Stress-induced molecules MICA as potential target for radioimmunotherapy of cancer

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Abstract. Improving the treatment of cancer, increasing their effectiveness and safety is the main objective in the medicine. Molecular nuclear medicine plays an important role in the therapy of cancer. Radioimmunotherapy (RIT) involves the use of antibodies conjugated with therapeutic radionuclides. More often for RIT use the radiolabeled monoclonal antibodies against tumor-associated antigens. Encouraging results have been achieved with this technology in the management of hematologic malignancies. On the contrary, solid tumors have been less responsive. Despite these encouraging results, new potential target for radioimmunodetection and RIT should be found. It was found to increase the level of tumor-associated molecules MICA in the serum of cancer patients. Use of anti-MICA monoclonal antibodies capable a specifically attach to cancer cell via NKG2D ligands and destroy it, is a very promising direction, both therapeutic and diagnostic standpoint.

1. Introduction
One of the most urgent problems of modern oncology is early diagnosis and more effective treatment of patients with malignant tumors. Currently, methods of radionuclide therapy are developing rapidly around the world, which has become an effective tool, both independently and combined treatment. Radioimmunotherapy (RIT) - a promising trend that combines advances in modern nuclear medicine, immunology and biotechnology. Molecular nuclear medicine plays an important role in the diagnosis and therapy of cancer and infectious diseases. It is based on targeted therapy when the therapeutic monoclonal antibody (mAb) coupled to radionuclides. Antibodies bind specifically to antigenic determinants on the tumor or other cells and destroys these cells radionuclide by topical exposure [1].

Receptor NKG2D is expressed by all NK cells but is not limited to NK cells, as it is also expressed by many T cells, including all CD8⁺ T cells in humans, subsets of γδ T cells, and subsets of NKT cells. In humans, the NKG2D ligands include MICA and MICB (MHC class I chain–related proteins A and B). These ligands are typically induced in response to stress, commonly found within cancers. However, some cancers are known to develop escape mechanisms that reduce the levels of these
ligands on their surface, including down regulation of expression or internalization of the ligands; or shedding of their soluble extracellular domains (sMICA, sMICB). Recently, we demonstrated that MICA is released as a soluble form from the cell surface of tumor cells and can be detected at high levels in sera of patients with malignancies but not in healthy donors [2].

A combination of measurements of circulating MICA/B domains and tumor volume can be combined to determine the rate of shedding, and that this data, could be used to determine likely response to RIT or cell therapy. Immunizing animals with recombinant human MICA protein can be obtained anti-MICA antibodies that will serve as a basis for joining radionuclide and subsequent RIT. The developed method of radioimmunotherapy can improve the treatment of cancer patients, as well as help to reduce the risk of metastasis and recurrence of tumor growth [1].

Purpose to this work was to justification find surface molecules of tumor cells as targets for radioimmunotherapy.

2. Material and methods

In the study were involved 80 patients with a variety cancer: melanoma (n=44), prostate cancer (n=16), colon cancer (n=20) and healthy volunteers (n=20). For the collection of biological material used was taken a peripheral blood samples. Before treatment, all patients performed an enzyme immunoassay for the presence of serum label of soluble forms of molecules MICA. A concentration of sMICA protein in sera of patients were determined by ELISA and reagent MICA Human ELISA Kit according to manufacturer's instructions (Abcam, USA).

3. Result and discussion

In Russia the first studies using radionuclide-labeled anti-tumor antibodies for the diagnosis and therapy of tumors were performed by Anokhin Yu N in 1985 year. The results of experimental studies on animals with transplants of solid tumors have been encouraging and have shown high specificity of $^{131}$I-labeled anti-tumor antibodies to target antigens in comparison with non-specific immunoglobulins, labeled with $^{125}$I. The concentration of the antitumor antibodies in tumor tissue is 7-10 times greater than the content of these antibodies in normal tissues [3, 4].

In this study we revealed the presence of soluble forms of highly polymorphic stress-induced MICA molecules in a number of cancers. The significant increase of level sMICA in the amount of 4.6 times in serum in patients with melanoma, colon cancer and prostate were found compared with the control group (figure 1).

These molecules, related to MHC class I antigens, have been described in the late '90s by American scientists [5]. It is shown that in neoplasia expression of these molecules is one of the earliest events that are manifested in the process of malignant transformation of cells. On the surface of normal cells they are absent or are contained in the insignificant amounts, but their expression can be induced under conditions of cellular stress, viral or bacterial infections and in tumor growth.

The adapter molecule DAP10 accept impulse occurring upon binding of NKG2D surface receptor on lymphocytes with stress-induced molecules on the surface of target cells, and carry out further transfer a signal that induces lymphocyte cytotoxic effect. Thus, expression of stress-induced molecules should lead to immunological destruction of the infected or transformed cells by the virus.

Therapy approaches are tailored to enhance or redirect NK cell— and/or T cell—mediated cytotoxicity by utilizing agents that may increase NKG2D ligand expression or bifunctional fusion proteins including an NKG2D ligand and a single-chain Ab fragment (scFv) targeting a specific tumor cell surface marker. Among pharmaceuticals tested for upregulation of NKG2D ligands on tumor lines in vitro are demethylating agents, proteasomal inhibitors, and genotoxic drugs used for chemotherapy such as histone deacetylase inhibitors [6]. Objectives for the bifunctional fusion proteins are to target NK cells and T cells onto tumor cells and engage NKG2D.
Another type of bispecific T cell and NK cell engager, thus far tested in mice only, combines the extracellular domain of NKG2D with either CD3ε or the Fc portion of IgG2a for binding to CD16. In an in vivo tumor model, administration of the NKG2D–CD3ε fusion protein reduced growth of tumors bearing NKG2D ligands and promoted T cell infiltration. Likewise, the NKG2D–Fc region fusion protein triggered lysis and reduced growth of ligand-positive lymphomas [7, 8].

This study confirms our earlier results and gives reason to conclude that in the serum of cancer patients with solid tumors multiply exceeded the levels of soluble forms of the NKG2D ligands [9]. This claim suggests that stress-induced molecules MICA may serve as markers of cancer.

4. Conclusion
We can assume that NKG2D ligands the antigenic determinants MICA or MICB will serve as an attractive target for radioimmunotherapy [10]. Conjugation of anti-MICA mAbs to these stress-induced molecules with a specific radioisotope may be broadly used in the treatment of many tumor types. Radioimmunotherapy can be used in chemoresistant patients and patients are unaffected to the action unlabeled antibody. Also RIT can be effective for elimination of multiple-drug-resistant tumor cells. Main goal of such drugs is associated with the purposive destruction certain cells, such as tumor. NKG2D ligands may present a useful target for immunotherapeutic approaches in cancer. A monoclonal antibodies NKG2D with radionuclide capable of specifically accede to the cancer cell and destroy it. Broad applicability of these therapies to many types of tumors offer great promise for the both treatment and diagnostic of cancer.

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