ISSN 1019-3316, Herald of the Russian Academy of Sciences, 2019, Vol. 89, No. 2, pp. 171–178. © Pleiades Publishing, Ltd., 2019.
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Scientific Session of the General Meeting of the Russian Academy of Sciences

Could Oncolytic Viruses Provide a Breakthrough in Oncology?

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Received December 3, 2018; revised February 3, 2019; accepted February 14, 2019

Abstract—Despite long-standing and large-scale studies on the nature of cancer and the development of numerous antitumor drugs, the incidence of cancer is growing and the five-year survival rate of cancer patients diagnosed at the advanced stages of the disease remains unacceptably low. The author considers the causes of such failures, which lie in the very nature of malignant cells: they can adapt to and resist practically any systemic therapeutic intervention. In this context, considerable hopes are pinned on oncolytic viruses, which are medical agents of a new type, able to produce an integrated effect on the disease. In addition to their direct ability to kill tumor cells selectively, oncolytic viruses stimulate natural processes of immune surveillance and removal of cancer cells. Besides, oncolytic viruses can kill tumor-initiating cancer stem cells that are highly resistant to chemo- and radiotherapy, and overcome the immunosuppression of the tumor microenvironment. These features make oncolytic viruses unique anticancer agents that combat cancer cells by multiple natural mechanisms. To incorporate viral cancer therapy into mainstream medical practice, it is necessary to intensify basic research on viral oncolysis mechanisms, to develop new therapeutic viral strains and tests for their personalized selection, and to improve methods of the local and systemic delivery of oncolytic viruses to tumor locations. Trials of drugs that would accelerate the introduction of new viral strains into medical practice will also require cardinal changes. Achievements in this sphere will help to overcome many old problems in the therapy of metastatic forms of malignant diseases.

Keywords: oncolytic viruses, viral oncolysis, antitumor therapy, cancer immunotherapy, immune checkpoint inhibitors, tumor progression, resistance to therapy, tumor stem cells, cancer recurrences, personalized therapy.

DOI: 10.1134/S1019331619020023

Problems of systemic therapy of malignant diseases. The brilliant advances of medicine in the fight against infectious diseases in the mid-20th century highlighted the problem of fighting cancer. The most effective approach to its therapy has always been radical surgical tumor removal, which allows to attain recovery in case of early diagnosis. However, if the malignant process has spread in the form of metastases, surgery becomes palliative, while an effective therapy requires specific systemic measures.

Over the past 70 years, unprecedented intellectual and material resources have been spent on finding effective methods of cancer treatment. Cancer problems have become the main locomotive and the main goal of financial investment in basic research in biology and medicine. During this period, the basics of the functioning of living systems were studied at the molecular, cellular, and organismic levels. As characteristic features of cancer cells and their fundamental differences from normal ones were revealed, numerous approaches of systemic therapy were developed, both broad spectrum and acting on the unique targets of individual forms of malignant diseases (targeted therapy). The treatments include cytotoxic chemotherapeutic agents, radiation therapy, hormones and their inhibitors, specific chemical inhibitors of metabolic processes, and monoclonal antibodies.

Significant success was achieved in developing neoplastic process imaging techniques and determining characteristic genetic molecular autographs of tumor cells, allowing for a fine personalized assessment of individual cases, dividing them into subgroups that have differences in prognosis and response to therapy. All this laid the foundation of the individual approach to treatment, making systemic therapy more effective.

Unfortunately, despite all these achievements, over the last 30 years, the main indicator that characterizes

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the effectiveness of therapy, the five-year survival rate, has increased by only 2–7% [1] depending on the type of cancer. Morbidity, as well as mortality, continues to grow even in developed countries with state-of-the-art medical infrastructures. The five-year survival rate is especially alarming among patients in whom cancer was detected at the stage of metastatic spread. For example, in the United States, lung cancer is first detected at this stage in 57% of cases, five-year survival remaining at 4.3% [2].

Evidently, the question how to treat metastatic cancers remains open. The existing schemes of their systemic therapy with the targeted elimination of tumor cells proceeding from their individual specific properties, including the use of target drugs, have only a short-term effect; in essence, they are palliative. If cancer goes beyond a primary organ, it predominantly remains incurable.

Toward naturelike approaches to the treatment of malignant diseases. Modern oncology is at the threshold of qualitative changes in the strategy of therapy. The recently obtained wealth of knowledge about the biology of normal and cancer cells, systems of control over the genetic stability of cells in a multicellular organism, and mechanisms of carcinogenesis and tumor progression shows that it is useless to create new ingenious and expensive target drugs. They will inevitably fail to surpass in effectiveness the existing ones. Even when causing remissions of various lengths, these drugs make no substantial contribution to the five-year survival index, let alone complete recovery.

We must reconsider the negative experience of systemic cancer therapy, reject illusions of the past, and develop new trends that would create hope for a qualitative breakthrough. The failures of therapy based on the killing of tumor cells with account for their characteristic, sometimes unique, properties follow from the nature of the tumor cell with its colossal variability. Instead of searching for new target drugs, we should turn to the use of antitumor defense mechanisms created by nature itself. These mechanisms defend us throughout life, and only their weakening creates conditions for the accumulation of the critical mass of transformed cells that trigger tumor progression processes.

Oncological diseases are not programmed by nature; they are a result of failures, violations of the natural mechanisms that ensure the preservation of health throughout life. Hence, studies on the natural mechanisms of antitumor defense can give a clue to the creation of effective means of struggle against cancer. Observable of late in oncology has been a gradual turn to biotherapy. The treatment of the future should be aimed at the recovery and strengthening of natural mechanisms of the recognition and elimination of pathological cells, excluding the possibility of their accumulation and tumor formation.

Reasons for the low effectiveness of systemic therapy of malignant diseases. As opposed to normal cells, the cancer cell ceases to be a part of the organism; it ceases to obey the signals that determine its role, place, functions, and adequate behavior. This occurs as a result of genetic or epigenetic damages in the intracellular control system, which assesses the behavior of each cell and makes decisions on its future fate. The central component of this system is tumor suppressor p53 [3], the function of which is to integrate the signals that come from numerous processes inside and outside the cell with the subsequent launch of pivotal decisions, which either favor the adaptation of the cell to the changing conditions or trigger irreversible processes of cellular suicide [4].

The system of the intracellular control of genetic stability reliably protects cells against genetic and epigenetic changes, guarantees defense against malignant transformations. However, if the mutagenic process affects the components of the p53-dependent control system itself, a catastrophe takes place, leading to the emergence of cells that embark on the path of autonomy and unlimited evolution inside the organism. In essence, such a cell becomes an independent parasitic unicellular organism, which competes with normal cells and other cells of the tumor. Acquiring additional mutations, the tumor cell seeks expansion and develops techniques to maintain a sufficient blood supply and overcome tissue seals and actions of the immune system. As a consequence, numerous genetically distinct subpopulations of cells form within the tumor, which can respond to the applied therapy differently; under its action, there occurs permanent selection of the most stable subpopulations.

Another reason for resistance to systemic therapy is that the tumor contains a subpopulation of cells in a special physiological state. They are usually identified as cancer stem cells [5], since they have a number of markers of stem cells and can divide asymmetrically, when one daughter cell preserves stem properties, while the other can perform the typical symmetrical divisions and does not differ from the cells of the bulk of the tumor. The stem cells are characterized by metabolic properties, are extremely stable to radiation and cytotoxic therapy, and lack characteristic targets that can be affected by target drugs. Such cells can be characterized as spores able to ensure the survival of a unicellular organism under unfriendly conditions. After a successful course of chemotherapy, when the tumor largely dies, some stem cells remain viable, which is later manifested as a recurrence.

Summing up the disappointing result of the description of the existing state of cancer systemic therapy, we can state that tumor cells originating in a patient's organism inherit a practically unlimited arsenal of mechanisms, which they can combine for adaptation to preserve viability and expansion under any conditions and effects.

Natural mechanisms of antitumor defense. An organism has three levels of mechanisms that ensure reliable defense against a malignant pathology. At the cellular level, this is the above-mentioned system controlled by tumor suppressor p53, which effectively stops or kills the emerging pathological cells. In addition to the p53 system, there exist some other intracellular mechanisms that restrict the possibility of an autonomous existence of changed cells.

The next level of defense relates to the function of innate (not adaptive) mechanisms of the immune system. Their mission is immune monitoring of the appearance of pathological cells. The immune cells with NK cells as their main component can recognize damaged and changed cells by characteristic molecular autographs and kill them at the place of their detection in real time.

If changed (tumor) cells accumulate, the third level of defense gets involved, the adaptive immune system, which recognizes neoantigens of tumor cells, emerging as a result of genetic changes or violations of the regulation of gene expression. T cells form an immune response to such antigens, manifested in the attack of cytotoxic T lymphocytes. The arsenal of immune mechanisms also includes some auxiliary factors, represented by numerous cytokines, which coordinate the functioning of immune cells and tissue reactions and the manifestation of cytotoxicity.

Role of the tumor microenvironment in advance of the neoplastic process. A malignant tumor forms as a result of the weakening and failures of defense mechanisms, which may be associated with genetic factors, toxic effects, age-related changes, and chronic pathologies. The emergent tumor begins to manifest additional mechanisms that favor its development. Tumor cells begin to form a friendly microenvironment, which provides for the inflow of nutrients and oxygen and defense against attacks of the immune system [6]. The deficit of oxygen forces tumor cells to secrete angiogenic factors, which favor the formation of a network of blood vessels. Chemokines secreted by tumor cells attract immature myelocytes (myeloid-derived suppressor cells), which heavily infiltrate the tumor, stimulate cell division, and protect the tumor against attacks of the immune system.

The immunosuppressive microenvironment is formed by tumor cells, among other things, at the expense of their ability to express on their surface so-called proteins of immune checkpoints [7], which are part of the physiological mechanism of weakening an immune attack by cytotoxic T lymphocytes at sites of chronic inflammation. Thus, tumor cells begin to express surface proteins, characteristic of antigen-presenting cells and able to decrease the activity of T cells. The proteins of the B7 family (CD80 and CD86), expressed on the surface of a tumor cell, interact with the CTLA-4 receptor of CB8+ T lymphocytes and the PD-L1 protein, with the PD-1 receptor; as a result, activation signals are blocked and the programmed death of the lymphocyte is triggered. Owing to tumor cell–induced immune tolerance, the tumor becomes able to develop and evolve, fixing its presence in the organism despite the counteraction of the immune system.

The discovery of the mechanism of defending tumor cells against an immune attack, awarded by the Nobel Prize of 2018, appears to be a landmark that makes it possible to increase substantially the effectiveness of cancer biotherapy through reactivating natural mechanisms of immune defense. At present, a number of therapeutic monoclonal antibodies have been developed that block the ability of T cells to switch themselves off under the action of immunosuppressive signals of the tumor microenvironment [8]. As opposed to most antitumor drugs, these preparations are not toxic for tumor cells themselves, but they create conditions for their effective removal at the expense of natural reactions of the immune system.

The appearance of checkpoint inhibitors can be considered as a milestone on the way toward effective therapy, an important component of future therapeutic schemes. The synergism of the therapeutic effect is expected, for example, under the inclusion of checkpoint inhibitors in schemes that use antitumor vaccines based on immune cells and adaptive immunotherapy with chimeric antigenic receptors (CAR) [9]. Even now, checkpoint inhibitors sometimes demonstrate impressive results both under monotherapy and in combination with traditional chemotherapeutic and antigenic drugs.

Viruses as potential antitumor agents. The ability of viruses to kill tumor cells has been known since the beginning of the 20th century. In 1904, an observation about leukemia remission after influenza was published [10]. Later several other reports appeared dealing with the relation between viral diseases and the amelioration of oncological patients. In the 1920s, it became clear that tumor cells can be used for the reproduction and development of viruses. The first attempts to use the ability of viruses to kill cancer cells took place soon after the end of WWII. All the viruses noted at that time were disease-causative agents; hence, the use of pathogens for cancer therapy was accompanied by serious complications and for many years formed a negative attitude of doctors and society to the very idea of viral therapy [11]. Interest in viral oncolysis began to revive after the appearance of vaccinal attenuated viral strains, the establishment of the antitumor activity of animal viruses, and the discovery of naturally nonpathogenic human viruses. In the Soviet Union, such studies used nonpathogenic human enteroviruses isolated from the intestinal tract of healthy children and developed as living enteroviral vaccines for the nonspecific prevention of seasonal viral infections. The trials of these viruses demonstrated cases of long remissions in some patients [12].

However, the limited knowledge about the fundamental mechanisms of viral oncolysis and the unpredictability of the therapeutic effect of viruses led to a pause in studies on this trend.

Systematic and large-scale studies on viral oncolysis at the present stage have been resumed on the basis of recent knowledge about the nature of viruses, the causes of their pathogenicity, and fundamental differences between normal and tumor cells. Artificial modifications of the virus genome have become possible, leading to a loss of pathogenicity and the acquiring of selectivity relative to tumor cells. Since the early 1990s, several dozen oncolytic viruses have been created and tested, and the main mechanisms of their therapeutic action have been established [13]. At present, three preparations on the basis of oncolytic viruses have already been approved for clinical use in various countries, and several others are at closing stages of clinical trials [14]. In some cases, clinical tests demonstrate a breathtaking effectiveness, leading to long-term remission and even the full recovery of patients with absolutely fatal diseases, for example, glioblastoma multiforme [15, 16]. All this testifies to the unquestionable good prospects for viral cancer treatment. However, the use of viruses in clinical practice is still associated with numerous problems, warranting further investigation.

Viruses as antitumor agents of a fundamentally new type. Viruses as living organisms, and, hence, their use in treatment, is fundamentally different from the use of all other medicaments. Since viruses can replicate in a patient's organism, the notion of the maximum tolerable dose for them is extremely conditional. An optimal dose must ensure the introduction of an oncolytic virus into tumor cells, its overdosing being less dangerous than that of other medications. Upon finding itself in the cell, the virus replicates, and its dose can increase substantially, ensuring the desired action.

The virus has a complex and combined effect on the patient's organism. It was assumed earlier that the therapeutic effect is predominantly based on the ability of the virus to replicate selectively in tumor cells and kill them. However, subsequent studies have shown a no less important role of viral infection in the stimulation of antitumor immunity processes and the action on the microenvironment inside the tumor, which removes immune suppression. To understand better the mechanisms of the therapeutic effect of oncolytic viruses, let us consider them individually.

Mechanisms of viral oncolysis. In the process of tumor progression, tumor cells undergo evolution toward an increase in autonomy, the removal of control on the part of the organism, the acceleration of divisions, and acquiring the ability to spread. Although this favors the development of the disease, at the same time, many of the changes lead to a higher susceptibility to viruses; tumor cells become more accessible to penetration [17]. Many tissue seals, which under normal conditions prevent the spread of viruses, become destroyed. Thus, in a quickly growing tumor, the blood-supply vasculature develops rapidly. As a result, the newly formed vessels are leaky [18] due to numerous holes through which viruses can penetrate the tumor from the bloodstream. The structure of the tumor is chaotic; some tumor cells lose contacts with each other, which is characteristic of normal tissues. As a result, the surface of cells is unprotected from contact with the virus. Using surface transmembrane receptors, viruses can easily penetrate a cell, but this does not guarantee their effective replication.

Interaction between the virus and the cell is a confrontation of two organisms, one of which (the virus) tries to seize control over the biosynthetic apparatus of the cell and use it for the needs of its own replication, while the other (the cell) uses defense mechanisms against viral pathogens. A normal cell has a reliable system of recognizing alien viral components, in response to which the cell begins to secrete type I interferons. These antitumor signaling proteins bind with specialized receptors on the surface of cells, activating a signaling cascade, which leads to the acquisition of insusceptibility to viral infection. In addition to specific antiviral action, the interferon induces a delay in cell division and a slowdown in metabolism, restricting the possibilities of the synthesis of viral proteins and nucleic acids; hence, along with defense against viruses, interferon mechanisms impose potential limitations on the expansion of tumor cells. For this reason, under tumor progression, there occurs the selection of cells in which the interferon mechanisms are suppressed as a result of mutations or epigenetic disorders [19], which predetermines the selectivity of viruses belonging to different families to replication in tumor cells. As a result, it becomes possible to maintain several cycles of reinfecting tumor cells with viral particles formed after the primary infection of a small number of tumor cells with the preparation introduced. Owing to this process, some tumor cells die on a scale that depends on the speed of virus replication; its ability to penetrate hard-to-reach regions of the tumor; and the time of the development of antiviral immunity, which neutralizes viral particles.

However, the direct cytolytic action of viruses on tumor cells is only a part of the viral oncolysis mechanism. The local viral infection developing in the tumor stimulates the innate and adaptive mechanisms of antitumor immunity. While the virus replicates in the tumor predominantly in tumor cells, the cells of the tumor stroma (blood vessels, connective tissue) are continuously affected by the virus; as a result, they actively secrete interferons and other cytokines, which attract cellular components of the innate immune system (NK cells, macrophages, and monocytes) to the tumor, as well as components of the adaptive immune system (antigen-presenting dendritic cells and T lymphocytes). The death of tumor cells under the direct action of viruses, as well as NK cells, leads to the

release of tumor neoantigens and a number of intracellular proteins and metabolites serving as adjuvants for accepting and recognizing tumor antigens by antigen-presenting cells. As a result, there forms a specific T-cellular immune response to the tumor's neoantigens, while cytotoxic T lymphocytes, able to recognize tumor cells, rush to kill them to tumor lesions even after the virus has ended its action because of neutralization by antiviral antibodies [20]. This antitumor action can be long term.

Infection of tumor cells with viruses also favors the removal of the immunosuppressive effect of the tumor microenvironment, which limits the ability of the immune system to kill pathological cells as soon as they are detected. Although many details of this process remain understudied, it has been revealed that, owing to the secretion of cytokines, there are changes in the character of infiltration by components of immune cells in the infected tumor microenvironment, including a substantial decrease in infiltration by suppressor myeloid-derived cells [21]. These processes are somewhat paradoxical. For example, under the action of an interferon, tumor cells can increase the level of the expression of the PD-L1 protein, which is accompanied by an increase in the infiltration of the tumor by CD8-positive T cells. Against this backdrop, the use of antibody blockers of immune checkpoints—Pembrolizumab (Keytruda) and Nivolumab (Opdivo)—is accompanied by the massive activation of cytotoxic T lymphocytes and the elimination of tumor cells. This mechanism underlies the observed synergism of the combined action of oncolytic viruses and immune checkpoint inhibitors, especially in the case of PD-L1-negative tumors, characterized by a low level of infiltration by T cells [22, 23]. Thus, owing to the complex action of viral infection, natural mechanisms of immune surveillance and timely removal of defective and tumor cells become restored, which helps overcome protective techniques of tumor cells and shift the balance of the tumor process from the accumulation of tumor cells to their elimination.

An important property of oncolytic viruses is their ability to kill tumor-initiating stem cells, which are extremely tolerable to therapeutic interventions and are the main source of relapses after massive chemotherapy, radiation therapy, and targeted therapy [5]. The existing data testify to the ability of oncolytic viruses of different virus families to kill tumor stem cells effectively [24]. This is demonstrated in the therapy of glioblastoma multiforme, relapses of which are inevitable due to the ability of glioblastoma stem cells to travel significant distances along axons from the location of the primary tumor [25]. The use of oncolytic viruses is the only way to prevent such relapses [15, 26].

What impedes the prompt introduction of oncolytic viruses into clinical practice? At present, many laboratories in the world actively study viral oncolysis and create new strains of oncolytic viruses based on several virus families. To create safe and effective strains, they introduce into the virus genome certain modifications that suppress the ability of viruses to overcome systems of the cell's antiviral defense and activate its metabolism to ensure the viruses' replication. Such strains are characterized by a high oncoselectivity, since tumor cells are, as a rule, devoid of antiviral defense and their metabolism is high to ensure continuous divisions. Additional genes are introduced into many strains of oncolytic viruses, strengthening the antitumor effect. They include, for example, some cytokines to stimulate antitumor immunity, enzymes favoring a better spread of viruses inside the tumor, proteins with oncotoxic properties, and microRNAs affecting the physiology of the tumor cell. In addition to genetically modified viruses, there are trials of animal viruses, safe for people but oncolytically active against human tumor cells, as well as those of some vaccinal viral strains, weakly or low pathogenic human viruses. However, because of the difficulty and expensiveness of preclinical and clinical trials, biotechnological companies promoting viral cancer therapy, as a rule, focus on a single therapeutic strain.

At present, only three viral strains have been allowed for clinical use. These are the recombinant adenovirus Oncorine (H101), obtained in China; the naturally nonpathogenic human enterovirus Rigvir, obtained in Latvia; and the recombinant herpes virus T-Vec (Imlygic or Talimogene Laherprepvec), obtained in the United States. The latter was permitted for use in 2015 for the therapy of metastatic types of malignant melanomas. Several viral strains are at the final stages of clinical trials: the recombinant poxvirus Pexa-Vec, the unmodified reovirus Reolysin, the recombinant poliovirus PVSRIPO, the unmodified coxsackievirus A21 CAVATAK, and some others [27]. Each of these viruses can in some cases demonstrate impressive therapeutic effects. For example, the recombinant poliovirus is effective in 20% of patients; it causes long-term remission with no relapses for at least one year [16]. On the one hand, this is an outstanding achievement because glioblastoma is an absolutely fatal disease. On the other hand, 80% of the patients do not respond to treatment by this viral strain, and such a picture is characteristic of all known oncolytic viruses. However, this does not mean that viral therapy has no prospects for patients who do not respond to a definite strain because another viral preparation may be effective for them.

The absence of a response to a viral preparation can largely be explained by the fact that the individual susceptibility of tumor cells to individual viral strains can vary significantly. Hence, to increase the efficacy of therapy, it is necessary to develop technologies of testing the susceptibility of tumor cells to a panel of several strains of oncolytic viruses. This can be accomplished either by the direct determination of the ability of

viruses to infect tumor cells obtained from a removed tumor or as a result of biopsy or by searching for and analyzing predictive biomarkers. The establishment of such biomarkers will make it possible to select a suitable personalized set of therapeutic strains using a relatively quick and simple test.

The use of a single viral strain has another drawback. In response to viral infection, the patient's organism develops antiviral immunity, which gradually neutralizes the virus and weakens its therapeutic effect. Although a virus can trigger the process of immune recognition and the elimination of tumor cells, the immune component alone may be insufficient under significant tumor loads. To prevent a recurrence, it would be preferable to use immunologically unrelated strains of oncolytic viruses in a series of courses.

Another serious obstacle on the way toward successful viral therapy is the insufficient efficacy of the delivery of an oncolytic virus to tumor lesions. Only few tumors are accessible for a direct injection of a virus; the tumor process spreads, and it is necessary to introduce the preparation on a systemic basis. An intravenous or intramuscular administration quickly activates the virus because it is quickly absorbed by endothelium cells and binds with specific factors of defense against pathogens. For effective delivery to a tumor, multiple injections of excessive doses of viruses are necessary, and even this is sometimes insufficient. To overcome this problem, the "Trojan horse" approach was proposed [28], when the virus is administered systemically using a carrier—virus-susceptible cells infected in advance in vitro. Upon entering the bloodstream, they migrate to the tumor, where the virus is released. At present, a number of cell types are being tested as carriers of oncolytic viruses, the most attractive being the use of the patient's own immune cells. Our laboratory has established that dendritic cells can be infected and can replicate on a limited basis various strains of oncolytic viruses. Intravenous administration of such cells to mice delivers viruses to the tumor much more effectively compared to the administration of high doses of a viral preparation. This makes dendritic cells a promising universal vector for the delivery of viruses to tumors.

Panel approach to viral therapy of malignant diseases. To overcome problems of the individual insusceptibility of tumors to viral therapy, we proposed the panel approach, which implies that the therapeutic arsenal consists of several strains of oncolytic viruses with different specificity relative to tumor cells. The panel formed predominantly consists of representatives of nonpathogenic human enteroviruses isolated from the intestinal tract of healthy children, which were tested as live enteroviral vaccines in the 1970s [29]. In addition, the panel includes vaccinal type I–III poliovirus strains, nonpathogenic for people animal paramyxoviruses (Sendai virus and Newcastle disease virus, which is an avian virus, attenuated and used in agriculture to vaccinate commercial poultry), and three strains of nonpathogenic orthoreoviruses. The representatives of the panel have different host tropisms in relation to individual human tumors, because they use different receptors to penetrate the cell, as well as specific elements of the virus genome, responsible for a range of action on various types of cells. The strains of the panel are devoid of pathogenicity but preserve the ability to damage tumor cells and stimulate natural antitumor mechanisms.

It is possible to select a set of strains suitable for a patient in the course of a direct trial of the ability of each virus in the panel to infect cells of the tumor in a culture. Such a test is possible in sampling viable cells from the tumor through biopsy or under surgery. Enzyme-treated tumor cells are placed in the broth and are infected with viruses, after which one observes the cytopathic effect of the virus and titers of newly formed viral particles. The strains able to replicate on tumor cells form a personalized panel, which is subsequently used for therapy through successive administration at intervals of one to three weeks. If tumor cells are inaccessible for analysis, treatment is also possible through successive administration of preparations of oncolytic viruses from the existing arsenal of strains, hoping that some of the preparations will be effective.

The studies of our laboratory are also aimed at the development of fast tests accessible for most patients that predict the susceptibility of their tumors to a therapeutic viral strain. To this end, it is necessary to identify biomarkers the level of expression of which in tumor cells can correlate with the ability to support the replication of certain viral strains. We seek such biomarkers by analyzing the data of sequencing the transcriptome and proteome of tumor cells and subsequently establishing correlations with the susceptibility of viruses to various viral strains. At the current stage of research, such tests reveal the dependence of the ability of cells to maintain the replication of viruses on the level of the expression of genes of the signaling pathways of the interferon response [30]. However, among predictive biomarker candidates, there are several products of genes responsible for processes necessary for the penetration of viruses into the cell, their uncoating, and their interaction with biosynthetic and energy processes of the cell [31].

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The treatment of oncological diseases using oncolytic viruses is fundamentally different from the existing treatment practices. Most medications rely on the use of small doses of toxic substances (poisons) that remove violations in the balance of processes. Such preparations can be effective relative to static, unchangeable targets. In oncology, tumor cells evolve continuously, while toxic action leads to the selection of cells devoid of therapeutic targets. Hence, the term

of the therapeutic effect of the existing and, most likely, future target preparations aimed at the direct elimination of tumor cells is quite short. As for viral therapy, it consists of two components. On the one hand, viruses also exert a direct toxic effect on cells using specific targets. Tolerance to the action of viruses can be formed just like that to typical therapeutic means. However, viruses also trigger the complex process of immune interactions; as a result, tumor cells lose the ability to escape elimination through natural mechanisms of antitumor defense. Owing to their complex action, viruses can be seen as agents affecting not a particular manifestation of a disease but its cause, which lies in the weakening of natural mechanisms of antitumor defense.

Despite the good prospects for viral therapy, many tasks still await solution in order to allow this approach to cure malignant diseases predictably. Of special interest is the combination of viral therapy with modern immunotherapy using immune checkpoint inhibitors. Our panel approach with its broad arsenal of strains of oncolytic viruses, able in aggregate to affect most tumor cells, allows us to hope that the efficacy of viral therapy will soon increase substantially. However, there are still many questions that require investigation to increase the effectiveness of viral delivery, to predict on a personalized basis the effect of individual viral strains, and to increase their therapeutic activity by giving additional functions to the virus genome. To answer these questions, it is necessary to activate basic research on mechanisms of viral oncolysis and possibilities of its control. The vectors of this search must follow the priorities dictated by the necessity to apply quickly in practice the technologies under creation. Of great importance is also the possibility to restructure substantially the practices of controlling the creation of new viral drugs, rules of clinical trials, and the removal of excessive barriers for the quick introduction of new technologies into medical practice. The actualization of these vectors can favor a breakthrough in cancer therapy in the decades to come.

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Translated by B. Alekseev