# The biological effects of microwave irradiation in mouse tumor

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### Abstract

This work studies the efficacy of the chitosan and electromagnetic wave treatment for the liver tumor in mice. The liver tumor model was applied to 70 mice, divided into 7 groups. 5 groups were treated by feeding different molecular masses of chitosan. One group was treated by 3GHz electromagnetic wave. From the experimental results of the survival rate, the lowest molecular mass of chitosan (Oligo) obtained the best survival result of 9 mice. In addition, according to the histopathological slices, the cell differentiation was observed in the electromagnetic wave group.

#### Introduction

There are more and more wireless applications emerging for the convenient communications of human beings. However, the known safety of the electromagnetic wave is also increasing the fear in people. Hence, there are many researchers studying the detrimental effect of the electromagnetic wave to human beings. But no definite conclusion is made.

We also didn't find any obvious unhealthy results in the previous study of the microwave exposure to rats. In fact, millimeter electromagnetic waves (MMWs) are widely used for the treatment of many diseases in Russia and East European countries. They employ 42.2GHz, 53.6GHz and 61.2 GHz MMWs to treat pain, cardiovascular diseases, wound healing, skin disorders and cancer. Hence, we used a liver tumor model to study the efficacy of the electromagnetic wave treatment in mice.

#### Material and methods

70 of 4-week-old mice (BioLASCO Taiwan Co., Ltd.) were involved in this work. They were divided into 7 groups. One is control group and others are experimental groups. Six out of the experimental groups were treated with different molecular masses of chitosan as shown in Table I. The other experimental group was treated with electromagnetic wave.

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The mice of each group were housed to a cage in a room. The laboratory was maintained on a 12-hour light-dark cycle(light on 6:00-18:00h) and at an ambient temperature of  $20\sim22^{\circ}$ C and a relative humidity of 65%. Animals were given food and water ad libitum.

Until the mice were at eight weeks of age, Smmu 7721 hepatoma cells ( $1 \times 10^7 / 0.2 \text{ ml}$ ) was subcutaneously injected in the back of the mice. Once the size of the liver tumor grew up to 3 mm, the treatments of chitosan and electromagnetic wave exposure started. The 6 chitosan experimental groups were oral fed by 0.2 ml chitosan solution 6 times in one week for four weeks.

The electromagnetic wave experiment mice were housed in a cage placed above the microwave radiator as shown in Fig.1. The microwave radiator consists of a signal generator, a power amplified, a horn antenna and a power supply module. The signal generator is a frequency synthesizer controlled by a micro-controller. The generated 3-GHz signal is sent to the power amplifier, which deliver a 3-W output power to the following antenna, which is a double ridged (TEM) broadband horn antenna with an antenna gain of 12dBi. The experimental group is irradiated at the average irradiation power density of 0.1mW/cm<sup>2</sup> for 6 months.

After treatments were conducted for four weeks, the mice were anatomized under anesthesia for tumor imaging. The whole resected tumors were placed together as revealed in Fig.2.

For the examination of histopathology, the tumor obtained from mice was placed in a fixative which stabilizes the brain to prevent decay. The fixative is formalin (10% formaldehyde in water). The brain tissue was transferred to a cassette, a container designed to allow reagents to freely act on the tissue inside. This cassette was immersed in multiple baths of progressively more concentrated ethanol, to dehydrate the tissue, followed by toluene or xylene, and paraffin. During this 12 to 16 hour process, paraffin will replace the water in the tissue, turning soft, moist tissues into a sample miscible with paraffin, a type of wax. The processed tissue was then taken out of the cassette and set in a mold. Through this process of embedding, additional paraffin was added to create a paraffin block which was attached to the outside of the cassette. The process of embedding then allows the sectioning of tissues into very thin (2 - 7  $\mu$ m) sections using a microtome. The microtome slices the tissue ready for microscopic examination. The slices are thinner than the average cell, and are layered on a glass slide for staining.

### Results

The survival rate is listed as shown in Table 1. Fig. 2 shows the remaining tumors, which were resected from the survived mice.

The whole histopathological slices are all demonstrated in Fig. 3. Fig. 3.1 is the control group showing the hyperplastic tumor cells are surrounded by blood vessels. Mitosis is also observed. Fig. 3.2-3.6 show the histopathological slices of tumors, that were treated with different chitosan molecular masses. Basically, there is no significant difference observed in these slices except thrombosis, tissue necrosis and bleeding. Fig. 3.7 is the slice of the 3GHz

exposure treatment. It is interesting that the cell differentiation was observed.

## Discussions

According the survival rate shown in Table 1, the molecular mass of the chitosan has significant influence on the survival rate and the tumor size. The lowest molecular mass Oligo can effectively suppress the tumor and prolong the life of the mice.

In addition, the beneficial treatment of the electromagnetic wave is observed in the histopathological slice. Although the electromagnetic wave group only obtained a moderate survival rate, the cell differentiation was observed. Although there are still many tumor cells, part of the cells are likely to differentiate to normal liver cells. Therefore, we will design more electromagnetic wave conditions and try to obtain better cell differentiation and tumor suppression. Besides, a further biological mechanism of the experimental results will be studied in the future work.

# References

[1] Pakhomov, A. and M. Murphy, "Low intensity millimeter waves as a novel therapeutic modality," *IEEE* Trans. Plasma Sci., Vol. 28,34-40, 2000.

[2] Rojavin, M. A. and M. C. Ziskin, "Medical applications of millimeter waves," QJ Med., Vol. 91, 57-66, 1998.

[3] Pletnev, S. D., "The use of millimeter band electromagnetic waves in clinical oncology," Crit. Rev. Biomed. Eng., Vol. 28, 573-587, 2000.

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	Control	Chitosan	FM80	FL80A	FL80B	Oligo	3GHz
Molecular		$3.3 \times 10^5$	$3.2 \times 10^5$	$1.73 \times 10^{5}$	8.6×10 <sup>4</sup>	$8.2 \times 10^{3}$	
mass							
Survival	5	4	7	4	7	9	6

Table I Survival rate of the experimental groups

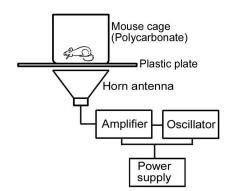


Fig.1 Microwave radiator.



Fig.2 Photograph showing the remaining tumors of survival mice.

Fig. 3.1 Control group	Fig. 3.2 Chitosan group				
Fig. 3.3 FM80 group	Fig. 3.4 FL80A group				
Fig. 3.5 FL80B group	Fig. 3.6 Oligo group				
Fig. 3.7 3GHz microwave group					