



CONTROL OF EHRlich TUMOR METASTASIS IN MICE LIVER BY ELECTROMAGNETIC WAVES, CERATES CERATES SNAKE VENOM AND METHOTREXATE

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Abstract

The use of chemotherapy for cancer treatment is a successful modality. However, this treatment has its side effects, which limits its applicability. The aim of the present work is a comparative study of side effects of treatment of *Cerastes cerastes* snake venom and methotrexate on tumor progression in the primary site (thigh of the animal) and in secondary sites (metastasis) formation in the liver of mice. This will be achieved through histological, biochemical and tumor volume investigations on two main groups, (healthy animals and tumor bearing animals) to monitor side effects of the used methodologies on normal tissue and cancerous tissue. A total of 122 mice were used, divided into two groups, "A" and "B". Group A: normal control mice, was given saline 0.2 ml. Group B: tumor bearing mice, were subdivided into 3 subgroups: B1: received no treatment; B2: was interperitoneally injected with snake venom (0.75 µg/g); B3: was given 2.8 mg/kg methotrexate as standard chemotherapy. Histological examination of liver using hematoxyline and eosin (H& E), as well as blood analysis were performed. Results showed that exposure to ELF- MWs at resonance frequency 4.5 Hz, 2G was quite effective in limiting tumor growth and proliferation, with minimal effect on non- involved tissues. Its use proves to be much safe for treatment of primary tumor site and to secondary sites.

Keywords: Ehrlich Tumor Metastasis, Cerates Cerates Snake

Introduction

Scientific research has been concentrated mainly on cancer and its treatment,

most treatment protocols start with surgery followed by chemotherapy and/or, radiotherapy. Recent research is now targeting tumor cells with

low frequency) magnetic waves on biological cells is the generation of induced electric currents from ionic movements (Feytching *et al.*; 2005). These biocurrents will generate bioelectric potential in the cells, which will interfere with its running metabolic process (Hashish *et al.*; 2008). Therefore, one may find that ELF magnetic wave can have two effects: either an increase of metabolic activities within the cells, and/or changing or stopping these activities. The first reasoning is a rationale medical treatment based on magnetotherapy (Newell *et al.*, 2000). While the later reasoning supports the new findings of toxicity of ELF magnetic fields (Behari *et al.*, 2010). The frequency and form of the magnetic wave used in the present work is 4.5 Hz and its shape was square. These waves induce electric currents and waves inside the treated tissue, which inhibit Ehrlich tumor growth (Fadel, 2005 and 2010). At this frequency the applied induced electromagnetic waves inside the tissues cause destructive interference with the metabolic bioelectromagnetic waves generated during malignant cellular division, which affects the ionic motions evolved in the process (Fadel, 1998). Moreover, epidemiological studies include cases of intrauterine children and newly born children who were in continuous exposure to the magnetic field (Feytching *et al.*, 2005), however in the present investigation, treatment of tumors by magnetic waves in adult animals continued for a limited period of one week at a rate of 2h/day.

Significant decrease of serum glucose level was observed in normal animals exposed to 4.5 Hz, 2 G magnetic field. The significant decrease of glucose level in blood serum exposed to the magnetic field, indicates a high consumption rate of glucose in order to provide energy to those injured cells and to enhance the repair mechanism (Tu and Giltner, 1974). Significant decrease of serum glucose level was also observed in normal animals exposed to 4.5 Hz, 2 G magnetic field in contrary with these results are the findings of Elferchichi *et al.*, (2010) who used static magnetic field on rats 1hr/day for 15 consecutive days which displayed increase in both plasma glucose level and cholesterol level this may be due to different animal species and different type of magnetic field. In this study significant increase in granulocyte % was observed and non-significant difference in total leukocyte and lymphocyte in normal mice exposed to magnetic field. Further support to the present results was the finding of Hashish *et al.*, (2008) using static magnetic field and extremely low frequency electromagnetic field for whole body exposure in mice, in addition of the results of Cakir *et al.*; (2009) using ELF-EMF 50 Hz on rats that were exposed to EMF for 50 .The increase in granulocyte % indicates that there is relation between exposure of extremely low frequency electromagnetic field and the oxidative stress through distressing redox balance leading to physiological disturbances Hashish *et al.*, (2008). The tumor bearing mice were concerned with the

effects of chemotherapy (Methotrexate) and 4.5 Hz, 2 G magnetic waves on Ehrlich tumor implanted in the thigh (primary site) and in the liver (secondary site)metastasis. These treatments were consequently followed by investigation of tumor growth at the primary site (thigh) and histological examination of liver sections as a secondary site (metastasis), in addition to estimation of some biochemical parameters of the blood. This is clear from the rate of tumor growth in the primary site in the thigh, where aggressive growth of the tumor occurred in a similar manner to that of untreated animals. The results also indicate that the growth of tumor volume in the thigh of animals treated with either 4.5 Hz, 2G magnetic waves or with Methotrexate was minimal as compared to that of untreated group. Similar growth rate of tumor in the primary site for both Methotrexate and 4.5 Hz magnetic waves occurred. However, as a result of the toxic effects of chemotherapy (Mycek *et al.*, 2000), a massive destruction of hepatocytes at the hepatic lobules occurred and supported by the findings of Zachariae, (1990), an observation, which didn't occur in animals, treated with magnetic waves. The effect of Methotrexate differs from the results obtained from using Methotrexate in combination with hydroxychloriquine for its antireumatic effect as the tested drugs didn't show hepatic toxicity and this is due to using hydroxychloroquine that reduced the dose of methotrexate, thereby reducing its hepato-toxic effects (Shashikumar *et al.*; 2010). In

this study significant increase in total leukocyte count was observed in all tumor bearing groups. However this result is compatible with the significant increase in total leukocyte count that was observed by Nakamura *et al.*; (2002) who used methotrexate on pulpal inflammation in rats.

The increase in total leukocyte count of tumor bearing mice treated with Methotrexate but no change was observed in plasma albumin level, these results were in compatible with the result of Rofe *et al.*; (1994) using Methotrexate therapy in tumor bearing rats where there are decrease in total white cell count and decrease in plasma albumin level. This incompatibility may be due to difference in dose administrated, species of animals and environmental conditions. Also, non significant change in serum cholesterol level was noticed for tumor bearing mice in all treatments.

The formation of aggregates of tumor cells in the liver of group exposed to magnetic field in-addition to the formation of cells exhibiting multinucleoli are markers for the start of repair mechanism (Fadel *et al.*, 2003). Further support to the present results(repair mechanism)was the finding of Truler *et al.*; (1998,2000) who have studied the effectiveness of low level direct current in liver metastasis tumor growth, and Traitcheva *et al.*; (2003) who have studied the lethal effect of extremely low frequency pulsed electromagnetic field on human cancer cells. More investigations are needed to confirm the afore-mentioned observations. The

results of the change in tumor size after exposure to magnetic waves are in agreement with other published data (Fadel *et al.*, 2009).

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