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QUANTITATIVE ASSESSMENT SOFTWARE DEVELOPMENT FOR THE CHLORELLA VULGARIS MICROALGAE POPULATION

Purpose. *Software development to quantify the population growth for the microalgae Chlorella.*

Methods. *Software was developed using Python programming language, external libraries: Numpy, Xlsxwriter, Matplotlib and PIL. The developed software suitability to quantify the population growth of the microalgae Chlorella and its verification was determined. During this verification, the population growth of the microalgae with different stages of biomass growth were evaluated. Samples of the microalgae Chlorella were obtained over a period of several weeks' population growth.*

Results. *Software that allows estimating the population growth of the microalgae Chlorella in the aquatic environment has been developed. Using this software, data dependence of the heterogeneity coefficient of microalgae Chlorella vulgaris biomass growth was obtained. The population growth leads to a decrease in this heterogeneity. This effect can be due the optimal conditions presence for the cultivation of microalgae Chlorella vulgaris.*

Scientific novelty. *The effect of microalgae Chlorella population growth on the system homogeneity has been determined. The increase of microalgae Chlorella vulgaris biomass content within 4 weeks leads to coefficient of heterogeneity decrease from 92% to 47%, due to the aggregation of microalgae Chlorella vulgaris in the aquatic environment.*

Practical significance. *Developed software allows quantify the growth of the microalgae Chlorella vulgaris population in the aquatic environment and can be used to control the algae biomass rise in industrial production.*

Conclusion. *Software that allows assessing the uniformity distribution of Chlorella vulgaris microalgae biomass population growth has been developed. It is applicable to study the level of microorganisms' concentration in the aquatic environment. The results of its verification indicate the developed software suitability for practical use. Using the developed software, the influence of Chlorella vulgaris microalgae biomass population growth in the aquatic environment was determined. It is shown that microalgae Chlorella vulgaris population growth from 1 to 4 weeks leads to decrease in the mixing index from 93% to 47%, which is due optimal conditions presence for the cultivation of the microalgae Chlorella vulgaris. The research methodology can be used in the automated monitoring complexes development for the ecological state of water resources and for controlling the industrial production microalgae Chlorella vulgaris population.*

Keywords*: environmental monitoring software; heterogeneity mixing index; Chlorella vulgaris.*

Intro. Today long-term voyages by submarines and, in the near future, by spacecrafts, pose a problem in the diet of personnel. One solution to this problem is the cultivation of unique types of algae, namely the green microalgae Chlorella vulgaris. Microalgae contain more than 60 trace elements, the concentration of which is much higher than in the terrestrial plants. The dry biomass of C. vulgaris contains more than 45–50% of proteins, including essential amino acids; 30–35% of carbohydrates, including mainly starch, cellulose, hemicellulose and soluble sugars; 5–10% of lipids. Chlorella vulgaris biomass includes vitamins of groups B, C, PP, E, and carotene. The use of chlorella biomass solves the issue of an additional source of useful elements in the diet of personnel on long journeys. Chlorella also produces a large amount of oxygen and consumes carbon dioxide, which can be used to create a microclimate in cramped conditions [1–3].

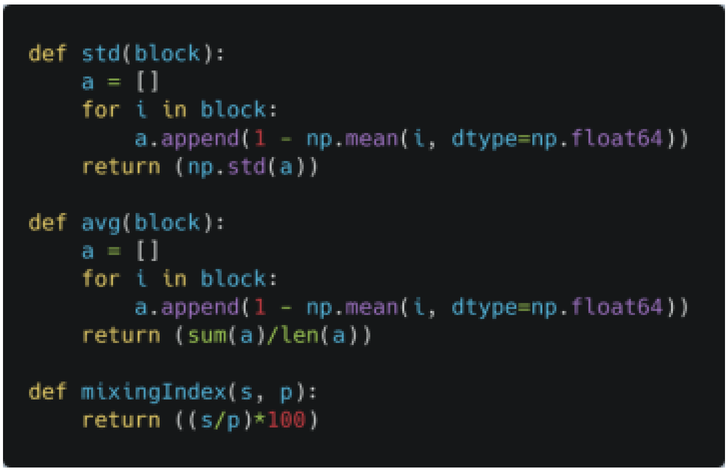
Thus, the quantifying the microalgae Chlorella population in industrial production under cyclic biomass growth conditions is acute. To assess the biomass homogeneity particles aquatic environment distribution, the segmentation method can be used. It is based on image division into regions for which a certain homogeneity criterion is met, for example, approximately highlighting areas equal brightness or pixel count ratio in the image. The image area concept is used to define a image related group elements that have certain common features (properties) [4, 5].

The most difficult tasks are non-trivial images segmentation in digital image processing. The final result of computer image analysis is often depending on the segmentation accuracy, so to improve algorithm of the segmentation reliability considerable attention should be noted. Building segmentation through the use of a threshold is one of the primary and straightforward methods. The threshold, serving as a distinguishing feature, aids in categorizing the target signal into different classes. The thresholding operation is based on the brightness value comparison of each image pixel with a given threshold value. Achieving the suitable threshold value enables the possibility to highlight areas of a certain type in the image. Binarization is the thresholding operation that produces a binary image. The goal of this operation is to significantly diminish the amount of information present in the image [6, 7].

During the binarization process, the initial halftone image, featuring a specific number of brightness levels, undergoes conversion into a black and white image where pixels are assigned only two values – 0 and 1. Due to the reduced information content in a binary image compared to a halftone image, processing, storage, and transmission of binary images are more straightforward. The lack of software implementation of the above method for assessing the homogeneity of the distribution of biomass particles in the aquatic environment hinders its widespread use in practice, presented in this article study which determines the relevance [8].

The task statements. The primary objective of this project is to create software designed to evaluate the distribution of Chlorella vulgaris biomass homogeneity in aquatic environments. In order to accomplish this objective, it was essential to examine information sources for the selection of a suitable programming language. Furthermore, functions needed to be developed for converting input data into a binary image, clustering binary images, and calculating the mixing index for the distribution of biomass particles in the aquatic environment. Finally, the suitability of the developed software was assessed through verification processes.

Research results. For the development of software to evaluate the homogeneity of Chlorella vulgaris biomass distribution in aquatic environments, the Python programming language and libraries such as Numpy, Xlsxwriter, Matplotlib, and PIL were used. Standard deviation calculating functions of the particle concentration in clusters (std), the average particle Chlorella vulgaris biomass concentration (avg) in the aquatic environment, and mixing index (mixingIndex) are shown in Fig. 1.



```
def std(block):
    a = []
    for i in block:
        a.append(1 - np.mean(i, dtype=np.float64))
    return (np.std(a))

def avg(block):
    a = []
    for i in block:
        a.append(1 - np.mean(i, dtype=np.float64))
    return (sum(a)/len(a))

def mixingIndex(s, p):
    return ((s/p)*100)
```

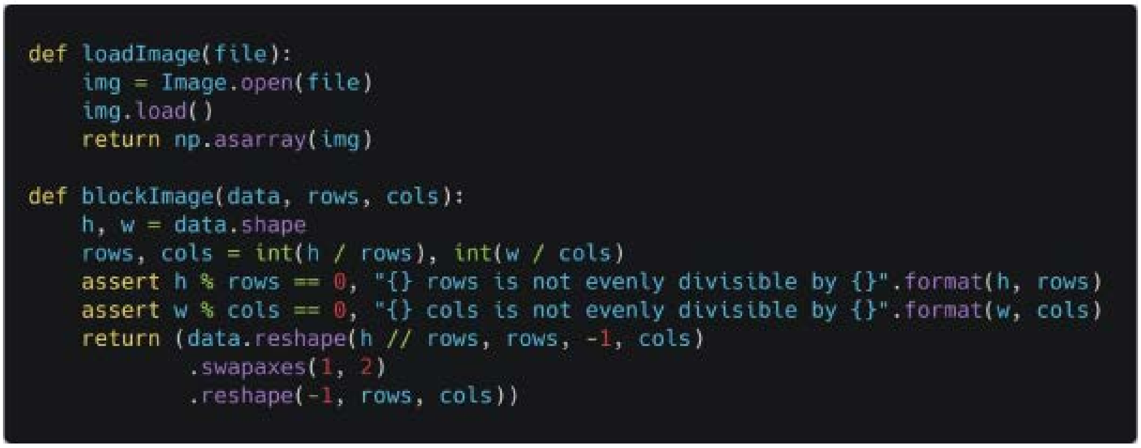
Fig. 1. **Functions for mixing index calculation**

The obtained microphotographs are converted into binary images using the Image module and convert method of the PIL library (Fig. 2).

```
def imageToBMP(file, num):
    img = Image.open(file)
    bw = img.convert(mode="1", dither=Image.NONE)
    return bw.save("%s.bmp" % num)
```

Fig. 2. **Function for image conversion to binary**

The binary images acquired undergo conversion into a data array through the use of the loadImage function in conjunction with the numpy library's asarray method. Following this conversion, the data array is segmented into subarrays, with the blockImage function determining their size. The above functions are shown in Fig. 3.



```
def loadImage(file):
    img = Image.open(file)
    img.load()
    return np.asarray(img)

def blockImage(data, rows, cols):
    h, w = data.shape
    rows, cols = int(h / rows), int(w / cols)
    assert h % rows == 0, "{} rows is not evenly divisible by {}".format(h, rows)
    assert w % cols == 0, "{} cols is not evenly divisible by {}".format(w, cols)
    return (data.reshape(h // rows, rows, -1, cols)
              .swapaxes(1, 2)
              .reshape(-1, rows, cols))
```

Fig. 3. **Functions for converting binary images to a data array**

The suitability of the developed software was determined by its verification. The Chlorella vulgaris microalgae population growth was subjected to evaluation. To assess the biomass distribution homogeneity in the aquatic environment, the image segmentation method was used. We analyzed microalgae Chlorella vulgaris microphotographs population for 4 weeks (Fig. 4).

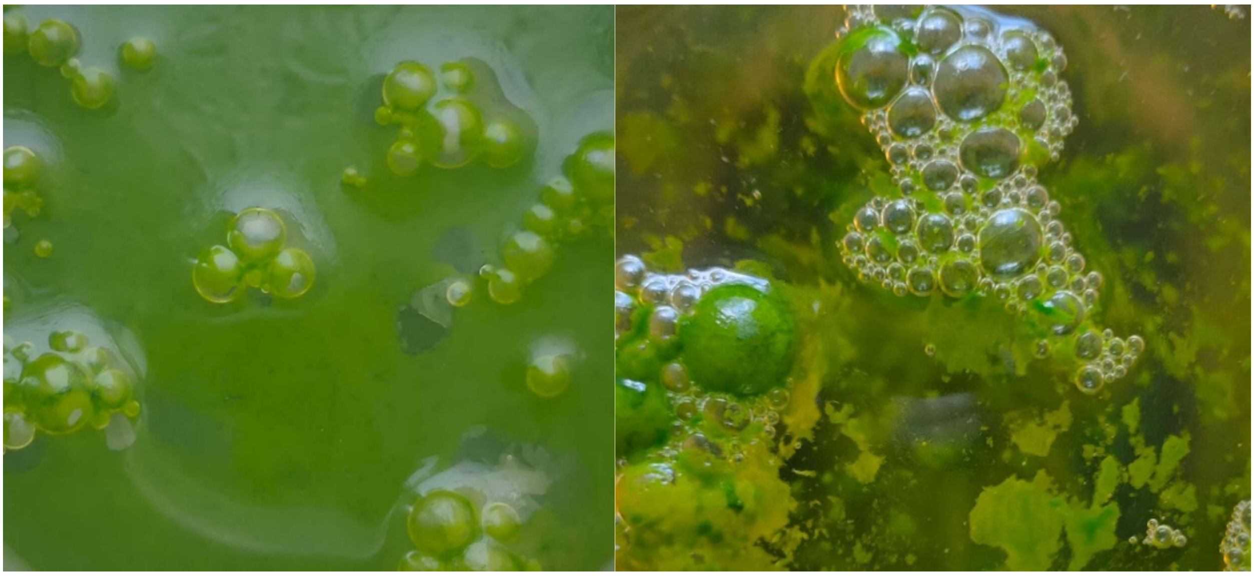


Fig. 4. **Photo of the microalgae Chlorella vulgaris population**

Changes in the microalgae population concentration over 4 weeks can be seen in Figures 5–7.

Fig. 5. **Chlorella vulgaris population week 1**

Fig. 6. **Chlorella vulgaris population week 2–3**

Fig. 7. **Chlorella vulgaris population week 4**

The cluster image analysis method was used to assess the uniform Chlorella vulgaris microalgae distribution population and for further obtained data statistical analysis [9]. The microalgae population micrographs were conditionally divided into a sections specified number and the dispersed phase concentration in each cluster was determined. The the particle distribution uniformity in the microalgae population was estimated by the mixing index (Kn), which is theparticle concentration standard deviation ratio in the clusters (S) to their average concentration in the sample (Pm):

$$Kn = (S / Pm) \times 100\%.$$

Chlorella vulgaris microalgae evaluation biomass results are shown in Fig. 8. The microalgae Chlorela vulgaris in the first week of population growth are characterized by the highest particle distribution heterogeneity (Kn ≈ 93%). The microalgae population heterogeneity at the second week growth (Kn ≈ 85%) is higher than at the third week (Kn ≈ 57%). Comparison of the data shows that the population growth at the fourth week (Kn ≈ 47%) is characterized by the highest homogeneity. The decrease in the Kn value for population growth from one to fourth weeks can be explained by the optimal conditions presence for the Chlorella vulgaris cultivation. The maximum biomass growth, corresponding to 0.5 days, occurs at a temperature of 32.5 °C and at pH range 6.31–6.84.

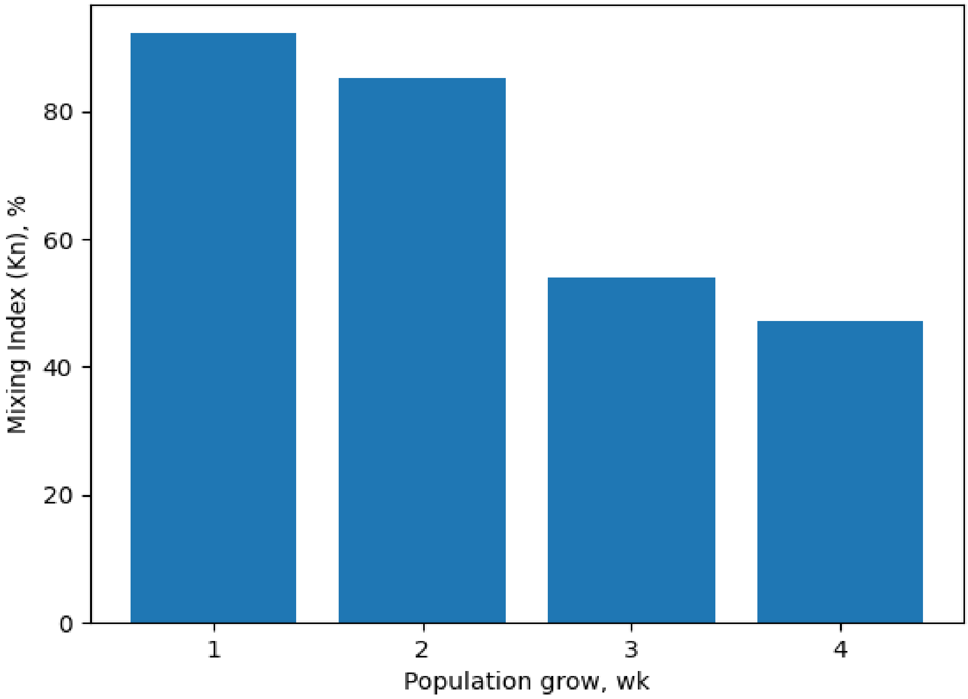


Fig. 8. **Heterogeneity coefficient histogram (Kn) Chlorela vulgaris microalgae biomass in aqueous solution from population changes in weeks (wk)**

It is worth mentioning that Chlorella vulgaris possesses higher quantities of folate and iron compared to other plant-derived foods, making it a potential supplement for mammals, including humans. Studies have reported its diverse pharmacological activities, encompassing immunomodulatory, antioxidant, antidiabetic, antihypertensive, and antihyperlipidemic effects [10].

The evaluation of homogeneity results for the population growth of Chlorella vulgaris microalgae was conducted using the developed software mentioned above. These results were then contrasted with data acquired through the manual homogeneity assessment method, without the aid of computer automation. The findings from this comparison affirm the practical suitability of the developed homogeneity assessment software.

Conclusions. Software that allows assessing the uniformity distribution of Chlorella vulgaris microalgae biomass population growth has been developed. It can be used to study the level of microorganisms' concentration in the aquatic environment. The results of its verification indicate the developed software suitability for practical use. Using the developed software, the influence of Chlorella vulgaris microalgae biomass population growth in the aquatic environment was determined. It is shown that microalgae Chlorella vulgaris population growth from 1 to 4 weeks leads to decrease in the mixing index from 93% to 47%, which is due optimal conditions presence for the cultivation of the microalgae Chlorella vulgaris. The research methodology can be used in the automated monitoring complexes development for the ecological state of water resources and for controlling the industrial production microalgae Chlorella vulgaris population.

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РОЗРОБКА ПРОГРАМНОГО ЗАБЕЗПЕЧЕННЯ ДЛЯ КІЛЬКІСНОЇ ОЦІНКИ ПОПУЛЯЦІЇ МІКРОВОДОРОСТЕЙ CHLORELLA VULGARIS

Мета. *Розробка програмного забезпечення для кількісної оцінки росту популяції мікроводорості Chlorella.*

Методика. *Програмне забезпечення було розроблено за допомогою мови програмування Python та зовнішніх бібліотек: Numpy, Xlsxwriter, Matplotlib та PIL. Визначено придатність розробленого програмного забезпечення для кількісної оцінки росту популяції мікроводорості Chlorella та проведено його верифікацію. Під час верифікації оцінювали ріст популяції мікроводоростей на різних стадіях нарощування біомаси. Зразки мікроводорості Chlorella були отримані протягом декількох тижнів росту популяції.*

Результати. *Розроблено програмне забезпечення, яке дозволяє оцінювати ріст популяції мікроводорості Chlorella у водному середовищі. За допомогою цього програмного забезпечення отримано залежність коефіцієнта гетерогенності приросту біомаси мікроводорості Chlorella vulgaris від часу. Зростання чисельності популяції призводить до зменшення цієї гетерогенності. Цей ефект можна пояснити наявністю оптимальних умов для культивування мікроводорості Chlorella vulgaris.*

Наукова новизна. *Визначено вплив росту популяції мікроводорості Chlorella на гомогенність системи. Встановлено, що збільшення вмісту біомаси Chlorella vulgaris протягом 4 тижнів призводить до зниження коефіцієнта гетерогенності з 92% до 47%, що пов'язано з агрегацією мікроводорості Chlorella vulgaris у водному середовищі.*

Практичне значення. *Розроблене програмне забезпечення дозволяє кількісно оцінювати ріст популяції мікроводорості Chlorella vulgaris у водному середовищі та може бути використане для контролю росту біомаси водоростей у промисловому виробництві.*

Висновок. *Розроблено програмне забезпечення, яке дозволяє оцінити рівномірність розподілу приросту популяції мікроводоростей Chlorella vulgaris у біомасі. Його можна використовувати для дослідження рівня концентрації мікроорганізмів у водному середовищі. Результати його перевірки свідчать про придатність розробленого програмного забезпечення до практичного використання. За допомогою розробленого програмного забезпечення визначено вплив на зростання біомаси мікроводоростей Chlorella vulgaris у водному середовищі. Показано, що зростання популяції мікроводоростей Chlorella vulgaris від 1 до 4 тижнів призводить до зниження індексу змішування з 93% до 47%, що зумовлено наявністю оптимальних умов для культивування мікроводоростей Chlorella vulgaris. Методика дослідження може бути використана при розробці автоматизованих комплексів моніторингу екологічного стану водних ресурсів та контролю промислової продукції мікроводоростей Chlorella vulgaris.*

Ключові слова: *програмне забезпечення екологічного моніторингу; індекс змішування неоднорідності; Chlorella vulgaris.*