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MODIFICATION OF THE METHOD OF SPECTROPHOTOMETRIC DETERMINATION OF CHLOROPHYLL *a* IN THE SUSPENDED MATTER OF WATER BODIES

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This article describes a modification of the spectrophotometric method for the determination of chlorophyll a in seston (suspended matter), which is convenient for hydroecological monitoring. It is proposed to use nuclear filters, as they have a number of advantages over fiberglass and bulk filters. An important difference of our approach from the standard methods is the use of the short-term (up to 30 min) drying of nuclear filters with seston in a desiccator at a temperature of 50–55^{o}C immediately after the suspended matter is deposited on the filter. A comparative analysis of the standard methodology and the proposed modification for assessing the content of chlorophyll and phaeopigments in laboratory cultures of algae (Euglena gracilis, Chlorella vulgaris) and natural plankton from a natural reservoir was carried out.

Keywords: *chlorophyll a, phaeopigments, spectrophotometric method, suspended matter.*

Introduction. One of the key approaches to assessing the productivity and ecological state of aquatic ecosystems is to determine the concentration of chlorophyll *a* (Chl *a*), the main photosynthetic pigment, an integral element of the plant cell. Numerous works of the Belarusian photosynthetic school have been devoted to studying the processes of chlorophyll formation, biogenesis, and organization of the photosynthesis apparatus in plant organisms [1]. Of the available methods for determining chlorophyll — spectrophotometric, fluometric, chromatographic, and remote [2] — the extraction spectrophotometric method, originally developed by Richards and Thompson [3], is mainly used in hydrobiological practice due to its simplicity and accessibility. Over time, the method was optimized and then was recommended as a standard [4] and served as the basis for the development of individual state and international standards [5–7].

In addition to chlorophyll, products of its transformation (pheophytins, pheophorbides, etc.) are constantly present in nature. The content of pheophytin *a* and pheophorbide *a* is comparable with the content of Chl *a*, which must be taken into account when calculating the concentration of the latter. To assess the total content of phaeopigments, the methods described in [8, 9] are usually used. From the ratio of Chl *a* and the products of its decomposition, one can make judgements about the state of the producers, since during the death of the cell, the photosynthetically active chlorophyll decomposes quite quickly to pheophytin, changing the absorption spectrum of the pigment extract [2, 9]. However, since the spectral maxima of the phaeopigments are close to the maxima of chlorophylls, spectrophotometric measurements of the contents of phaeopigments and "pure" chlorophyll in the general extract can be significant, and the results obtained are only approximate [6, 10]. Currently, the determination of Chl *a* concentration in seston is an indispensable element in assessing the ecological state of water bodies, and therefore, much attention is paid to methodological issues [2, 10, 11].

The modification of the spectrophotometric method for determining the concentration of Chl *a* proposed in this work involves the use of nuclear filters to concentrate seston, the simplification of the procedure for preparing the extract, and the possibility of long-term storage of filters until analysis (without significant loss of chlorophyll). The weighing of a filter with a precipitated suspension dried to a constant mass allows for parallel determination of the content of chlorophyll and seston in water on the same filters.

Research Methods. The spectrophotometric method for determining chlorophyll and its derivatives is based on measuring the amount of light absorbed by a solution of pigments in the region of the spectral maximum. Since the extract from seston contains pigments with overlapping absorption spectra, the assessment of their concentrations is complicated. So, to calculate the content of chlorophylls *a*, *b* and *c*, a system of three equations with three unknowns is solved [4]:

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