



Binase Do Not Change Level of Intracellular RNA in Breast Cancer Cells

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Abstract We analyzed changes of intracellular RNA level in breast cancer cells using BT-20, BT-474, and MCF-7 cell lines. It is the continuation of our former studying the effect of bacterial ribonucleases on physiological and biochemical parameters of breast cancer cells. For the first time, we tested ability of *Bacillus pumilus* RNase (binase) to effect the level of intracellular RNA in these cells. Differences in total RNA level under of the binase action on the cells of triple negative, triple positive, and double positive breast cancer during 48 h of treatment were not detected. The sensitivity of cells to binase was not associated with a reduction of the intracellular RNA level.

Keywords Breast cancer · BT-20 · BT-474 · MCF-7 · Binase · Intracellular RNA

1 Introduction

Breast cancer is a socially significant cancer in females worldwide. The possibilities of breast cancer treatment are primarily determined by the biological characteristics of the tumor. Breast cancer has several sub-types: one of these sub-types is known as triple negative breast cancer (TNBC). TNBC does not grow in response to the hormones estrogen, progesterone, or HER2/neu [1]. Therefore, TNBC does not respond to hormonal therapies that target the receptors of these hormones. For this type of breast cancer, targeted treatments are not

available as for other sub-types of breast cancer. Thus, patients with hormone-sensitive tumors and with triple negative tumors differ in their susceptibility to therapy. Approximately 10–20% of breast cancers are triple negative [2].

Cytotoxic ribonucleases of various organisms were known to be used as potential antitumor agents. The commercial preparation of RNase from the frog *Rana pipiens* oocytes (onconase) is now at the third stage of clinical trials as a drug against lung mesothelioma [3]. Antitumor activity was found to be characteristic of RNase from bovine testes (BS-RNase), RNases of human eosinophilic granules, and of a number of RNases of lower organisms, including fungi, actinomycetes, and bacteria [4]. Binase is an enzyme that has been isolated from a bacterium *Bacillus pumilus*. We have shown previously that it has selective antitumor properties for different types of malignant tumors [5, 6], include breast cancer tumors [7]. The aspects of enzyme action toward cells of the immune system [8] and non-malignant cells [9] have also been described.

Here, we demonstrate for the first time that sensitivity of breast cancer cells to binase, include triple negative, does not depend on cellular RNA catalytic degradation.

2 Materials and Methods

Bacterial guanyl-specific RNase from soil bacterium *Bacillus pumilus*, binase (12.2 kDa, 109 amino acid residues, pI 9.5), was homogenously isolated from culture liquid of *Escherichia coli* BL21 carrying the pGEMGX1/ent/Bi plasmid, according to Schulga et al. [10].

Cell cultures of breast cancer cell lines BT-20 (triple negative), BT-474 (triple positive), and MCF-7 (double positive) were obtained from all-Russian cell culture collection. Cells were cultivated in RPMI supplemented with penicillin (100 U/

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