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METHODS APPLIED FOR MEASUREMENT AND VISUALIZATION OF CHANGES IN BIODIVERSITY

METODY POMIARU I WIZUALIZACJI ZMIAN BIORÓŻNORODNOŚCI

Abstract: The article presents the possible methods for determining biological or statistically significant differences between taxocenoses compared with respect to biodiversity. To obtain a complete description of biological differences between the compared hypothetical communities, the following indices were calculated: *S* (taxon richness), *H'* (the Shannon index), H_{max} (the maximum value of the Shannon index for the richness of taxa represented by the same number of individuals), V_d (a percentage value of covering the structural capacity of community, "evenness deficiency"), *E* (the MacArthur index - a taxon number (*S*) in a community for which the observed value of *H* equals H_{max}), and P_s (a taxon richness shortage in percents). Moreover, a graphic profile method (Δ_β, T_j, and L_j profiles) was used for comparing the diversity of the communities. To obtain information about statistically significant differences in biodiversity between the analysed communities, rarefaction curves were applied. The curves are based on the null models and the Monte Carlo method. The rarefaction method resulted in determination of the statistical significance of the differences between taxon richness and Shannon's index values for the compared communities. The V_d and P_s indices and the profile method allowed concluding about the significance of the biological differences between taxocenoses, even when their values of Shannon's *H'* indices were numerically similar.

Keywords: biodiversity, Shannon's index, rarefaction curves, profile method

Introduction

While undertaking the 'strategy of protection and rational use of biological diversity' [1, 2], as well as biomonitoring processes in bioreactors with activated sludge [3-7] and surface water *eg* [8-12], an ideal solution would be creating conditions that would ensure 'control of strategy to be realized', where the monitoring, *ie* systematic measurements, would be carried out and the measurement results could be reliably compared. Biodiversity (biological diversity α, β, and γ) may be evaluated based on the richness of species *S* or taxa selected at own preference [13]. It may also be assessed in terms of the number of taxa and their relative abundance (or relative biomass, or relative coverage degree) by calculating

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Shannon's index of diversity *H*' [14-17], or other biotic indices [18-23]. While applying methods for description of communities based on *S* or *H*' indices, it is not possible to assess correctly the biological or statistically significant differences (or absence of such differences) between compared taxocenoses characterised by identical or subjectively different *S* and *H*' values. Nowadays, there are means to solve this problem [24-28]. It seems that these methods are not yet widely used in the field of environmental engineering. Hence, the objective of the present paper is to show them using a simple example of three hypothetical communities of living organisms.

Materials and methods

The material used in the investigations comprised three hypothetical communities, two of which had the same *S* values (species richness), but differed with respect to the *H*' values (Shannon's index). Two exhibited similar *H*' values, but differed in the *S* values; the problem is better illustrated by the fact that they had the same number of individuals $N = 20$ (Table 1).

Table 1

Taxonomic-biocenotic characteristics of three hypothetical communities A, B, and C

Taxa	**Community**		
	A	**B**	**C**
s_i	n_i	n_i	n_i
A	4	10	3
B	4	1	6
C	4	3	-
D	4	2	4
E	4	4	7

The formulas presented below were used in measurements and comparisons of the biological diversity of the communities (objects) characterised [27, 29-34].

The species richness *S* was established by simply summing all of the taxa belonging to the analysed community and the same method was used for *N* [32]:

$$S = \sum_{i=1}^{s} s_i \tag{1}$$

$$N = \sum_{i=1}^{S} n_i \tag{2}$$

where: *S* - species richness, number of taxa; s_i - distinguished taxon; *N* - total number of individuals in the sample (total number of individuals in taxocenosis); n_i - number of individuals of the i^{th} taxon.

Relative abundances, necessary for calculation of the Shannon index and derived indices, were determined on the basis of the following equation [32]:

$$\Pi_i = \frac{n_i}{N} \tag{3}$$

where Π_i - relative abundance of the i^{th} taxon.

The *H*' Shannon index was calculated on the basis of the following equation [32, 33]:

$$H' = -\sum_{i=1}^{S} \Pi_i \lg_2 \Pi_i \tag{4}$$

where *H*' - Shannon's index of biodiversity.

The maximum value of the Shannon index H'_{max} [30, 31] was calculated using the following formula:

$$H'_{\max} = \lg_2 S \tag{5}$$

where H'_{max} - maximum *H*' value for a given richness *S*, which would occur if all taxa were equally abundant.

The evenness deficiency index V_d was calculated based on the concept presented by Hurlbert and Magurran [30, 31]:

$$V_d = \left(1 - \frac{H'}{H'_{\max}}\right) \cdot 100 \tag{6}$$

where V_d - a percentage value of implementation of community capacity (evenness deficiency).

MacArthur's index *E* was calculated according to the following equation [29]:

$$E = 2^{H'} \tag{7}$$

where *E* - species richness of a community, for which the observed *H*' is the H'_{max} value.

The proportionality shortage index P_s was calculated based on the concept presented in [27]:

$$P_s = \left(1 - \frac{E}{S}\right) \cdot 100 \tag{8}$$

where P_s - expressed in percent species shortage.

Additionally, a graphic profile method of Δ_β, T_j, and L_j was applied to compare the biodiversity of the communities [32]. The Δ_β profiles were plotted on the basis of the points on the coordinate axes (β, Δ_β), where $\beta > -1$ and Δ_β can be described by the following formula [35, 36]:

$$\Delta_\beta = \sum_{i=1}^{S} \left\{ \frac{1 - \Pi_i^\beta}{\beta} \right\} \Pi_i = \frac{1 - \sum_{i=1}^{S} \Pi_i^{\beta+1}}{\beta} \tag{9}$$

The T_j profiles were plotted on the basis of the points on the coordinate axes (j, T_j), where $j = 1, \ldots, S-1, S$, and T_j is described by the following formula [35, 37]:

$$T_j = \sum_{i=j+1}^{S} \Pi_i^{\#} \quad (\text{for } j = S,\ T_S = 0;\ \text{for } j = 0,\ T_0 = 1) \tag{10}$$

where: $\Pi_i^{\#}$ - relative abundance of the i^{th} species, classified into the so-called ranked relative abundance vector ($\Pi^{\#}$), which covers the relative abundances of all species ordered from the greatest to the lowest.

The latter profiles mentioned above, *ie* the L_j profiles, were plotted on the basis of the points on the coordinate axes (*L*, *L*'). Calculations of the coordinates were performed based

on ranked relative abundance vectors $\Pi^{\#}$, $\Pi^{\#'}$ of the compared communities, as follows [38]:

$$\begin{gathered} p_0 = (L_0, L'_0) = (0,0) \\ p_1 = (L_1, L'_1) = (\Pi_1^{\#}, \Pi_1^{\#'}) \\ p_2 = (L_2, L'_2) = (L_1 + \Pi_2^{\#}, L'_1 + \Pi_2^{\#'}) \\ . \\ . \\ . \\ p_k = (L_k, L'_k) = (L_{k-1} + \Pi_k^{\#}, L'_{k-1} + \Pi_k^{\#'}) \\ p_s = (L_s, L'_s) = (L_{s-1} + \Pi_s^{\#}, L'_{s-1} + \Pi_s^{\#'}) = (1,1) \end{gathered} \tag{11}$$

Null models were applied to obtain information about the statistically significant differences between the communities analysed in terms of the taxa richness and diversity. These models (based on the Monte Carlo method) enable statistical significance to be determined even if the sizes of compared samples are not the same - $N_1 \neq N_2$ [25, 26]. Eco Sim 7.0 software [24-26] was used for calculations concerning the analysed communities A, B, and C, and data required for plotting rarefaction curves.

Results and discussion

Table 2 shows the results of calculations made according to formulas (1)-(4).

Table 2

Taxa richness, total abundance, and Shannon's index for communities A, B, and C

Index	Community		
	A	**B**	**C**
S	5	5	4
N	20	20	20
H'	2.322	1.923	1.926

Given the results presented above, it is hardly possible to state explicitly whether the communities compared, especially B and C, vary with respect to the biological differences. However, the data presented in Table 3 allow a presumption that, in the biological sense, community C is characterised by lower biodiversity than community A, but greater biodiversity than community B, as it exhibits a lower evenness deficiency V_d and a lower species shortage P_s. This is displayed by the AMOEBA-type graphs [39-42] - Figures 1 and 2.

Table 3

Values of H'_{max}, V_d, E, and P_s indices for communities A, B, and C

Index	Community		
	A	**B**	**C**
H'_{max}	2.323	2.323	2.0
V_d	0.0%	17.2%	3.7%
E	5.0	3.793	3.800
P_s	0.0%	24.2%	5%

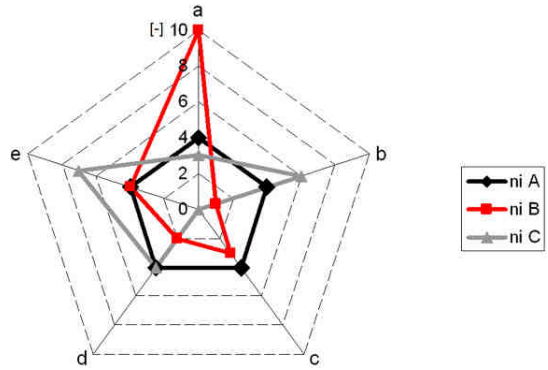

Fig. 1. Species richness *S* of communities A, B, and C

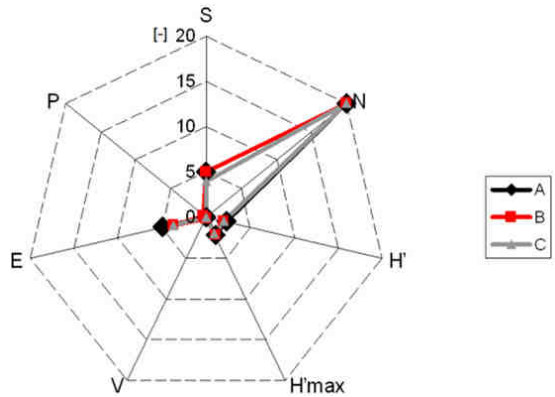

Fig. 2. Values of *H'*, *S* indices and derived indices for communities A, B, and C

Figures 3-7 show the results obtained with the profile method. It should be explained that the Δ_β profiles plotted for the compared communities show their ranking according to three indices: 'species count' $\Delta_{Si} = S - 1$, Shannon's index and Simpson's index $\Delta_{Si} = 1 - \sum_{i=1}^{s} \Pi_i^2$. If these profiles do not cross each other, all the indices used rank the compared communities in the same way (*ie* according to all indices, a given community X is more diverse than X'). If the profiles cross, the ranks of the communities will depend on the index applied. The T_j profiles have been developed based on the concept of *intrinsic diversity ordering* [36]. The mutual position of the T_j profiles plotted for the compared communities allow conclusions concerning the intrinsic diversity ordering. If the profile of the X community lies above the profile of the X' community, the X community is intrinsically more diverse, whereas the X' community is less diverse. When the profiles intersect, it is impossible to determine which of the communities compared is more intrinsically diverse. The Δ_β profiles are isotonic to T_j profiles, which means that Δ_β profiles can inform about the intrinsic diversity ordering between the communities. It should be remembered, however, that sometimes the Δ_β profiles may not cross even if the T_j profiles do. The L_j profiles are curves, which are also based on the concept of intrinsic diversity

ordering. While plotting these profiles, the points form curve *p*. Its position with respect to the diagonal *d* (with the equation $y = x$) carries information concerning the intrinsic diversity ordering between the compared X and X' communities. When the curve *p* lies above the diagonal *d*, community X is intrinsically more diverse than X'. When the curve *p* lies under the diagonal *d*, X' is intrinsically more diverse than X. If the curve *p* and the diagonal *d* overlap, there is no difference in the diversity between the communities. When the curve *p* crosses the diagonal *d*, it is not possible to state which of the compared communities is more diverse. It should be noticed that the T_j and L_j profiles lead to the same conclusions. The selection of the type of the profile has no effect on the final result [32].

Based on the Δ_β profiles for the analysed communities, taxocenosis A can be regarded to be more diverse than taxocenoses B and C, whereas taxocenoses B and C cannot be compared due to the crossing courses of the profiles (Fig. 3).

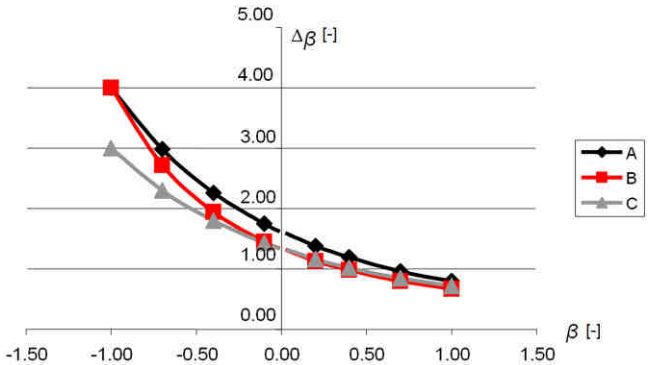

Fig. 3. The Δ_β profiles for communities A, B, and C

The T_j and L_j profiles, similarly to the Δ_β profiles, indicate that community A is characterised by higher biodiversity than communities B and C (Figs. 4-6). Taxocenoses B and C are incomparable due to the crossing of the graphs of the respective T_j and L_j profiles (Figs. 4 and 7).

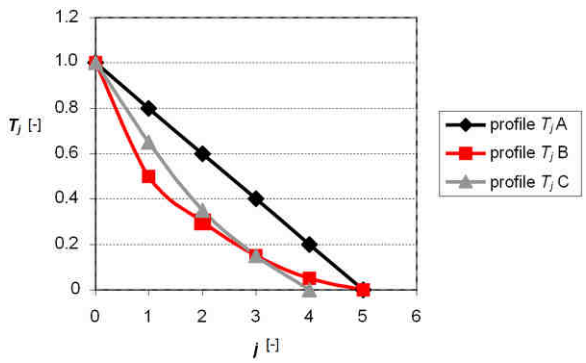

Fig. 4. The T_j profiles for communities A, B, and C

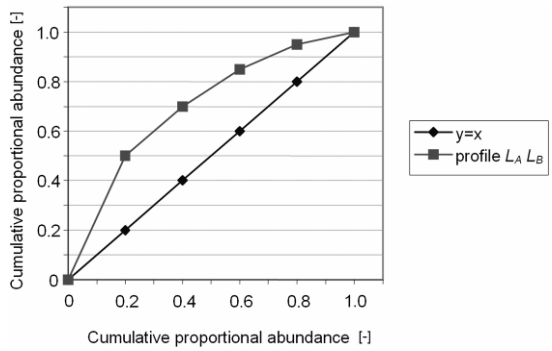

Fig. 5. The L_j profile for communities A and B

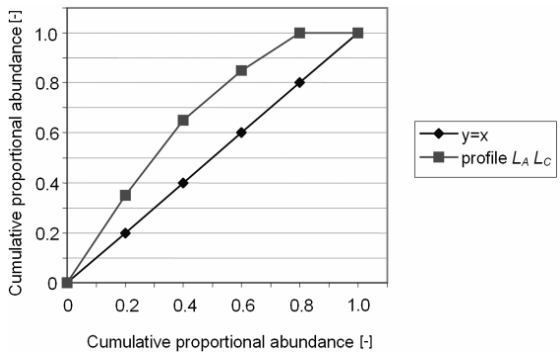

Fig. 6. The L_j profile for communities A and C

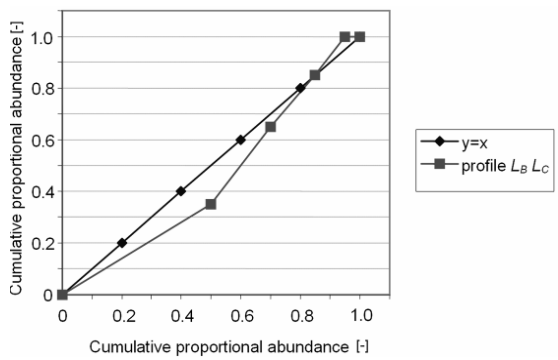

Fig. 7. The L_j profile for communities B and C

The methods presented above do not yield judgements concerning the statistical significance of the differences or confidence intervals - the latter indicate the statistical significance of the differences if they do not overlap. Thus, it is not possible to decide about the significance of the differences between the communities compared. The problem discussed may be solved using multiple sampling methods. With these methods, rarefaction

curves were obtained for the compared communities A, B, and C (Figs. 8 and 9). Figure 8 shows that multiple sampling of 4 individuals from 20 allows determination of the full number of species in community C; only 5 individuals from 20 belonged to community A (in the upper graph the black dashed curve is comparable to red dashed curve). However, to get the full number of taxa in community B, multiple sampling of 10 individuals is required. A low number of taxa and individuals lead to certain 'angularity' of the graphs obtained; this should disappear in the case of more abundant samples $N > 50$.

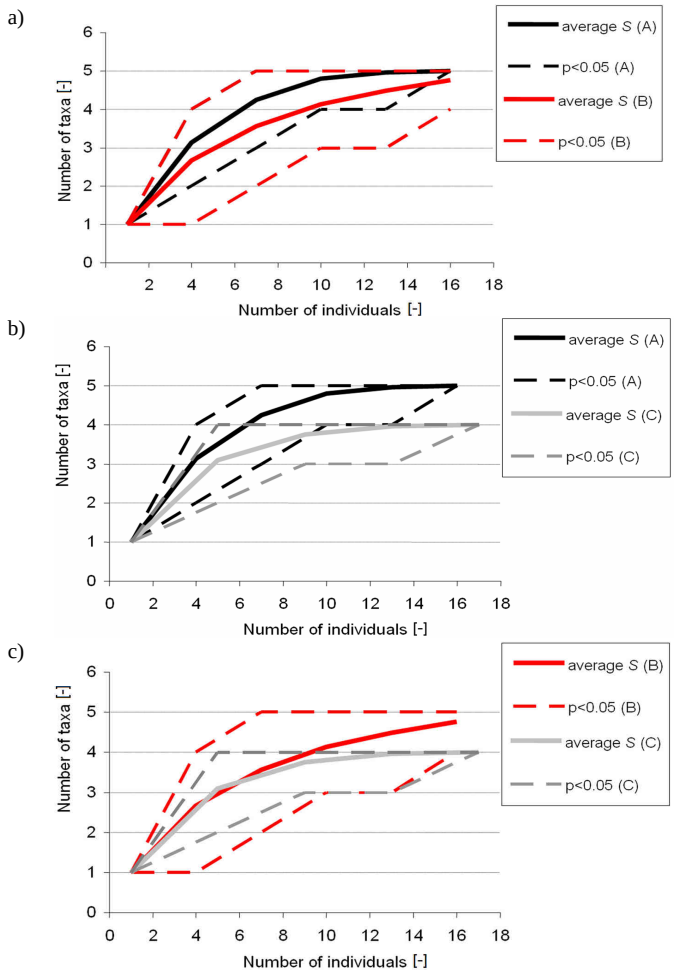

Fig. 8. Rarefaction curves of species richness for communities A, B, and C

The *H*' values calculated with the use of the EcoSim 7.0 software differ from those presented in Table 2. This can be explained by the fact that the EcoSim software calculates Shannon's index using a natural logarithm, while the base-2 logarithm is commonly applied in the theory of computer science. Hence, the *H*'(A) value is 1.61, and *H*'(B) and *H*'(C) equal 1.33.

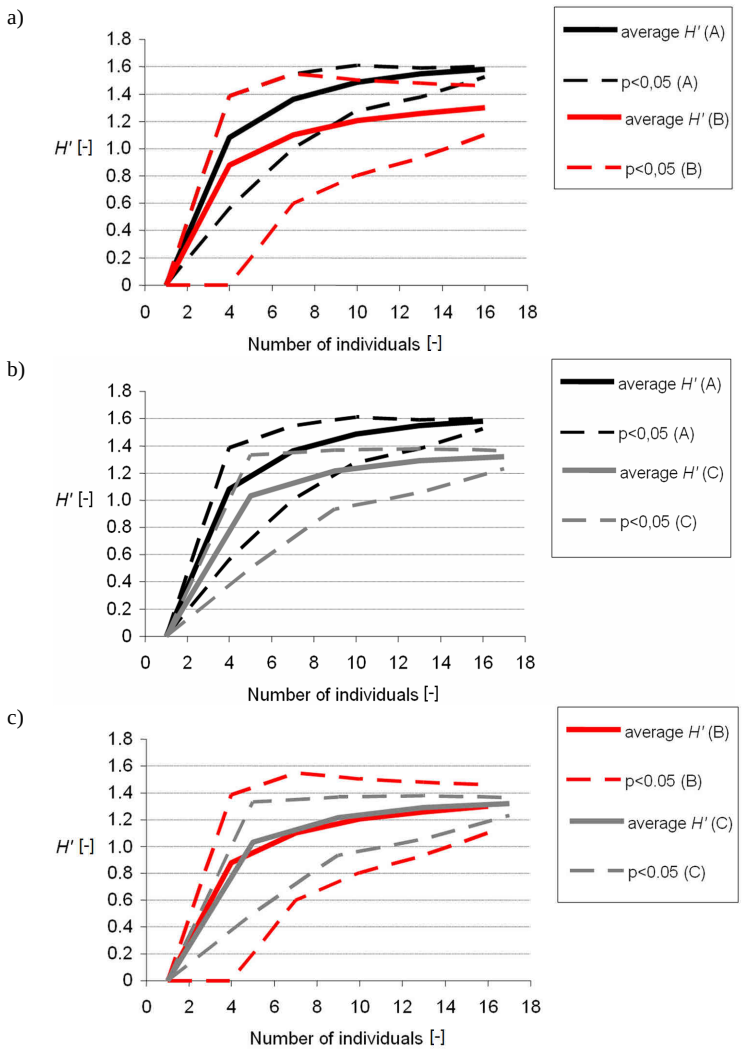

Fig. 9. Rarefaction curves of Shannon's index *H*' for communities A, B and C

Based on the results obtained, it may be presumed that in the case of Shannon's species diversity, taxocenosis A significantly differs from taxocenoses B and C (the confidence intervals do not overlap - Fig. 9a, b).Taxocenoses B and C do not differ in terms of the parameter discussed (the confidence intervals overlap - Fig. 9c).

The method of rarefaction presented in the article can be successfully applied not only for the number of taxa and Shannon's diversity index [24, 25, 28] but also for evaluation and analyses of other indices important in ecology. For instance, the effective number of species and, connected with this issue, problems of sample size, sample numbers, and sample coverage could be mentioned here [43].

Conclusions

The following conclusions can be drawn from the analysis of the results:

- It is possible to express biodiversity quantitatively, and after measurements thereof, taxocenoses and their changes in the time function can be compared with respect to this parameter.
- The indices developed, derivatives of the *H'* index (especially V_d and P_s), indicate the significance of the biological differences, even when at similar values of the Shannon's *H'* indices.
- The Δ_β profiles serving the comparison of the values of species richness, Shannon's and Simpson's indices in the investigated taxocenoses, have a basic shortcoming - the β values in the denominator of the fraction have to pass by 0 value, which is arithmetically inadmissible.
- The rarefaction methods facilitate determination of the statistical significance of the differences, or absence of the statistical differences, between Shannon's index values for the compared communities.

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Abstrakt: W artykule przedstawiono metody określenia biologicznych i statystycznie istotnych różnic między taksocenozami porównywanymi pod względem bioróżnorodności. W celu pełnego opisu różnic biologicznych pomiędzy porównywanymi, hipotetycznymi zbiorowiskami obliczono wskaźniki: *S* (bogactwo taksonów), *H*' (indeks Shannona), H_{max} (maksymalna wartość indeksu Shannona dla danego bogactwa taksonów charakteryzujących się takimi samymi liczebnościami), V_d (wyrażona w procentach wartość wypełnienia strukturalnych możliwości zbiorowiska; niedostatek „równomierności"), *E* (indeks MacArthura, czyli liczba taksonów *S* w zbiorowisku, dla którego dany indeks *H* przyjąłby wartość maksymalną) oraz P_s (wyrażony w procentach niedostatek bogactwa taksonów). Dodatkowo, dla porównania bioróżnorodności zbiorowisk użyto graficznej metody profili Δ_β, T_j i L_j. W celu uzyskania informacji o statystycznie istotnych różnicach między analizowanymi zbiorowiskami pod względem bioróżnorodności wykreślono krzywe rarefakcji, bazujące na modelach numerycznych i metodzie Monte Carlo. Metoda rarefakcji umożliwiła określenie statystycznie istotnych różnic między wartościami bogactwa taksonów i indeksu Shannona obliczonych dla porównywanych zbiorowisk. Metoda profili oraz indeksy V_d i P_s pozwalają wnioskować o istotności różnic biologicznych nawet wtedy, kiedy wartości indeksów *H*' Shannona są do siebie liczbowo zbliżone.

Słowa kluczowe: bioróżnorodność, indeks Shannona, krzywe rarefakcji, metoda profili