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THE STORAGE OF ANHYDROBIOTIC CULTURES OF MICROALGAE AND CYANOBACTERIA OF A. O. KOVALEVSKY INSTITUTE OF BIOLOGY OF THE SOUTHERN SEAS OF RAS

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Reliable preservation of microalgae cultures and creation of genetic banks of strains is one of the important tasks of modern biology. To date, 792 collections of various cultivated organisms from 76 countries are registered in the catalog of the World Federation for Culture Collections in the WDCM CCINFO database. This is the most extensive consolidated database of culture collections, which includes both well-known large collections and small repositories of research and educational institutions from all over the world. The database contains 47 algological collections and 80 collections of various microorganisms, which also include microalgae and cyanobacteria cultures. Only 30 biological collections are registered in Russia, from which only 13 contain algae strains. The most common technique of microalgae cultures storage is the method of their periodic re-sowing onto liquid media or agar. It is used in 127 collections (99 % of the total number in the catalog). Other methods used are: cryopreservation – in 33 collections (27 %), lyophilization – in 13 (11 %), L-drying – in 5 (4 %), freezing – in 19 (16 %), and immobilization in alginate beads – in 1 (0.8 %). However, when using these methods, there is a change in morphological and functional features of cells of the cultures stored, as well as their shredding. In addition, cultures maintaining in a viable state is time-consuming and requires expensive equipment. Preservation of microalgae, transferred to the state of anhydrobiosis by dehydration, is simple and cost-effective. Anhydrobiosis is a deep and long-term inhibition of metabolism, reversible under favorable conditions; it is a quite common phenomenon in nature. The only collection in the WDCM CCINFO database that applies the method of transferring cells to a resting state (for soil algae) is the collection of algae cultures of the National University of Kyiv (ACKU WDCM 994). Many years of experiments on the transfer of microalgae to the state of anhydrobiosis allowed us to develop a method of long-term preservation of microalgae without the use of nutrient media. This technique includes cells transfer to the state of anhydrobiosis, their preservation in a dehydrated state, and subsequent removal to an active culture. In order to preserve algological biodiversity, IBSS RAS created a repository of microalgae transferred to the state of anhydrobiosis, which can be converted to active cultures if necessary. The objects of the repository were marine unicellular algae, as well as freshwater and halobic species of lower phototrophs which are perspective for biotechnology and aquaculture. The cultures were obtained as an inoculum from IBSS RAS collection of live cultures of planktonic microalgae. The algae were grown in an accumulative mode under constant lighting. The biomass was collected during cultivation of algologically pure microalgae cultures at the growth retardation or at the stationary stage. Cells were separated from the culture medium by centrifugation or by filtering them on a plankton sieve. Then the algae were dehydrated and maintained in hermetic zipper bags placed in plastic containers of 100 to 500 ml, at a temperature of +18…+21 °C in the dark in a specially equipped room. The main part of the collection is represented by strains from the phyla Chlorophyta, Cyanophyta, Bacillariophyta, and Rodophyta. The list of species, the number of isolates stored, and the information on preservation forms are provided

in this article. The technological regulations for maintenance and replenishment of the storage of anhydrobiotic cultures are described. The repository is at the stage of formation. Its future lies in the fund expansion to include marine, freshwater, and halobic species. Optimization of the dehydration method will allow the transfer of microalgae belonging to different systematic phyla to the state of anhydrobiosis.

Keywords: microalgae, anhydrobiosis, viability, dehydration, storage of microalgae and cyanobacteria

One of the important tasks of modern biology is the reliable preservation of microalgae cultures and the creation of genetic banks of strains. According to the catalog of the World Federation of Cultures, to date 792 collections of various cultivated organisms from 76 countries are registered in the WDCM CCINFO database [27]. The most common technique of microalgae cultures storage is the method of their periodic re-sowing onto liquid media [1, 2, 4, 15, 17, 23] or agar [3, 4, 15]. The catalog contains 47 algological collections and 80 collections of various microorganisms, which also include microalgae and cyanobacteria cultures. The method of periodic re-sowing of microalgae cultures onto liquid media or agar is used in 127 collections (99 % of the total number in the catalog). Other methods used are: cryopreservation – in 33 collections (27 %) [5, 10, 13, 14, 16, 18, 19, 22, 24, 25, 28], lyophilization – in 13 (11 %) [26], L-drying – in 5 (4 %) [21], freezing – in 19 (16 %), and immobilization in alginate beads – in 1 (0.8 %) [11, 12]. However, when using these methods, there is a change in morphological and functional features of cells of the cultures stored, as well as their shredding. In addition, cultures maintaining in a viable state is time-consuming and requires expensive equipment.

Preservation of microalgae, transferred to the state of anhydrobiosis by dehydration, is simple and cost-effective. Anhydrobiosis is a deep and long-term inhibition of metabolism, reversible under favorable conditions. This phenomenon, quite common in nature, formed the basis for the method of transferring cells to a resting state. The only collection in the WDCM CCINFO database applying the method of transferring cells to a resting state for soil algae is the collection of algae cultures of the National University of Kyiv (ACKU WDCM 994) [3].

Many years of experiments on the transfer of microalgae to the state of anhydrobiosis allowed us to develop a method of long-term preservation of microalgae without the use of nutrient media. This technique includes cells transfer to the state of anhydrobiosis, their preservation in a dehydrated state, and subsequent removal to an active culture [6].

The method was tested on prokaryotic and eukaryotic microalgae: on marine, halobic, and freshwater species. The technique is successfully used in IBSS RAS. A collection of anhydrobiotic cultures of lower phototrophs, reversible to a viable state and retaining the ability to divide, was created in the Biotechnology and Phytoresources Department in 2005. The initiator of the collection is PhD R. P. Trenkenshu.

The purpose of creating the collection is the reliable preservation of cultures of lower phototrophs that are also suitable for creating a genetic bank of strains. The practical importance of the repository is connected with the ability to constantly have viable cultures at our disposal to provide experimental research work. In future, it is planned to use the collection as a bank of microalgae and cyanobacteria for conservation of rare and endemic species, algae that are rich in biologically active substances and perspective for application in biotesting, biomonitoring, bioremediation, and scientific and educational process.

MATERIAL AND METHODS

The cultures were obtained as an inoculum from IBSS RAS collection of live cultures of planktonic microalgae [9]. The algae were grown in an accumulative mode under constant lighting. The biomass was collected during cultivation of algologically pure microalgae cultures at the growth retardation stage

or at the stationary stage. Cells were separated from the culture medium by centrifugation at 3000 rpm on a centrifuge OPN-3-UKhL 42 or by filtering them on a plankton sieve 100 PE [20]. The choice of filtration method depended on the size of the cells and the trichomes of the lower phototrophs.

The preparation for the preservation was carried out in three ways:

1) to remove salts, the cells were washed from the culture medium first with a solution of ammonium carbonate and then with distilled water, and after this, the cells were dehydrated;
2) the algae to be stored for a long period were dehydrated along with the culture medium;
3) the cells of lower phototrophs were suspended with protectors.

For each microalga, the preservation was carried out in different ways to be able to conduct a comparative analysis of the cells viability of the lower phototrophs stored for a long period and to identify the optimal method.

At the initial stage, the microalgae were dehydrated at a temperature of +20…+70 °C in increments of 10 °C. After a series of experiments, the temperature range of +30…+40 °C was selected [8]. The control of the residual moisture level was carried out during drying; its values were in the range of 10–17 % for most dehydrated cells [7]. The dehydrated algae were maintained in hermetic zipper bags placed in plastic containers of 100 to 500 ml, at a temperature of +18…+21 °C in the dark in a specially equipped room.

The repository consists of a box and a room that both serves as an incubation-stabilizing space in front of the entrance to the storage and is intended for handling samples. An air conditioner is installed in the box to dry the air and to maintain the set temperature.

All samples are labeled with information on name of the culture, dehydration conditions (temperature and duration), and date of transfer to the state of anhydrobiosis.

RESULTS AND DISCUSSION

The first collection samples were spirulina tablets given by manufacturers (“Agro-Viktoriya” LTD) and purchased in pharmacies of the city. The cyanoprokaryotic tablets were reactivated, adapted to cultivation conditions, and transferred to intensive culture. Then, the cultures were subject to re-putting into the suspended animation. The method was tested on microalgae of different phyla.

To date, 366 samples of dehydrated microalgae cultures from 4 phyla have been put for long-term maintenance: cyanobacteria Chlorophyta (Cyanobacteria), green microalgae Cyanophyta, red algae Rodophyta, and diatom algae Bacillariophyta (Fig. 1).

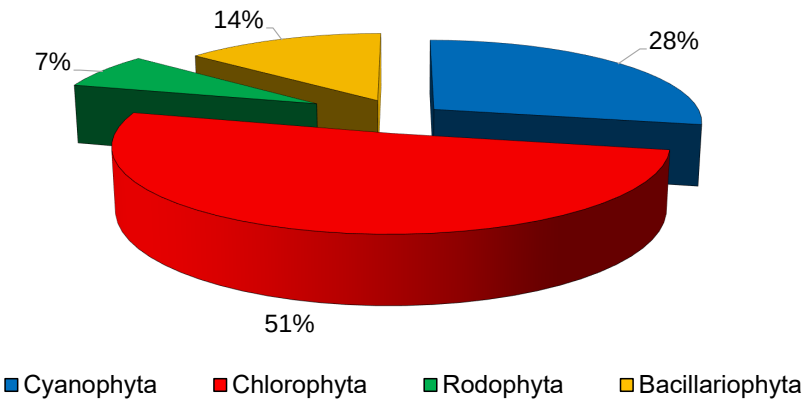

Fig. 1. Ratio of the number of algae strains from different phyla in IBSS RAS collection of anhydrobiotic cultures

The main part of the collection is represented by strains from the phylum Chlorophyta and contains species *Dunaliella salina* (Dunal) Teodorescu, 1905, *Tetraselmis viridis* Rouchijajnen, 1966, *Chlorella vulgaris* f. *suboblonga* V. M. Andreeva, 1975, *Chlorella* sp., and *Scenedesmus* sp. Phylum Cyanophyta is represented by four species (*Arthrospira (Spirulina) platensis* (Nordstedt) Gomont, 1892, *Synechococcus elongates* (Nägeli) Nägeli, 1849, *Oscillatoria amoena* (Kutzing) Gomont, 1892, and *Nostoc commune* var. *flagelliforme* Bornet & Flahault, 1886); phylum Bacillariophyta – by two species (*Phaeodactylum tricornutum* Bohlin, 1897 and *Cylindrotheca closterium* (Ehrenb.) Reimann et Lewin, 1964); phylum Rodophyta – by one species (*Porphyridium purpurerum* (Bory de Saint-Vincent) Drew and Ross, 1965) (Table 1).

Table 1. Taxonomic diversity of cyanobacteria and algae in IBSS RAS collection of anhydrobiotic cultures

Phylum	Order	Genus	Species	When and where from collected/identified	Number of samples stored
CHLOROPHYTA	Chlamydomonadales	*Dunaliella*	*Dunaliella salina*	Salt lakes of Syvash (Crimea)	126
	Sphaeropleales	*Scenedesmus*	*Scenedesmus* sp.	Accompanying the cultivation of chlorella	3
	Chlorodendrales	*Tetraselmis*	*Tetraselmis viridis*	The Black Sea	37
	Chlorellales	*Chlorella*	*Chlorella vulgaris*	The Institute of Botany (Kyiv, Ukraine)	12
			Chlorella sp.	"Ikhlyas-agroenergiya" LTD	20
CYANOBACTERIA	Oscillatoriales	*Oscillatoria*	*Oscillatoria amoena*	Identified while cultivating *Spirulina platensis*	2
			Spirulina (Arthrospira) platensis	MSU (Sochi)	94
	Synechococcales	*Synechococcus*	*Synechococcus elongates*	Accompanying the cultivation of spirulina	3
	Nostocales	*Nostoc*	*Nostoc commune*	The Institute of Botany (Kyiv, Ukraine)	3
BACILLARIOPHYTA	Bacillariales	*Phaeodactylum*	*Phaeodactylum tricornutum*	The Black Sea	23
		Cylindrotheca	*Cylindrotheca closterium*	Algobank (Caen, France), the Mediterranean Sea	3
RHODOPHYTA	Porphyridiales	*Porphyridium*	*Porphyridium purpurerum*	BRI (Saint Petersburg)	74

Microalgae and cyanobacteria stored were transferred to state of anhydrobiosis at different dehydration modes, including temperature and duration of dehydration. Samples of the same algae and cyanobacteria were dehydrated in different years; they were dehydrated with and without various protectors. This is due to the fact that the storage time limits have not yet been determined. Forms of lower phototrophs preservation are presented in Figs 2 and 3. In order to determine the physico-chemical changes of microalgae depending on the storage period, the aliquots of cultures were periodically removed from the collection, and their biochemical control and reactivation were carried out.

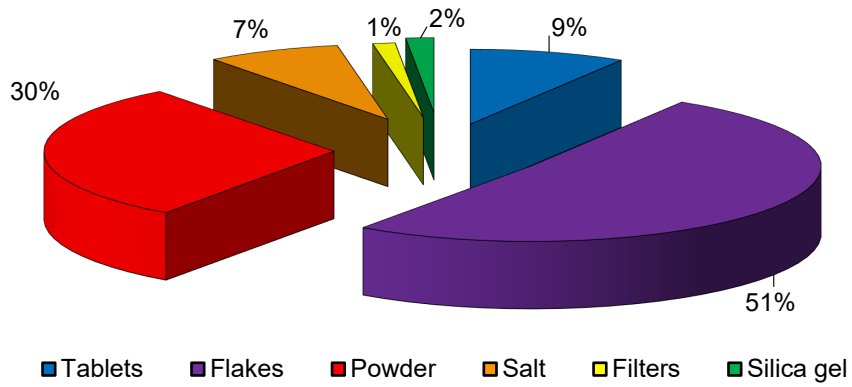

Fig. 2. Forms of microalgae and cyanobacteria preservation for long-term storage

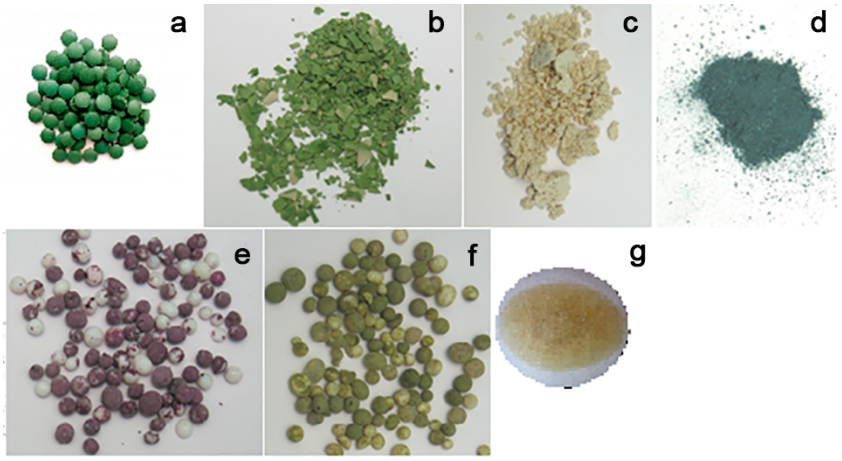

Fig. 3. Appearance of dehydrated samples of microalgae and cyanobacteria stored: a – tablets; b – flakes; c – salt; d – powder; e, f – silica gel; g – filters

In order to preserve the cultures of the lower phototrophs, the following technological regulations for servicing and replenishing the collection of anhydrobiotic cultures was applied (Table 2).

Optimization of the method makes it possible to transfer microalgae belonging to various systematic phyla to the state of anhydrobiosis. The technique can be recommended for use in scientific and educational institutions, as well as in biotechnology, where long-term preservation of strains of museum cultures is required. At this stage, IBSS RAS repository is unique and has no analogues. The collection of anhydrobiotic cultures is constantly replenished with new species of lower phototrophs.

Conclusion. IBSS RAS collection of anhydrobiotic cultures is at the stage of formation. Its future lies in the fund expansion to include marine, freshwater, and halobic species. The development of individual protocols for dehydration and reactivation will make it possible to transfer microalgae belonging to different systematic phyla to the state of anhydrobiosis.

Table 2. Technological regulations for servicing and replenishing the collection of anhydrobiotic cultures of microalgae and cyanobacteria

Stage number	Stage name	Manipulations carried out
I	Obtaining the anhydrobiotic culture	• Obtaining an algologically pure culture from a natural population; • culture certification; • culture adaptation to artificial growing conditions; • intensive cultivation; • transfer of culture to the state of anhydrobiosis; • preparation for long-term storage.
II	Biochemical control of microalgae and cyanobacteria species preserved	Complex biochemical analysis of the lower phototrophs to be stored for a long period (determination of the content of chlorophylls, total carotenoids, total proteins, carbohydrates, lipids, and nucleic acids).
III	Storage and control of viability of cultures preserved	• Identification of living and dead cells of lower phototrophs; • biochemical control of samples preserved; • reactivation and assessment of ability to grow on liquid media.
IV	Maintaining a catalog of anhydrobiotic cultures	Development of the electronic catalog. It includes information on strain name, number, preservation form, date of transferring to the state of anhydrobiosis and dehydration conditions, data on cultivation (including growth characteristics), results of biochemical analysis before the preservation and during the storage, information on previous reactivation, and morphological and biochemical characteristics of algae after reactivation.

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REFERENCES

1. Aizdaicher N. A. Collection of marine microalgae at the A. V. Zhirmunsky Institute of Marine Biology. *Biologiya morya*, 2008, vol. 34, no. 2, pp. 152–155. (in Russ.)
2. Vladimirova M. G., Ignat'evskaya M. I. Izuchenie vliyaniya uslovii khraneniya kul'tur *Chlorella* v kollektsii na posleduyushchuyu ikh produktivnost'. *Mikrobiologiya*, 1966, vol. 35, no. 3, pp. 539–548. (in Russ.)
3. Kostikov I. Yu., Demchenko E. N., Berezovskaya M. A. Microalgae culture collection at the Taras Shevchenko National University, Kyiv. Catalogue of strains (2008). *Chornomorskyi botanichnyi zhurnal*, 2009, vol. 5, no. 1, pp. 37–79. (in Russ.)
4. Novakovskaya I. V., Patova E. N. Collection of living microalgae strains of the Institute of Biology, Komi Science Centre, Ural Branch, RAS, and its perspective using. *Izvestiya Komi nauchnogo tsentra UrO RAN*, 2012, iss. 2, no. 10, pp. 36–41. (in Russ.)
5. Odintsova N. A., Boroda A. V. Cryopreservation of the cells and larvae of marine organisms. *Biologiya morya*, 2012, vol. 38, no. 2, pp. 93–103. (in Russ.)
6. Pat. 2541452 Rossiiskaya Federatsiya. MPK6 S 12 N 1/04. *Sposob dlitel'nogo khraneniya mikrovodoroslei* / Kharchuk I. A. ; zayavitel' i patentoobladatel' Federal'noe gosudarstvennoe byudzhetnoe uchrezhdenie nauki "Institut morskikh biologicheskikh issledovanii imeni A. O. Kovalevskogo RAN". No. 2014149881/93; zayavl. 26.09.2014; prior. 17.03.2008; opubl. 10.02.2015, Byul. no. 4. (in Russ.)

7. Kharchuk I. A. Khranenie mikrovodoroslei v sostoyanii angidrobioza. In: *Mikrovodorosli Chernogo morya: problemy sokhraneniya bioraznoobraziya i biotekhnologicheskogo ispol'zovaniya* / Yu. N. Tokarev, Z. Z. Finenko, N. V. Shadrin (Eds). Sevastopol : EKOSI-Gidrofizika, 2008, chap. 9, pp. 237–267. (in Russ.)
8. Kharchuk I. A. Dynamics of viability and components of biochemical composition *Arthrospira* (*Spirulina*) *platensis* (Nords) Gomont depending on the dehydration temperature transferring at anhydrobiosis state. *Voprosy sovremennoi al'gologii*, 2018, no. 1 (16). URL: http://algology.ru/1258 (accessed 23.07.2019). (in Russ.)
9. *Kollektsiya mikrovodoroslei otdela ekologicheskoi fiziologii vodoroslei FITs InBYuM* [Electronic resource] : site. URL: http://ibss-ras.ru/?page_id=2828 (accessed 24.07.2019). (in Russ.)
10. Cañavate J. P., Lubian L. M. Relationship between cooling rates, cryoprotectant concentrations and salinities in the cryopreservation of marine microalgae. *Marine Biology*, 1995, vol. 124, iss. 2, pp. 325–334. https://doi.org/10.1007/BF00347136
11. Chen Y. C. Immobilized microalgae *Scenedesmus quadricanda* (Chlorophyta, Chlorococcales) for long-term storage and for application for water quality control in fish culture. *Aquaculture*, 2001, vol. 195, no. 1–2, pp. 71–80. https://doi.org/10.1016/S0044-8486(00)00540-8
12. Chen Y. C. Immobilized *Isochrysis galbana* (Haptophyta) for long-term storage and applications for feed and water quality control in clam (*Meretrix lusoria*) cultures. *Journal of Applied Phycology*, 2003, vol. 15, no. 5, pp. 439–444. https://doi.org/10.1023/A:1026071714199
13. Crutchfield A., Diller K., Brand J. Cryopreservation of *Chlamydomonas reinhardtii* (Chlorophyta). *European Journal of Phycology*, 1999, vol. 34, iss. 1, pp. 43–52. https://doi.org/10.1080/09670269910001736072
14. Day J. G., Watanable M. M., Morris G. H., Fleck R. A., McLellan M. R. Long-term viability of preserved eukaryotic algae. *Journal of Applied Phycology*, 1997, vol. 9, no. 2, pp. 121–127. https://doi.org/10.1023/A:1007991507314
15. Hur S. B., Bae J. H., Youn J., Jo M. J. KMMCC – Korea Marine Microalgae Culture Center: List of strains, 2nd edition. *Algae*, 2015, vol. 30, suppl., pp. S1–S188. http://dx.doi.org/10.4490/algae.2015.30.S.S1
16. Kumari N., Gupta M., Singh R. Open encapsulation-vitrification for cryopreservation of algae. *Cryobiology*, 2016, vol. 73, iss. 2, pp. 232–239. https://doi.org/10.1016/j.cryobiol.2016.07.005
17. Marsalek B., Rojickova-Padrtova R. Long-term maintenance of alga strains for use in biomassays and biotechnology. *Archiv fur̈ Hydrobiologie. Supplementband: Algological Studies*, 1988, vol. 124, pp. 121–136.
18. Meyer M. A. *Cryopreservation of a Marine Diatom*. PhD Thesis. Texas A & M University, College Station, 1985, 112 p.
19. Poncet J.-M., Véron B. Cryopreservation of the unicellular marine alga, *Nannochloropsis oculata*. *Biotechnology Letters*, 2003, vol. 25, no. 23, pp. 2017–2022. https://doi.org/10.1023/B:BILE.0000004395.04116.45
20. Richmond A., Lichtenberg E., Stahl B., Vonshak A. Quantitative assessment of the major limitations on productivity of *Spirulina platensis* in open raceways. *Journal of Applied Phycology*, 1990, vol. 2, no. 3, pp. 195–206. https://doi.org/10.1007/BF02179776
21. Sakane T. Preservation of microorganisms by L-drying. *International Journal of Reefing*, 1982, vol. 57, no. 6, pp. 767–775.
22. Scarbrough C., Wirschell M. Comparative analysis of cryopreservation methods in *Chlamydomonas reinhardtii*. *Cryobiology*, 2016, vol. 73, iss. 2, pp. 291–295. https://doi.org/10.1016/j.cryobiol.2016.07.011
23. von Schwartzenberg K., Bornfleth S., Lindner A., Hanelt D. The Microalgae and Zygnematophyceae Collection Hamburg (MZCH) – living cultures for research on rare streptophytic algae. *Algological Studies*, 2013, vol. 142, pp. 77–108. http://doi.org/10.1127/1864-1318/2013/0131
24. Surek B. Meeting report: International Symposium on the Cryopreservation of Algae (Austin, Texas, USA, 16–17 April, 1988). *Protist*, 1998, vol. 149,

iss. 3. pp. 201–205. https://doi.org/10.1016/S1434-4610(98)70026-4

25. Tessarolli L. P., Day J. G., Vieira A. H. Establishment of a cryopreserved biobank for the Culture Collection of Freshwater Microalgae (CCMA-UFSCar), São Paulo, Brazil. *Biota Neotropica*, 2017, vol. 17, no. 2, e20160299. http://dx.doi.org/10.1590/1676-0611-bn-2016-0299
26. Tsuru S. Preservation of marine and fresh water algae by means of freezing and freeze-drying. *Cryobiology*, 1973, vol. 10, iss. 5, pp. 445–452. https://doi.org/10.1016/0011-2240(73)90074-6
27. *WDCM CCINFO. World Data Centre for Microorganisms, Culture Collections Information Worldwide. 2014.* [Electronic resource.] URL: http://www.wfcc.info/ccinfo/home (accessed 23.07.2019).
28. Zheng L., Lu Z., Zhang Q., Li T., Song L. A fluorescence ratio-based method to determine microalgal viability and its application to rapid optimization of cryopreservation. *Cryobiology*, 2018, vol. 81, pp. 27–33. https://doi.org/10.1016/j.cryobiol.2018.02.014

ХРАНИЛИЩЕ АНГИДРОБИОЗНЫХ КУЛЬТУР МИКРОВОДОРОСЛЕЙ И ЦИАНОБАКТЕРИЙ ИНСТИТУТА БИОЛОГИИ ЮЖНЫХ МОРЕЙ ИМЕНИ А. О. КОВАЛЕВСКОГО РАН

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Надёжное сохранение культур микроводорослей и создание генетических банков штаммов — одна из важных задач современной биологии. В каталоге Всемирной федерации культур в базе WDCM CCINFO на сегодняшний день зарегистрировано 792 коллекции различных культивируемых организмов из 76 стран. Это самая обширная сводная база данных, включающая как известные крупные коллекции, так и небольшие хранилища исследовательских и образовательных учреждений со всего мира. В базе представлено 47 альгологических коллекций и 80 коллекций микроорганизмов, которые также включают культуры микроводорослей и цианобактерий. В России зарегистрировано всего 30 биологических коллекций; фонды только 13 из них включают штаммы водорослей. Самый распространённый способ хранения культур микроводорослей — метод их периодических пересевов на жидкие среды или агар. Его используют в 127 коллекциях (99 % от общего количества в каталоге). Также применяют криоконсервацию — в 33 коллекциях (27 %), лиофилизацию — в 13 (11 %), L-высушивание — в 5 (4 %), замораживание — в 19 (16 %), иммобилизацию в альгинатных бусинках — в 1 (0,8 %). Между тем при использовании этих методов изменяются морфологические и функциональные свойства клеток сохраняемых культур и происходит их измельчание. Кроме того, поддержание культур в жизнеспособном состоянии трудоёмко и требует дорогостоящего оборудования. При этом хранение микроводорослей, переведённых в состояние ангидробиоза путём их обезвоживания, просто и экономически выгодно. Ангидробиоз — глубокое и длительное торможение метаболизма, обратимое при благоприятных условиях; это достаточно распространённое явление в природе. Единственная коллекция из базы WDCM CCINFO, для которой применяют способ перевода клеток в покоящееся состояние путём ангидробиоза (для почвенных водорослей) — коллекция культур водорослей Киевского национального университета (ACKU WDCM 994). Многолетние опыты по переводу микроводорослей в состояние ангидробиоза позволили разработать метод их длительного хранения без использования питательных сред, включающий перевод клеток в состояние ангидробиоза, их сохранение в дегидратированном состоянии и последующее выведение в активную культуру. С целью поддержания альгологического биоразнообразия на базе ФИЦ ИнБЮМ создано хранилище микроводорослей, переведённых в состояние ангидробиоза; их при необходимости можно вывести в активные культуры. Объектами стали морские одноклеточные водоросли, а также пресноводные и галобные виды низших фототрофов, перспективные для аквакультуры и биотехнологии. Культуры получены в виде инокулята из коллекции живых культур планктонных микроводорослей ФИЦ ИнБЮМ.

Водоросли выращивали в накопительном режиме при постоянном освещении. Биомассу собирали во время культивирования альгологически чистых культур микроводорослей на стадии замедления роста или на стационарной стадии. Клетки отделяли от культуральной среды центрифугированием или путём их фильтрации на планктонном сите. Затем водоросли обезвоживали и хранили в герметичных зиплок-пакетах, помещённых в пластиковые ёмкости объёмом от 100 до 500 мл, при температуре +18…+21 °С в темноте в специально оборудованном помещении. Основная часть коллекции представлена штаммами из отделов Chlorophyta, Cyanophyta, Bacillariophyta, Rodophyta. В статье приведены список видов и количество сохраняемых изолятов, представлена информация о формах хранения, описан технологический регламент обслуживания и пополнения хранилища ангидробиозных культур. Хранилище находится на стадии формирования. Его будущее связано с расширением фонда за счёт морских, пресноводных и галобных видов. Оптимизация способа обезвоживания позволит перевести в состояние ангидробиоза микроводоросли, относящиеся к разным систематическим отделам.

Ключевые слова: микроводоросли, ангидробиоз, жизнеспособность, дегидратация, хранение микроводорослей и цианобактерий