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# Influence of a Constant Magnetic Field on the Infectious Titer of Black Sea Algal Viruses

O. A. Stepanova<sup>*a*, \*</sup>, P. V. Gaisky<sup>*b*, \*\*</sup>, and S. A. Sholar<sup>*b*, \*\*\*</sup>

<sup>a</sup> Institute of Natural and Technical Systems of Russian Academy of Sciences, Sevastopol, 299011 Russia <sup>b</sup> Federal Research Center "Marine Hydrophysical Institute of Russian Academy of Sciences," Sevastopol, 299011 Russia

\*e-mail: solar-ua@ya.ru

\*\*e-mail: gaysky@inbox.ru

\*\*\*e-mail: sa.sholar@mail.ru

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Abstract—The change in the infectious titer of strains of the Black Sea algal viruses of three species of microalgae (*Tetraselmis viridis*—strain TvV-S21-1, *Dunaliella viridis*—strain DvV-S20-1 and *Phaeodactylum tricornutum*—strain PtV-S20-1) after incubation in a constant unidirectional magnetic field with a magnetic induction of 60 mT was studied experimentally. The studies were carried out using a laboratory installation with a duration of experiments ranging from 24–72 h to 6–8 days. A decrease in the infectious titer of algal viruses by 1–2 orders (10–100 times) was recorded after a magnetic load.

**Keywords:** Black Sea algal viruses of microalgae, *Tetraselmis viridis*, *Dunaliella viridis* and *Phaeodactylum tricornutum*, viral infectious titer, constant unidirectional magnetic field **DOI:** 10.1134/S000635092202021X

The rapid development of marine (aquatic) virology since the end of the 20th century has revealed a number of new directions of study, a brief description of which was presented in our publication [1]. Although the importance of natural aquatic (marine) viruses, the most numerous among hydrobionts, in the ecology of the hydrosphere of our planet is obvious, it has not been studied enough and the relevance of research in this direction is increasing. According to scientists, understanding the role of viruses in the organic carbon cycle in the hydrosphere and in the functioning of marine food chains, as well as in the biodiversity of unicellular aquatic organisms, which are hosts of aquatic (marine) viruses serves as the basis for assessing the stability of marine ecosystems and increases the predictability of the impact of global changes on biogeochemical processes in the world ocean [2]. Thus, the role of hydrosphere viruses in the ecology of our planet is clearly indicated in the title of a study of a Canadian scientist-"Marine virusesmajor players in the global ecosystem" [3].

It is logical to assert that changes in the viral communities of water bodies, caused by various biotic and, in particular, abiotic factors, due to the magnitude and prevalence of aquatic (marine) viruses, will lead to unpredictable consequences, taking into account their cascading impact on biogeochemical cycles, food chains and the metabolic balance of the world ocean [4, 5].

One of the tasks in studying the ecology of aquatic viruses is to analyze the impact of environmental factors on the viral component of the hydrosphere, including on its individual representatives [1, 4]. It is known that under natural conditions any biological object is located in a ubiquitous geomagnetic field, the basis of which is a relatively stable constant magnetic field  $(35-65 \ \mu\text{T})$ . Changes in the constant magnetic field (geomagnetic variations) can be caused by the impact of the solar plasma flow (solar wind), which intensifies in years of increased solar activity [6, 7]. In our studies, for the first time, a negative correlation between the abundance of marine viruses in the Black Sea viral plankton and the solar activity index was established, which probably reflects cause-and-effect relationships, caused, among other factors, by the action of increased ultraviolet background during active Sun years and an increase in the values of geomagnetic variations using the example geomagnetic index  $A_p$  [8, 9]. Data from the literature provided knowledge about the influence of a constant magnetic and alternating electromagnetic field on the reproduction of viruses and were used as the basis for proposals for the practical application of this physical factor [10–14]. Since information about the influence of the magnetic field on representatives of the marine viral component, including Black Sea algal viruses, is limited, our first studies were devoted to identifying the response by assessing the infectious titer of the Black Sea microalgal virus Tetraselmis viridis (strain TVV-SI1)



**Fig. 1.** (a) Scheme of a laboratory installation developed based on the laboratory of hydrophysical and bioelectronic measuring systems and technologies of the Center for Ecological Instrumentation and Eco-Energy of Institute of Natural and Technical Systems of the Russian Academy of Sciences, which creates an artificial constant magnetic field with a magnetic induction of 60 mT; (b) photograph of the laboratory installation.

to an additional magnetic load under experimental conditions [15]. The results obtained indicated a decrease in the infectious titer of the strains of algal viruses used in the experiment under the influence of the magnetic load.

The aim of our further studies, the results of which are presented here, was the investigation of the effect of magnetic load (constant unidirectional magnetic field) of different duration on the infectious titer of strains of the Black Sea algal viruses of three species of microalgae.

#### MATERIALS AND METHODS

The influence of a unidirectional constant magnetic field on the viral titer of the Black Sea algal viruses was studied experimentally using a laboratory installation based on the laboratory of hydrophysical and bioelectronic measuring systems and technologies of the Center for Ecological Instrumentation and Ecoenergy of the Institute of Natural and Technical Systems [15].

The basis of the laboratory installation was two neodymium magnetic disks rigidly fixed at a distance of 5 cm (50 mm in diameter with a height of 10 mm), between which a constant unidirectional magnetic field with a measured value of magnetic induction of 60 mT was created. In the space between the magnetic disks, the experimental samples of the viral suspensions were placed in glass bacteriological test tubes in a volume of 2.0 mL. The incubation of the studied material in the magnetic field ranged from 24–48 h to 6-8 days. As a control (without magnetic load), samples of viral suspensions were used, placed under conditions of illumination and temperature identical to those of the experimental conditions.

A diagram and a photograph of the developed laboratory installation, which creates an artificial constant magnetic field, are shown in Fig. 1.

Taking into account the fact that it was assumed in the experiments performed and described earlier [15] that the viral suspension may contain infected but not yet lysed host cells (microalgae), the viral suspensions used were purified from large particles (non-lysed cells) by ultrafiltration through nitrocellulose filters manufactured by Sartorius (Germany) with a pore diameter of 300 nm. Thus, the methodological part of the experiments described earlier [15] was corrected. The ultrafiltration of viral suspensions, further used for both experimental and control samples, was performed using a sterile device—a syringe and a filter holder with nitrocellulose filters.

For titration (determination of infectious titer), based on a tenfold dilution of experimental and control samples of suspensions of algal viruses, as described in [16], three liquid cultures of Black Sea microalgae were used: *Tetraselmis viridis* (Rouchijajnen. R.E. Norris, Hori & Chihara, 1980), *Dunaliella viridis* (Teodoresco, 1905) and *Phaeodactylum tricornutum* (Bohlin, 1897). The cultures were obtained from the Department of Ecological Physiology of Algae, Kovalevsky Institute of Biology of the Southern Seas of the Russian Academy of Sciences.

In the experiments, we used viral suspensions of algal viruses from the author's collection [16] algal viruses *T. viridis* (strain TVV-S2I-1), *D. viridis* (strain

Strain of the studied algal virus		Duration of incubation of the viral suspension of the studied algal virus strain in a magnetic field (experiment) and the infectious titer in IE/mL				Changes in infectious titer in the experiment compared
		24 h	48 h	72 h	6–8 days	
TvV-S21-1	Experiment	n/s	10 <sup>3</sup>	10 <sup>2</sup>	n/s	Decrease by 10 and 100 times after 48 and 72 h
	Control	n/s	104	104	n/s	
DvV-S20-1	Experiment	106	n/s	10 <sup>5</sup>	10 <sup>5</sup>	Decrease by 10 and 100 times after 24, 72 h and 6–8 days
	Control	107	n/s	107	107	
PtV-S20-1	Experiment	10 <sup>6</sup>	n/s	104	104	Decrease by 100 times after 72 h
	Control	106	n/s	10 <sup>6</sup>	10 <sup>6</sup>	and 6–8 days

**Table 1.** Indicators of the infectious titer of three Black Sea algal viruses of microalgae cultures *Tetraselmis viridis* (strain TVV-S21-1), *Dunaliella viridis* (strain DvV-S20-1) and *Phaeodactylum tricornutum* (strain PtV-S20-1)

n/s-studies were not conducted.

DvV-S20-1) and *R. tricornutum* (strain PtV-S20-1). The morphology, electron photographs, and properties of these Black Sea algal viruses, as well as data from the analysis of sequenced genomes of some viral strains, are described in [16, 17].

For each strain of the algal viruses, two or more experiments were carried out: two experiments for the algal virus strain *T. viridis* and four experiments each for algal virus strain *D. viridis* and *P. tricornutum*, during the incubation in a magnetic field for 6 to 8 days, two experiments were performed.

#### **RESULTS AND DISCUSSION**

The results of the experiments on changes in the infectious titer of Black Sea algal viruses *T. viridis* (TvV-S21-1), *D. viridis* (DvV-S20-1) and *P. tricornu-tum* (PtV-S20-1) are presented in the Table 1.

As can be seen from the data, in control experiments, algal viruses with different initial infectious titers were used for research  $-10^4$ ,  $10^6$  and  $10^7$  infectious units per 1 mL (IE/mL) for the TvV-S21-1. PtV-S20-1 and DvV-S20-1 strains, respectively. Such values of infectious titers used in experiments with Black Sea algal viruses are close to the natural values during the peak seasons of their abundance [16]. It was previously found that infectious titers in stored viral suspensions at room temperature and light, which correspond to the same conditions in the experiments in control and are identical to the conditions in the experiments, usually do not change for one month or more [16]. However, after the incubation in a magnetic field for 24 h, a 10-fold decrease in infectious titer (by one order of magnitude) was revealed for the algal virus D. viridis (strain DvV-S20-1) and after 72 h of magnetic load it decreased by 100-fold (by two orders of magnitude). Further exposure to the magnetic field within 6 to 8 days did not cause a decrease in the infectious titer, which remained at the level of  $10^5 \text{ IE/mL}$ , as it was after 72 h of exposure.

For the algal virus *T. viridis* strain TvV-S21-1 there was a decrease in the infectious titer by 10 and 100 times after 48 and 72 h, respectively. Determination of the infectious titer 24 h after magnetic load, as well as after 6 to 8 days for this algal virus was not performed.

Changes in the infectious titer were not recorded for the algal virus *P. tricornutum* strain PtV-S20-1 after 24 h in a magnetic field. Studies after 48 h were not performed. However, after 72 h of magnetic load, a decrease in the infectious titer in the PtV-S20-1 strain by two orders of magnitude (100 times) was observed. The same value of the decrease in infectious titer (100 times compared with the control) was seen after 6 days.

Thus, it was recorded that after 24–48 h of exposure to a constant magnetic field with a magnetic induction of 60 mT, the infectious titer of the studied Black Sea algal viruses decreased by one order for DvV-S20-1 and TvV-S21-1 and did not change for PtV-S20-1. A longer exposure to a magnetic field (72 h or more) led to a decrease in the infectious titer by two orders of magnitude (for DvV-S20-1 and PtV-S20-1).

Our results indicate the role of the magnetic field in reducing the infectious titer in natural Black Sea algal viruses and do not contradict the data of other researchers who have identified an inhibitory effect of magnetic and electromagnetic fields on viral reproduction [10, 11]. It has also been recorded that some effects of magnetic field exposure with an increase in magnetic induction may not differ from the data of control samples without a magnetic load [18], which corresponds to our observations on the invariance of the infectious titer of algal viruses after 72 h with an increase in the duration of the magnetic load (6–8 days). The influence of magnetic fields on biological systems and objects has been known since the time of Hippocrates, their miraculous properties were described by doctors from ancient China, India, and Egypt [19]. The accumulated knowledge about the impact of magnetic and electromagnetic fields on representatives of the biosphere with different organization - from microorganisms and viruses to higher vertebrates allowed a number of facts new to science to be established. However, the data obtained by scientists indicate the need for further research, since knowledge about the effects of magnetic and electromagnetic fields on biological objects, including viruses, seems to be relevant and promising both in theoretical and practical terms [6, 10-15, 18-22].

According to researchers [23–25], a magnetic field can affect the probability of formation of peroxyl radicals in biological systems, a change in the concentration of which leads to a change in the concentrations of reactive oxygen species (ROS) involved in the formation of the response of a biological system to an external magnetic effect and triggering biological mechanisms that lead to a change in metabolism. Reactive oxygen species are known to be trigger molecules for many biological processes. The key links of metabolism can be influenced by the control of their concentration by a weak external magnetic field. Thus, hydrogen peroxide and other reactive oxygen species, formed as a result of the action of a magnetic field, should be considered as damaging factors for the biological component of the planet, including marine viruses (algal viruses).

The antiviral activity of trace elements in a low oxidation state was also reported in a recently published review [26], where the mechanism of the antiviral action of microelements is explained by the suppression of the electrostatic interaction of the virus (its adsorption capacity) with the cell by neutralizing the charges on the surface of the virus and the cell. The authors of the review suggest the possibility of using trace elements in a low oxidation state for the treatment of viral infections, since "many potential targets that are inaccessible to antibodies and other large molecules are easily accessible to micronutrients."

Now, due to the outbreak of the Covid-19 pandemic, studies on the role of magnetic and electromagnetic fields in changing the reproduction of viruses are of particular interest [10–14, 26]. The relevance of the results obtained in this case is not only the use of such knowledge as the basis for new scientific hypotheses that explain the activation of viral processes on our planet, but also contribution to the development of new methods for the prevention and control of viral diseases, using physical methods for inhibition of viral reproduction [26–28]. Therefore, based on the current situation with the Covid-19 pandemic and the expected activation of other viral infections on our planet [27, 28], ongoing studies on the effect of magnetic load on the viral titer of natural marine viruses (algal viruses) are relevant. They will continue to be used in experiments of a wider range of methods of electromagnetic influence and a wider range of algal viruses (viruses of other microalgae species).

#### **CONCLUSIONS**

The change in the infectious titer of strains of Black Sea algal viruses of three species of microalgae has been experimentally studied (*Tetraselmis viridis* strain TvV-S21-1, *Dunaliella viridis*—strain DvV-S20-1 and *Phaeodactylum tricornutum*—strain PtV-S20-1) after incubation in a constant unidirectional magnetic field with a magnetic induction of 60 mT.

During the experiments, the following patterns were established:

(1) After 24–48 h of magnetic load the infectious titer decreases by one order of magnitude in the studied Black Sea algal viruses, (for DvV-S20-1 and TvV-S21-1), or does not change (for PtV-S20-1);

(2) Longer exposure to a magnetic field (72 h or more) leads to a decrease in infectious titer by two orders of magnitude (for DvV-S20-1 and PtV-S20-1);

(3) The invariability (stability) of the infectious titer of algal viruses was revealed after 72 h of magnetic load, even with a further increase in duration (up to 6 to 8 days).

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### COMPLIANCE WITH ETHICAL STANDARDS

*Conflict of interest.* The authors declare that they have no conflicts of interest.

*Statement on the welfare of humans or animals.* This article does not contain any studies involving animals performed by any of the authors.

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