Research of Antiblastic Effects of Melatonin by Using ATP-TCA Method on Prostate and Breast Cancers Cell Culture, with Artificial Activation of Melatonin Receptor Genes

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Abstract

Using exclusive technology we have developed biologically (animal) derived apyrogenic (assessed by LAL-Test system) "cocktail" that increased by 41% expression of Melatonin receptor MT1A in cellular culture of prostate cancer (after the 10 day incubation) and by 57.5% in case of breast cancer (after the 10 day incubation). Exposure of adequately marked so called activated cancer cell cultures to Melatonin and assessment of its anti-cancer activity using ATP-TCA method demonstrated the following results: In experiment Melatonin in a dose of 0.4 mg/kg suppresses metabolic activity of prostate adenocarcinoma cellular culture by 51.4% and mammary ductal adenocarcinoma cellular culture – by 67.9% compared to standard control. Melatonin in a dose of 0.8 mg/kg suppresses metabolic activity of prostate cancer cells by 56.7% and mammary ductal adenocarcinoma cells – by 71.1% compared to standard data.

Introduction

Small fir tree cone is seen in the geometric center of the brain. This is exactly what epiphysis – special endocrine organ looks like, its weight is just 0,1 gram. 4 thousand years ago old Hindu called it conoid body. Old peoples thought conoid (pineal) body's functions were fortunetelling, clairvoyance, and also judgment and analysis of former reincarnations. According to above mentioned peoples, epiphysis is a "third eye". French philosopher René Descartes developed tract on pineal body, where called it "a place for soul". Earlier Leonardo da Vinci expressed the same idea.

Function of pineal body was unclear to medical society till the mid 20th century, when americian dermatologist A. Lerner (who was looking for effective cosmetic lightening substance to treat pigmentary dermatosis) paid attention to article published in 1917 by British scientists F. Allen and K. McCord, who stated that tadpoles body color lightens if they are fed with pineal body extract [6]. Pineal body is proved to a biological clock (pineal body's extract – hormone melatonin – regulates pigment metabolism, sexual functions [1], circadian and seasonal rhythm [2], cellular maturing and division processes [4], participates in formation of visual and color perception, sleep and wakefulness, etc.; besides, it has demonstrated anticancer activity in experiment: the cancer growth slows under artificial lighting; melatonin level in the serum of patients with early stage cancer is increased 1,5-2 times compared to normal, and is significantly decreased after metastatic

growth [5]. Diurnal excretion of melatonin in cancers is changed [2]. According to I. Kvetnoi (1988): 1.One third of cancer tissue cells synthesize different biogenous amines, including melatonin; 2. Cancers producing melatonin grow slowly and clinically progress in a more benign manner (it might be manifestation of organism's self-protection). A systematic review of unblended clinical trials involving a total of 643 cancer patients using melatonin found a reduced incidence of death [7].Women with the brightest bedrooms have an increased risk for breast cancer [8]. Reduced melatonin production has been proposed as a likely factor in the significantly higher cancer rats in night workers [10]. Many biological effects of melatonin are produced through activation of melatonin receptors [1], while others are due to its role as a pervasive and powerful antioxidant [3], with a particular role in the protection of nuclear and mitochondrial DNA [9].

It was suggested to "attach" cytostatic substances to melatonin antibodies so that after administration they would accumulate in cancer cells and destroy them. Still there is a question: are cancer cells producing melatonin or are they accumulating it? Though the experiment demonstrated [5], that they also synthesize biogenous amines. Still it is not excluded that they accumulate them, or both mechanisms work together. Cancer cell (gradually) has less ability to produce something – as it is a classical mutant and eventually it can't even divide!

Melatonin was found in endothelial cells, mast cells, gastrointestinal tract, heart, retina, genital system, thymus, leucocytes and thrombocytes. It dissolves in water-soluble, fat-soluble, passes through all barriers, penetrates everywhere, has four receptors (one of them nuclear); is found in every live organism; seems to be universal.

Material and Methods

The aim of our research was to study the antiblastic effects of melatonin on cancer cell culture. Experiments were performed on 22 cases of prostate cancer (adenocarcinoma)cell culture and 30 cases of breast cancer (mammary ductal adenocarcinoma) cell culture. Normal epithelial cells of prostate and breast have been used by control. By using exclusive technology, from developing animal tissues we have derived the apyrogenic (assessed by LAL-Test System ((Limulus Amoebocyte Lysate – Endotoxin Testing)) "cocktail", which (in our opinion) should increase the expression of melatonin receptor MT1A in tissues. Mentioned "cocktail" was used for activation of melatonin receptor genes in cancer cell cultures; besides, melatonin was used in a doses of 0,4mg/kg and o,8mg/kg. Exposure of adequately marked so called activated cancer cell cultures to melatonin and assessment of its anticancer activity have been done by using ATP-TCA (Tumor Chemosensitivity Assays) method; standart data (of normal tissues) were used by control. Expression of universal nitric oxide synthase (u-NOS) after stimulation of melatonin receptor MT!A expression was studied as well; standart data (of normal tissues) were used by control. Results of research was analysed statistically by using computer program SPSS 12.

Results

Exposure of adequately marked so called activated cancer cell cultures to melatonin and assessment of its anti-cancer activity by using ATP-TPA method demonstrated the following results:

I. "Cocktail" increases by 41% expression of melatonin receptor MT1A in cellular cultures of prostate cancer and by 57,5% in cellular cultures of breast cancer compared to the control data (Fig. 1-2);

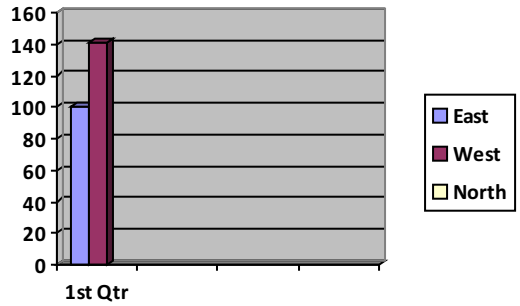

Figure 1. Diagram. "Cocktail" increases by 41% expression of melatonin receptor MT1A in cellular cultures of prostate cancer compared to the control data

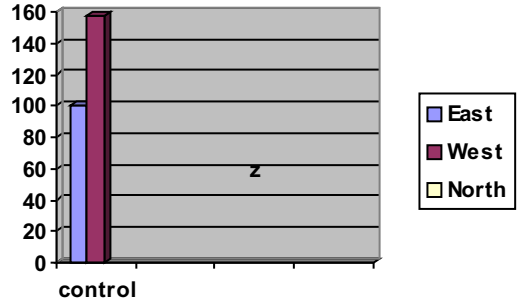

Figure 2. Diagram. "Cocktail" increases by 57,5% expression of melatonin receptor MT1A in cellular cultures of breast cancer compared to the control data

II. Melatonin in a dose of 0,4 mg/kg suppress metabolic activity of prostate adenocarcinoma cellular cultures by 51,4% and mammary ductal adenocarcinoma cellular cultures – by 67,0% compared to standard control (Fig. 3-4);

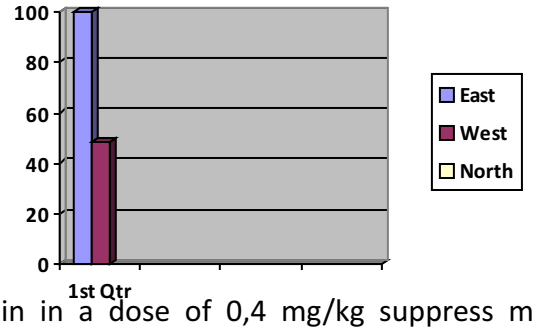

Figure 3. Diagram. Melatonin in a dose of 0,4 mg/kg suppress metabolic activity of prostate adenocarcinoma cellular cultures by 51,4% compared to standard control

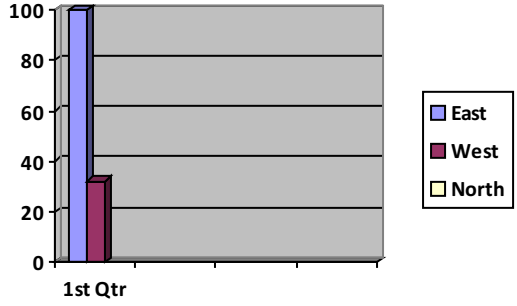

Figure 4. Diagram. Melatonin in a dose of 0,4 mg/kg suppress metabolic activity of mammary ductal adenocarcinoma cellular cultures – by 67,0% compared to standard control

III. Melatonin in a dose of 0, 8 mg/kg suppress metabolic activity of prostate cancer cells (in cultures) by 56,7% and breast cancer cells (in cultures) – by 71, 1% compared to standard data (Fig. 5-6);

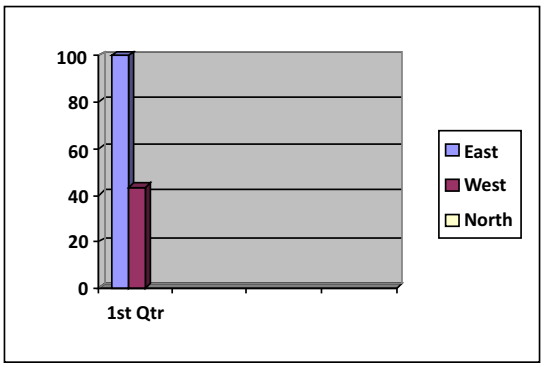

Figure 5. Diagram. Melatonin in a dose of 0, 8 mg/kg suppress metabolic activity of prostate cancer cells (in cultures) by 56,7% compared to standard data

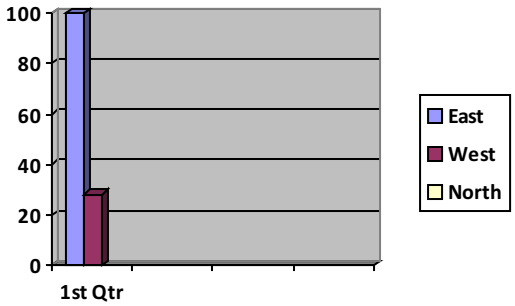

Figure 6. Diagram. Melatonin in a dose of 0, 8 mg/kg suppress metabolic activity of breast cancer cells (in cultures) by 71, 1% compared to standard data

IV. Expression of universal nitric oxide synthase (u-NOS) after stimulation of melatonin receptor MLTA1 expression increases in cases of prostate adenocarcinoma in total 29,7 times and in cases of mammary ductal adencarcinoma 32,3 times compared to control (Fig, 7-8).

Figure 7. Diagram. Expression of universal nitric oxide synthase (u-NOS) after stimulation of melatonin receptor MT!A expression increases in cases of prostate adenocarcinoma in total 29,7 times compared to control

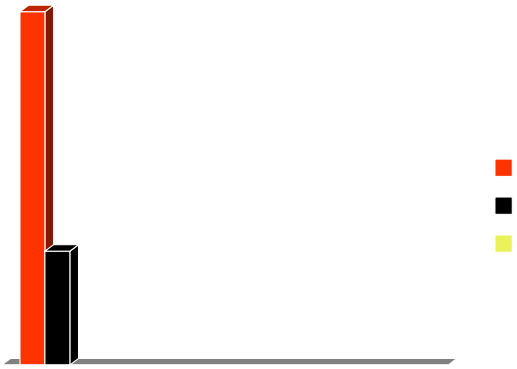

Figure 8. Diagram. Expression of universal nitric oxide synthase (u-NOS) after stimulation of melatonin receptor MLT1A expression increases in cases of mammary ductal adencarcinoma 32,3 times compared to control

Conclusion

We suppose that melatonin receptor genes' activation can be effective in the therapy (treatment) against the neoplastic processes.

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