQE designated the range of acceptable values. It can be easily understood that the higher the x (phytoplankton) coefficient value, the more important is phytoplankton for the OOA principle. Similarly, the larger the second coefficient is, the wider the acceptable ranges. It should also be noted that for every new added value in the dataset (i.e., values for 2020, 2021, etc.), the regression can produce a slightly differentiated equation, meaning new semi-widths and thus new acceptable EQR ranges.

3. Results

3.1. BQEs Comparison and Status Classification Approaches

The ecological status assessed for the case-studied lakes differ up to two quality scales (Table 3). The macrophyte-based index appears to be the most lenient, while the index based on macroinvertebrates of the sublittoral and profundal zones appears as the strictest. On the other hand, the index based on the macroinvertebrates of the littoral zone remains more “austere” than phytoplankton, except in the cases of *Vegoritis* and *Lysimachia* (Table 3). The results for both monitoring periods using the OOA principle end in 16.7% of the cases in poor and good ecological quality and 66.7% in moderate, while when using the median and average approach, the results were similar, classifying the cases at 50% moderate and 50% good ecological quality. As for the final classification, the OOA principle, selecting the worst case, showed the poorest ecological status, downgrading up to two scales the status assessment, whereas the median and average indices’ EQRs were more kind in total, reflecting the quality distance among BQEs mentioned above.

Table 3. Ecological quality of six Greek lakes, based on multi-metric indices for different biological quality elements. The ecological status assessed by the “One Out–All Out” (OOAO), median, and average approaches. Quality color scale and Ecological Quality Ratio values are presented. P, phytoplankton [29]; M, macrophytes [30]; B-SP, sublittoral/profundal zoobenthos [32]; B-L, littoral zoobenthos [33].

Lake Type	Lake	Monitoring Period	P	M	B-SP	B-L	OOAO	Median	Average
Deep	Vegoritis	1st (2012–2015)	0.66	0.75	0.54		0.54	0.66	0.65
		2nd (2016–2019)	0.64	0.62		0.69	0.62	0.64	0.65
	Volvi	1st (2012–2015)	0.45	0.70	0.41		0.41	0.45	0.52
		2nd (2016–2019)	0.46	0.71		0.44	0.44	0.46	0.53
	Yliki	1st (2012–2015)	0.77	0.69	0.34	0.48	0.34	0.59	0.57
		2nd (2016–2019)	0.75				0.75	0.75	0.75
	Doirani	1st (2012–2015)	0.56	0.77	0.69		0.56	0.69	0.68
		2nd (2016–2019)	0.57	0.77			0.57	0.67	0.67
Shallow	Lysimachia	1st (2012–2015)	0.59	0.59	0.39	0.51	0.39	0.53	0.51
		2nd (2016–2019)	0.53	0.42		0.63	0.42	0.53	0.52
	Ozeros	1st (2012–2015)	0.71	0.45	0.53	0.49	0.45	0.51	0.55
		2nd (2016–2019)	0.72	0.62		0.52	0.52	0.62	0.62

3.2. Implementation of the Proposed Methodology

3.2.1. Fuzzy Regression Scenario A

In the cases of the Vegoritis and Volvi lakes, the estimated OOAO values form a tube parallel to the xx' axis (Figure 5a,b). This means that the phytoplankton index has no significant influence on the water quality classification for the examined lakes. To be precise, phytoplankton has a negligible coefficient (0.0025) for Lake Vegoritis, while for Lake Volvi, phytoplankton has no effect in shaping the OOAO values (Figure 5a,b). In this case, it is obvious that all the other indices rather than the phytoplankton index influence the OOAO. In contrast, the OOAO equation for Lake Yliki is proportional to the phytoplankton index, while the semi-width indirectly indicates the influence of the other indices (Figure 5c) encompassing the generated uncertainty (0.3123), which is also dependent on phytoplankton EQR.

Accordingly, Figure 6 depicts the fuzzy regressions for shallow natural lakes. A simple line is produced for Lake Doirani (Figure 6a) since the phytoplankton index has the lowest value among the indices, and hence, it is identical with the OOAO values, demonstrating no uncertainty. In Ozeros and Lysimachia lakes, the produced fuzzy curves represent a tube almost parallel to the xx' axis (Figure 6b,c). In Lake Lysimachia (Figure 6b), a fuzzy coefficient (0.4038) is produced from the EQR values generated from all other indices than the phytoplankton one. Hence, the final ecological status would be within the range of 0.4038 ± 0.0138 . The OOAO values for Lake Ozeros are slightly dependent on phytoplankton (0.0038) (Figure 6c).

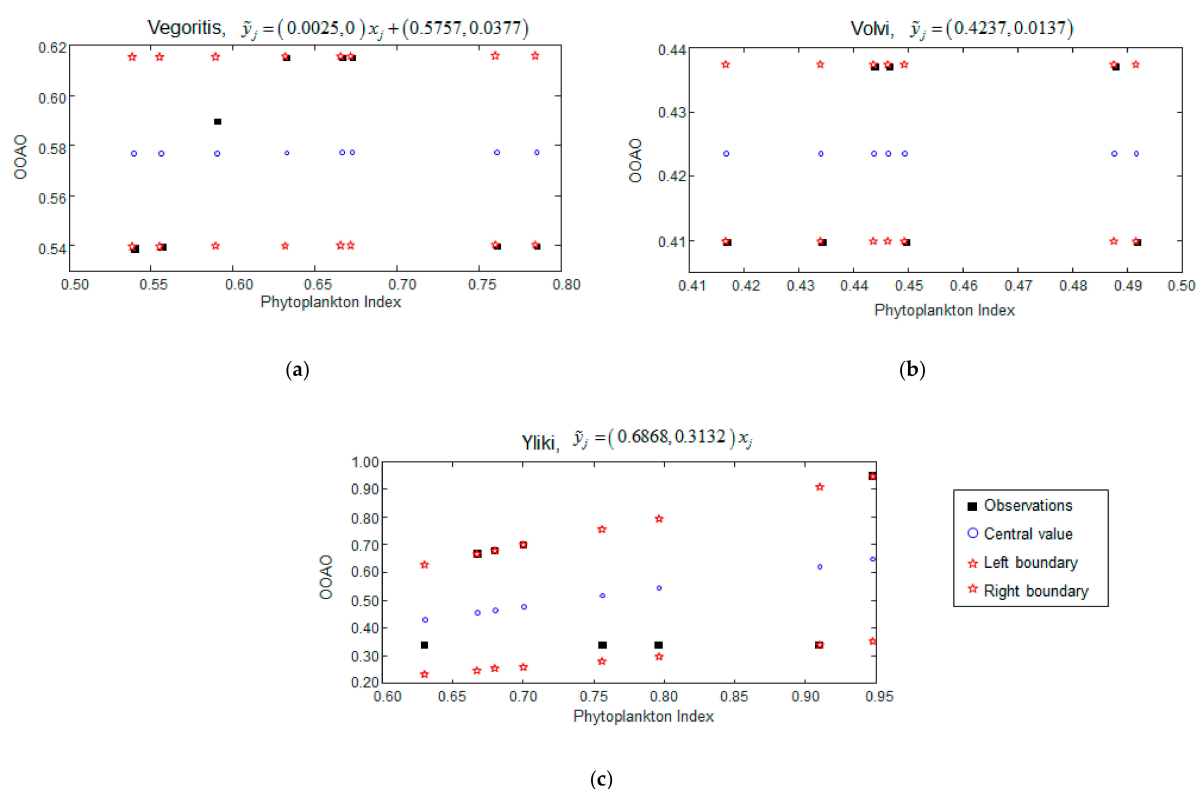


Figure 5. Graphical representation of the fuzzy regression for the OOAQ ("One Out–All Out") principle (dimensionless) with respect to phytoplankton index (dimensionless) in case of Scenario A for three deep natural lakes: (a) Vegoritis, (b) Volvi, and (c) Yliki.

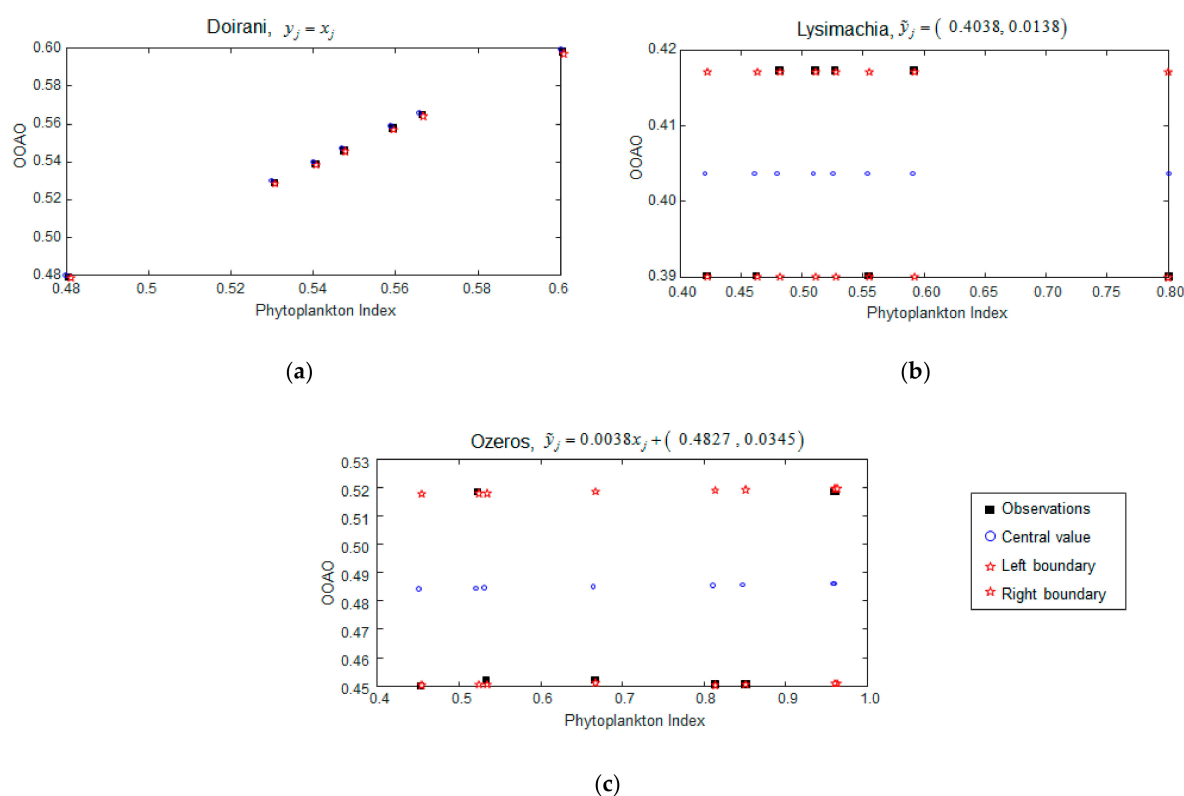


Figure 6. Graphical representation of the fuzzy regression for the OOAQ ("One Out–All Out") principle (dimensionless) with respect to phytoplankton index (dimensionless) in case of Scenario A for three shallow natural lakes: (a) Doirani, (b) Lysimachia, and (c) Ozeros.

3.2.2. Fuzzy Regression Scenario B

It seems that performing a fuzzy regression with phytoplankton EQR values (one for each year of the monitoring period) and two values for all other indices (one per four years) shapes the OOAo in favor of phytoplankton due to reduced yearly EQR comparisons. So, regardless of the lake type (shallow or deep), the independent variable (phytoplankton index) becomes a driving factor for the regression (Figures 7 and 8). In all lakes, the OOAo values are almost equivalent to that of the phytoplankton index, except in the case of Lake Ozeros (Figure 8). In that case, the phytoplankton coefficient is also high (0.7865), and the uncertainty is phytoplankton related, in contradiction with Lysimachia, Yliki, and Volvi lakes, where the uncertainty is an independent term.

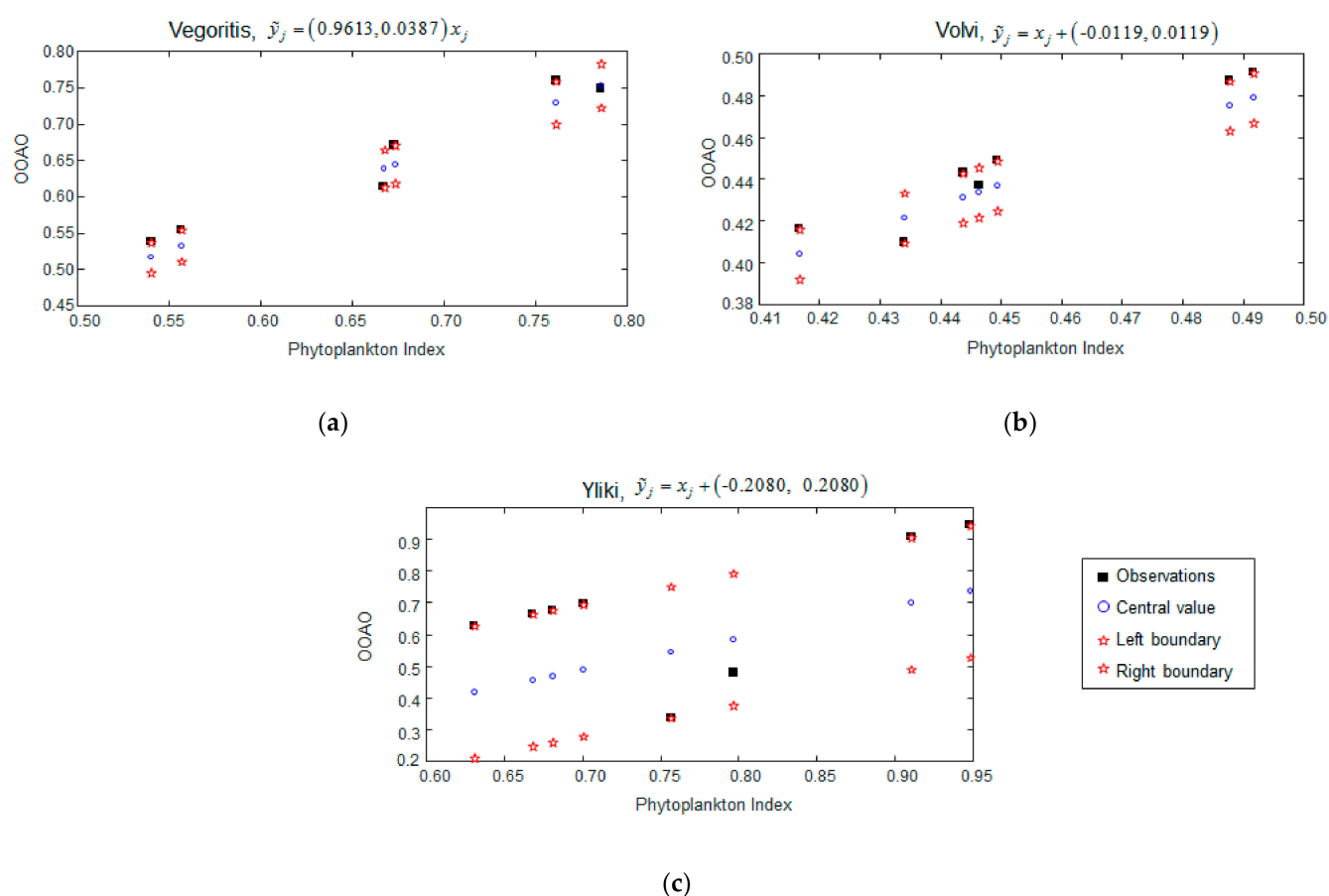


Figure 7. Graphical representation of the fuzzy regression for the OOAo (“One Out–All Out”) principle (dimensionless) with respect to phytoplankton index (dimensionless) in case of Scenario B for three deep natural lakes: (a) Vegoritis, (b) Volvi, and (c) Yliki.

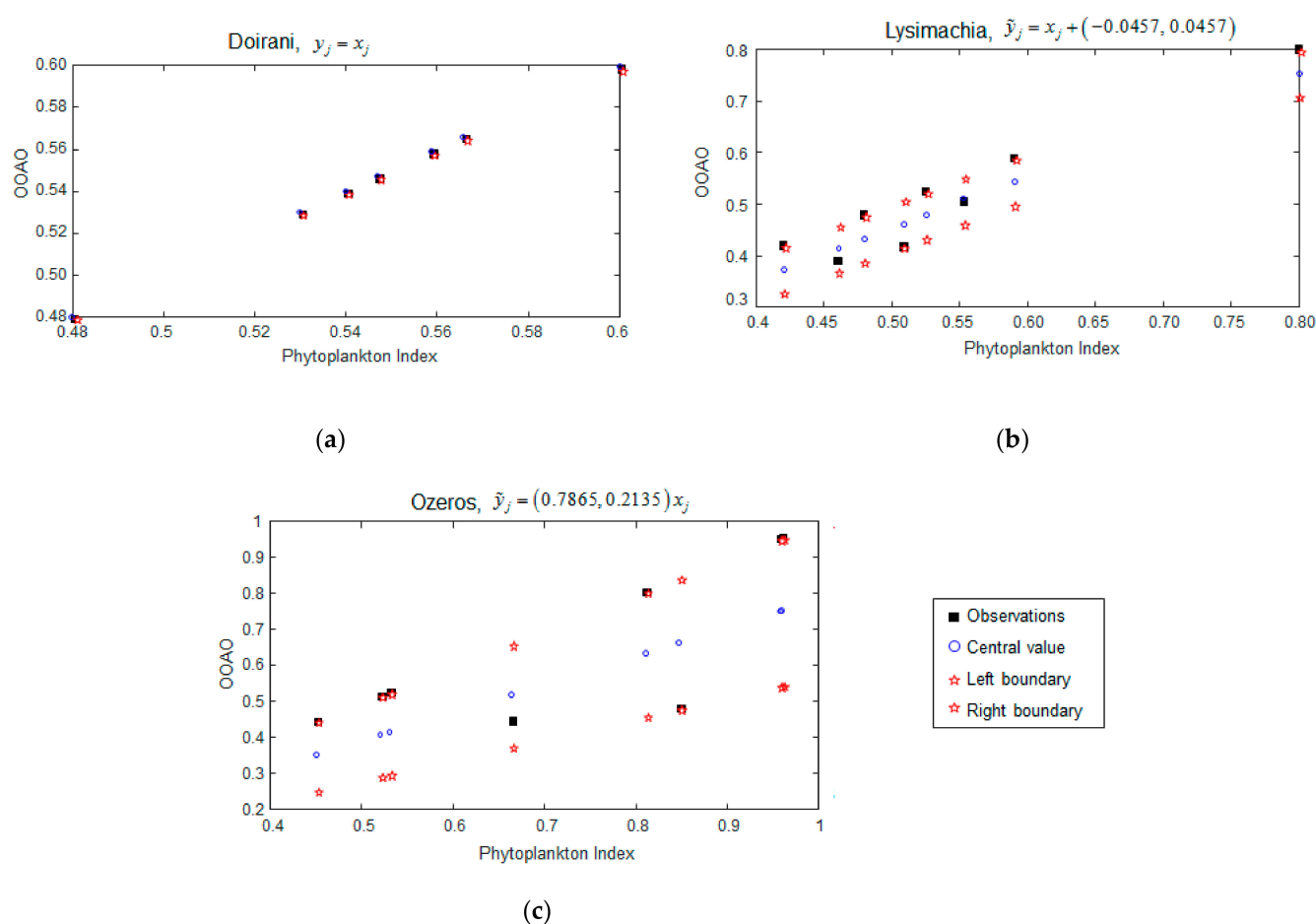


Figure 8. Graphical representation of the fuzzy regression for the OOAo (“One Out–All Out”) principle (dimensionless) with respect to phytoplankton index (dimensionless) in case of Scenario B for three shallow natural lakes: (a) Doirani, (b) Lysimachia, and (c) Ozeros.

4. Discussion

Assigning nature to boxes is difficult, and therefore, it is a challenge to fulfil the requirements of the WFD to define an overall ecological status, considering the ecosystem as a whole and using multiple BQEs in different waterbody types. The OOAo principle, where the status is based on the lowest score of any of the BQEs, is a precautionary rule that can lead to the strictest classification of waterbodies’ ecological status [52]. In this study, we attempted to consider combinations for multiple BQEs to assess ecological status in two different lake types. Moreover, we performed a fuzzy method to overcome the obstacle of the blind application of the OOAo approach, taking into account the mandatory monitoring periods according to the WFD.

As for the mere EQR values comparison, the macrophyte-based index appears as the less strict index, with an exception in the case of Lysimachia, possibly because it focuses on the euphotic littoral zone. It is considered that the abundance and the composition of plant species very often have a quite wide ecological scale [19,20]. Additionally, which BQE reflects better the lake quality is a matter of the lake itself, since all differ according to their morphometry and hydrology, and they are usually subjected to different pressures. Even within a lake, multiple BQEs can respond differently in various lakes’ parts, seasons, and time scales [53,54]. According to previous studies [21,52,55] regarding the different approaches to evaluate the overall status, the OOAo principle was the most conservative, downgrading the ecological status of lakes. The OOAo principle is also considered as an inadequate tool for addressing progress or deterioration in status [56], as it masks the real changes that WFD implementation can achieve, especially in cases where PoMs have

been implemented with delay. The suggested fuzzy method in all scenarios retains the strictness of the OOA principle but in a way that the main BQE (i.e., phytoplankton) can be boosted and the character of each lake showcased. This approach comes in line with the most voted choices in a query posed to all experts and attendees during EU WFD 2017 e-conference [11]. Moreover, the fuzzy regression can keep learning the waterbody as the dataset expands. In Scenario A, using repeatedly the same value for the indices of macrophytes and benthic invertebrates for four consecutive years, is a bias against the main BQE (i.e., phytoplankton) for lakes status classification as the regression frames any picture that this BQE could give. This was depicted by the fuzzy regression that generated tubes parallel to xx' axis and the equations that lessened the importance of phytoplankton. The only exception was Lake Ylik because EQR values were based on macroinvertebrate and macrophyte indices, which were not available during the second monitoring period (2016–2019), hence affecting the OOA and consequently the fuzzy regression. This bias is imposed by the WFD recommendations requiring different monitoring frequencies for each BQE responding to different life spans and community variability. Regarding Scenario B (using only the value when the BQEs were monitored), one would claim that it is biased in favor of phytoplankton, but we deemed that this approach allows the main BQE (i.e., phytoplankton) to express the yearly quality variation, respecting the OOA principle when other BQEs were also monitored. In any of the scenarios, this fuzzy method enables the experts to assess if the most often monitored BQE (i.e., phytoplankton) plays a crucial role in defining the OOA values. This enables the assessment of the waterbody with the uncertainty expressed by the boundaries, which work as a safety net. Additionally, it guides the experts to check which of the other BQEs characterize better the qualitative status of the lake.

As for the progress reporting of individual BQEs, according to Carvahlo et al. [11], this requires a larger (lakes and monitoring years) dataset to apply a statistically solid fuzzy regression. However, to evaluate the progress as expressed by indices is an ambitious task, given the result is practically often a circumstance of the relationship among the nature of administrative governance, the nature of PoMs taken [57], and the minimum of WFD obligations (along with the expected lag from the waterbody). Thus, the framework for overall ecological status classification should in fact include a form of elasticity. It is expected that different governance approaches for the same nature of measures could end in diverse results due to design and implementation progress. Tsakiris [58] highlighted that the first RBMPs showed a significant progress on the fulfilment of environmental objectives, while the second reporting period did not support the same rate in progress [59,60]. Following the OOA principle as safer, only 20% of the EU waterbodies showed improvement in their status [61] despite costly measures [62]. Instead, there was a call for more stringent nutrient thresholds [63,64], signifying the need for new approaches for the sustainability of WFD 20⁺. Moreover, Greece and especially its lake basins host mainly agroecosystems, where diffuse pollution cannot easily be tackled, and green infrastructure measures need more time to pay off in quality status terms [8,57]. This issue is aggravated under the Mediterranean climate prism and some specific hydromorphological characteristics (i.e., shore alterations, high water retention time). Moreover, a six-year period for RBMP cycles renewal can be considered as adequate for reporting progress in ecological point of view, but such a period can hinder trends and the effectiveness of policy measures [56] or include the possible benefits from action plans in a management point of view. The next step that goes beyond the OOA principle, and is nowadays under consultation [56], is the use of new quality indicators which are focused on implementation shifts (i.e., pressure reduction, PoMs application) [56]. Novel reporting protocols can perhaps improve the digitalization and administrative simplification but add extra burden on MSs to follow the changes. It is unclear, though, if this change can positively impact the OOA principle in any means.

In this study, we suggest the development of an overall assessment that will be based on the precautionary OOA principle through the fuzzy OOA approach, which we consider that it enables a better implementation of the WFD and allows for a case-

specific evaluation, including the uncertainty in classification as an asset. We argue that such an approach offers a deeper system understanding through self-learning processes based on the existing datasets, while it also recognizes the different character of the lakes and adds a special gravity to the most sensitive and most frequently monitored BQE. However, we believe that all BQEs should be monitored, as freshwater ecosystems are highly complex systems [65], and an efficient management should depend on multiple components [66]. Additionally, all BQEs samples should be taken in a timing able to support a more proper application of OOA (as it was treated in Scenario B) under the same environmental conditions (i.e., meteorological, hydrological) to establish more stable stressor-response relationships.

5. Conclusions

1. The inclusion of a fuzzy regression among the frequently monitored BQE (phytoplankton) and the outcome of OOA (determined by the comparison of four BQE indices) application in lakes encompasses the uncertainty and the possibility to broaden the acceptable final EQR based on the character and status of each lake;
2. The fuzzy OOA is an approach that seems to allow a better understanding of the WFD implementation and case-specific evaluation, including the uncertainty in classification as an asset;
3. It offers a deeper understanding through self-learning processes based on the existing datasets;
4. As for the progress reporting of individual BQEs, this requires a more complete dataset to apply a statistically solid fuzzy regression.

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Conflicts of Interest: The authors declare no conflict of interest.

Appendix A

Table A1. Reference values of phytoplankton, macrophytes, and littoral invertebrates indices.

Index	Metrics	Lake Type GR-DNL	Lake Type GR-SNL
HeLPhy	Total Phytoplankton Biovolume ($\text{mm}^3 \text{L}^{-1}$)	1.29	0.74
	Cyanobacteria Biovolume ($\text{mm}^3 \text{L}^{-1}$)	0.01	0.01
	modNygaard Index	1.03	1.11
	Chlorophyll a ($\mu\text{g L}^{-1}$)	1.56	3.59
HeLM	TIHelm	7.14	7.14
	Cmax (m)	12.2	6.1
	ASPT	5.47	5.47
HeLLBI	Odonata (% AC)	16.67	16.67
	Simpson	0.80	0.80

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