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DEVELOPMENT OF EFFECTIVE SUBTLE ENERGY PATTERNS FOR PREVENTION AND TREATMENT OF CANCER AND INCREASING LONGEVITY

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A. TARGETING CANCER

Background:

In many tissues, a cellular hierarchy exists in which a small population of stem cells is responsible for the production of the mature cells of the organ. The stem cells maintain themselves through a process known as self-renewal. Similarly, tumors contain a minority population of <u>cancer stem cells (CSC)</u> that maintain the tumor. The CSC can give rise to both more CSC as well as cancer cells without the "stemness". In marked contrast to the CSC, these latter cells have either no or a markedly diminished capacity to form new tumors. <u>Therefore, to treat cancer effectively, the CSC must be eliminated.</u> Otherwise, the tumor will rapidly reform if the therapy eliminates cancer cells but spares a significant population of the cancer stem cells.

Immunotherapy of cancer is a promising approach, which can be applied in conjunction with the existing treatment strategies, i.e. surgery, chemotherapy and radiation therapy. The addition of immunotherapy with cancer vaccines would help in the elimination of the remaining tumor cells. The most important feature of the cancer vaccine approach is the fact that it can target micro metastases in the body of the patient. Micro-metastatic disease is one of the main reasons for the poor outcome in many cases. Because the immune system can localize and destroy small cancers in the body, immunotherapy is a very important addition to the standard treatment options. Ultimately, multiple vaccine strategies, applied in synergy, will most likely be responsible for the future success of cancer immunotherapy.

Proposed Studies:

We will perform advanced research with cancer cells and cancer stem cells to determine the most effective Subtle Energy Patterns developed with Vital Force Technology (VFT) for suppressing cancer cells & cancer stem cells' proliferation and transforming them into non-cancer cells. This research will be performed at the state-of-the-art facilities at UCSD: one of the top research universities in the World: (http://ucsdnews.ucsd.edu/newsrel/general/10-09NSFRankings.asp); (http://ucsdnews.ucsd.edu/about/).

Specifically, we will design precise assays to determine the efficiency of suppressing the growth of the cancer cells and the cancer stem cells in test tubes as well as in laboratory animals with experimental tumors. In these experiments we will treat the cancer cells and the animals with energy-infused medium and supplements. We will then determine the effects of these Subtle Energy Patterns on the genetic and protein-expressing profiles of treated cells (genomics and

proteomics analyses) utilizing array technologies. These studies will include analysis of telomerase expression (an important enzyme related to cancer cell survival and longevity), anti-apoptotic proteins (proteins related to cell death), immunological markers of cancer, stem cell markers, and other important elements of the cell structure and immune function.

Significance:

The importance of these studies is two-fold: 1) As a result of this research, we will develop products in the form of energy infused supplements that can be used for preventing and treating cancer. 2) In addition, energy patterns found in this research will be used as complementary tools to existing cancer treatment methods.

The identification of CSC and the design of nanoparticle-based vaccines for specific targeting of CSC will undoubtedly contribute to the development of new therapeutic strategies aimed at eradicating the tumorigenic stem cell subpopulation within cancer. The ability to prospectively identify, isolate and study cancer stem cells will significantly alter the way we think about, prevent, and treat cancer. The design of nanoparticle vaccines targeting cancer stem cells is highly innovative and holds great promise. This approach has never been applied to the prevention or treatment of any type of cancer. Therefore, the findings of this study will be translated to clinical applications, which will benefit many patients with cancer and will reduce the human suffering and the impact of this disease.

Initial results:

We performed several initial research experiments with cancer cells to determine the most effective Subtle Energy Patterns developed with Vital Force Technology (VFT) for suppressing cancer cells' growth.

Specifically, we determined the efficiency of suppressing the growth of the cancer cells in test tubes. In these experiments we treated 2 types of Human cancer cells with energy-infused growth medium: i) The Breast Cancer cell line MDA231, and ii) The Acute Monocytic Leukemia cell line THP-1. We determined the effects of these Subtle Energy Patterns on the growth characteristics of the treated cells.

The energy-treated media were prepared at the Energy Tools International, LLC by infusing standard growth media RPMI-1640. The media were then shipped to Minev lab for testing. An effort was made to spray the tissue culture room, tissue culture hood, incubators, and all flasks with Clean Sweep energy formula during the experiment to establish an energetically favorable atmosphere.

REPRESENTATIVE EXPERIMENTS WITH THE CANCER CELL LINE THP1

Results with initial energy formula presented in Fig. 1: From this chart one can see that energy infused liquids suppressed cancer cell development, even at very low concentrations.

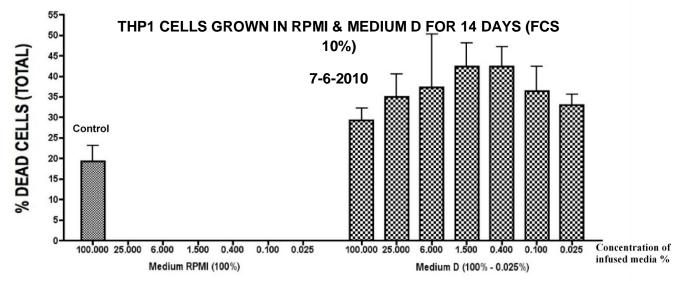


FIG. 1: Percentage of Dead Cells

This experiment demonstrated that even at a significant dilution (0.025%) the energy-infused medium is able to kill the cancer cells. This novel finding might be used directly to treat patients with cancer, or in combination with other cancer-targeting strategies.

After the first experiments the energy formula was modified and improved, resulting in a much stronger suppression of the proliferation rate of cancer cells. Presented below are the results of a 2 week experiment

THP1 CELLS GROWN IN RPMI & MEDIUM TA 65 FOR 14 DAYS (FCS 10%)

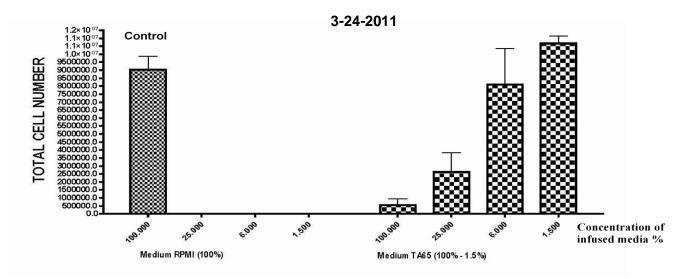
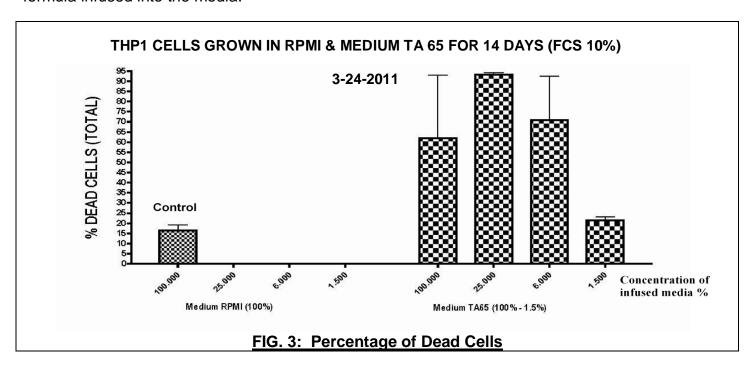


FIG. 2: Total Number of Cells Grown

The initial number of cancer cells was 15,000. After 14 days the control had a total of 9 million cancer cells, and the 100% concentration of VFT infused media had a total of 500,000 cells. This indicates a very strong suppression of the proliferation rate of cancer cells by the energy formula infused into the media.



From Fig. 2 one can see that in the media infused with the VFT energetic formula the number of cancer cells that grew is 18 times less than in the control. From Fig. 3 it follows that at least 60% of these cells are already dead, whereas in the control up to 20% of the cells were dead. This means that in the energy infused media, in comparison with the control, less than 3% of the cancer cells are alive.

These results demonstrate the potential for using VFT energetic formulas in a variety of ways such as creating additives to existing and future anti-cancer vaccines, and as a main component in cancer preventative remedies and supplements.

B. INCREASING LONGEVITY

Background:

The extension of life expectancy is one of the most dramatic achievements of the 20th century. The elderly population, over 65 years, is the fastest growing age group. In this project we will attempt to develop new approaches to increasing longevity by utilizing two strategies: (1) Stimulating the human immune system to prevent and fight cancer, and (2) Increasing longevity of healthy cells and tissues.

- 1. Cancer has become a leading cause of death in our aging society. That is why the efforts to find new ways to fight cancer are very important. Among the body's potential defenses are white blood cells called cytotoxic T lymphocytes (CTL). CTL can recognize and kill cancer cells, but only if they see complexes of protein fragments (peptides) attached to transplantation or major histocompatibility complex (MHC) molecules on the surface of the cancer cells. We found that peptides derived from the sequence of human telomerase are naturally expressed on the surface of cancer cells in association with MHC molecules where they serve as targets for CTL. Since numerous studies show that telomerase is the most widely expressed and specific tumor marker presently known, we believe that human telomerase could serve as an excellent target tumor antigen. Telomerase is a ribonucleoprotein, which when activated, synthesizes telomeric DNA and compensates for its loss with each cell division. Maintenance of a constant telomere length ensures chromosomal stability, prevents cells from aging, and confers immortality. Therefore, telomerase is an excellent target for immunotherapy specifically directed against cancer, and free of unwanted side effects.
- 2. <u>Increasing the longevity</u> of healthy cells and tissues is an essential component of slowing down the aging process, improving quality of life, especially for the elderly, and preventing diseases, including cancer. We have begun experiments to program the activities of cells and their essential components, like telomerase, with subtle energy patterns developed using Vital Force Technology™. Increasing the activity of telomerase in healthy cells would prolong their life, while decreasing its activity in cancer cells can help to fight cancer.

Proposed Studies:

1. We will design, test and optimize <u>telomerase-based vaccines</u> for prevention and treatment of cancer. First, we will determine the telomerase activity in several cancer cell lines. We will then group the cell lines based on the level of the telomerase activity. In the next phase of this project we will use natural and modified telomerase peptides to try to generate cancer-specific CTL *in vitro*. We will immunize *in vitro* lymphocytes obtained from normal volunteers and from patients with cancer.

Importantly, we will grow these lymphocytes in media, infused with specific subtle energy patterns to stimulate their growth and reduce cell death. We will determine if these peptides can induce potent cytotoxic lymphocytes able to kill cancer cells *in vitro*. We will detect if telomerase-positive cancer cells are better targets for our CTL than the cancer cells not expressing telomerase. We will also use human dendritic cells (DC) loaded with a variety of natural and modified telomerase peptides to induce cancer-specific CTL. Since the dendritic cells are the most efficient antigen-presenting cells, we expect to be able to generate very potent and specific CTL. Peptide-pulsed dendritic cells are

currently being used in clinical protocols for cancer immunotherapy. In this project we will apply our new strategy for the design and synthesis of <u>polymeric nanoparticles</u> that enhance the delivery of our vaccines into the DC. These acid-sensitive nanoparticles, 200-500 nm in diameter, are chemically stable at pH 7.4 but degrade into linear polymer chains and small molecules under mildly acidic conditions. The molecular design of these nanoparticles should allow them to circulate in the blood, at pH 7.4, but then rapidly hydrolyze after internalization into the DC. These experiments will provide important insights on the possibility to improve the efficiency as well as to shorten the duration of the treatment as a result of using vaccine-induced CTL expanded in energy-infused media. Next, we will analyze all data and will compare the experiments for statistical significance.

2. We will test existing VFT energy patterns and develop new patterns targeted to regulate the activity of telomerase, the effectiveness of CTL, dendritic cells and the activity and proliferation rate of stem cells. These patterns will be incorporated into anti-cancer vaccines and other modalities. Patterns effective for prolonging the life of normal cells and increasing immunity can be used for further research in vivo to develop remedies for the prolongation of life.

Significance:

The use of immunogenic peptides derived from telomerase - the most common tumor marker, in the development of vaccines for cancer, is innovative and holds great promise. These peptide-based and nanoparticle-based vaccines might be used directly to immunize patients with cancer. Dendritic cells loaded with telomerase peptides can be used to elicit powerful anti-tumor immune responses in patients with cancer. In addition, telomerase-specific CTL might be extremely useful for cellular immunotherapy of cancer. Therefore, the findings of this study may be used both for prevention and for treatment of prostate, breast, ovarian and many other telomerase-expressing cancers.

Finding energy patterns for regulating the activity of telomerase, along with using energy patterns for programming stem cells can lead to the creation of unique, innovative and simple products (like energy infused supplements) for life extension and for preventing various aging related diseases.

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