THE EFFECTS OF HIGH FREQUENCY ELECTROMAGNETIC WAVES ON THE VEGETAL ORGANISMS*

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The influence of radiofrequency (RF) and microwave (MU) electromagnetic waves of non thermal power density upon the assimilatory pigments in the vegetal tissues was studied. Increase in the photosynthetic pigment levels was found in both cases for relatively short exposure times (1 to 4 hours) while for longer exposure time (12 hours), in the plants exposed to microwaves, pigment content diminution was noticed The general increase of the photosynthesis efficiency was presumed to occur in the basis of indirect data provided by the enhanced ratio chlorophyll A/B. Slight stimulatory influence on the biomass accumulation was noticed in the case of MW exposure.

Key words: electromagnetic exposure, *Zea mays*, chlorophyll ratio, biomass accumulation.

1. INTRODUCTION

Since atmosphere "microwave window" allows penetration of cosmic radiation with frequencies ranging within GHz domain, there is always a microwave component within the environment radiation background. In the modern world, with the use of sophisticated microwave emitting devices for communications in the air, surveillance systems, industry, diagnostic and therapeutic purposes in medicine – the importance of electromagnetic pollution need not be over emphasized.

There is a rich literature describing the biological influence of low frequency electromagnetic fields upon vegetal organisms. But the responses elicited in various plant species appear to depend in a complex way on both the physical parameters of the irradiation source (frequency, power density, pulses or continuous waves, duration of exposure etc.) and the condition of the biological

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material magnetic (vegetation stage, pretreatment, environment etc.); nevertheless different biological parameters may lead to the revealing of different aspects of the interaction between the electromagnetic waves and the irradiated material. Some examples are illustrating the above statements.

Muraji *et al*. (1998) reported that an electromagnetic field of low frequency – near 10 Hz – has stimulated the growth of corn roots while for higher frequencies – above 240 Hz – the growth inhibition was noticed [1].

Fischer *et al.* (2004) have found that 16 Hz electromagnetic field of low intensity (20 microT) caused a small but significant increase in the fresh weight of young plantlets of *Helianthus annuus* and *Triticum aestivum* though germination was not influenced [2].

Pazur and Scheer (1992) published their findings related to the exposure to 7.8 Hz and 200 microT electromagnetic field of green algae *Chlorella*, underlying the inhibitory influence on cell division, while pigment content appeared to be unaffected [3].

Alexander *et al.* (1995) found that seed germination of onion and rice is accelerated if exposed to a weak electromagnetic field for 12 h and further, the seedlings showed significantly increased fresh and dry weight [4].

Yano *et al*. (2004) have shown that the main effects of low frequency electromagnetic fields (60 Hz frequency) on the photosynthetic activity in plant seedlings was consistent with the slightly perturbation of the early growth of the exposed plantlets, by changing the photosynthetic CO_2 uptake [5].

It seems that less literature is dedicated to the impact of high frequency electromagnetic waves upon vegetation since people is interested first of all in the risks of human professional exposure to radiofrequency waves and microwaves as well as in the assessing of the electromagnetic pollution health risks [6–7]. However, there are some experimental investigations on living bodies exposed to non-thermal microwaves and radiofrequency waves that focused on plants during early ontogenetic stages when the sensitivity to external constraints is higher – as in any other young organisms. The non-thermal effect of microwaves on the germination was reported in [8] on the basis of experiments with cabbage seeds which exhibited delayed germination in contrast with the expectation of stimulated germination – related to the well known thermal effect of microwaves. Biological effects of electromagnetic exposure on different biological parameters of the exposed seedlings were mentioned by Hamada (2007) that reported that under the influence of microwaves, wheat seedlings exhibited decreased ratio chlorophyll a/chlorophyll b, though increased protein accumulation occurred simultaneously [9]. Schmutz, P. *et al*. (1996) reported non-detectable changes in chlorophyll contents following long-term exposure of young spruce and beech trees to 2450-MHz microwave radiation [10].

Some data resulted from our own previous scientific work in the field of bioelectromagnetism have been communicated or published in the last decade. So, the positive influence of microwaves of non-thermal intensity on vegetal tissues at the level of chlorophyll biosynthesis was revealed in the case of two medicinal plant species: *Chelidonium majus* [11] and *Papaver pseudo-orientale* [12], while similar data were recorded also in cereal plant species [13–14]. The positive response of cereal plant species *Triticum aestivum* (wheat) and *Secale cereale* (rye) at the level of biomass accumulation and length growth following microwave exposure was also noticed [15–16]. Some evidences upon the genotoxicity of non-thermal microwaves were reported also by our multidisciplinary research group [17–19]. The biological effects of radiofrequency waves of non thermal level in plants – either grassy or woody – were also investigated, the slight photosynthesis stimulation being revealed for chronic exposures of young cereal plants [20] while the inhibition was evidenced in the case of tree seedlings [21].

In the next the authors present comparatively some effects of radiofrequency waves and microwaves on the plant growth, observable when the irradiation is carried out at low level of power density – as in the case of electromagnetic pollution impact on biosphere.

2. MATERIALS AND METHODS

BIOLOGICAL MATERIAL

Maize (*Zea mays* L.) grains were selected from single plant genitor in order to assure genophond uniformity: 25 grains in every sample. The plantlet growth was conducted within Angelantoni scientifica microclimate room at 12:12 hours light: dark cycle, at 24ºC thermostated environment. Pure distilled water was supplied to the maize plantlets in adequate aliquots all over the experiment. After 14 days, when the nourishing stocks from the initial caryopsides have been finished, the MW exposure and RF waves exposure have been carried out.

ELECTROMAGNETIC EXPOSURE

The device utilized for the exposure to radiofrequency waves was the transverse electromagnetic cell (TEM) designed to deliver low power density of 0.6 mW/cm^2, at a frequency of 418 MHz. The TEM device, built in aluminum, has adequate dimensions calculated to assure the characteristic impedance Z_0 of 50 Ω. The transverse propagation mode is the dominant one. The vegetal organisms were exposed to the TEM cell for various exposure times (continuous wave exposure):

1-2-4-12 hours. The microwave generator set-up was designed on the basis of an IMPATT diode. It is able to deliver microwave flow with the power density of about 10 mW/cm^2 at the level of its horn antenna at the frequency of 10.75 GHz. A probe detector measured the power density of electromagnetic waves flux, which was of about 1 mW/ cm^2 (low power density microwaves) in the plan of the plant grains (at the distance of 25 cm from the horn antenna).

SPECTRAL ASSAYS

Meterteck spectrophotometer type SP 870 Plus; chlorophyll and carotene extracts in 70% ethanol have been assayed on the basis of light extinctions at the wavelengths of 663 nm, 645 nm and 472 nm accordingly to Meyer-Berthenrath's modified method. The calculation formulae are:

$$chlA = \frac{12.3E(663) - 0.86E(645)}{1000dw}v \tag{1}$$

$$chlB = \frac{19.3E(645) - 3.6E(663)}{1000dw}v \tag{2}$$

$$t.c. = \frac{10E(472)}{2485dw}v \tag{3}$$

where:

– *chlA/B* = chlorophyll A/B content in mg/g;
– *t.c.* – total carotene content (mg/g);
– *E(l)* – light extinction to the wavelength *l*; *d* – cell width (=1cm);
– *v*– extract volume of ethanol 70% (ml); *w* – tissue weight (g).

STATISTICAL ANALYSIS

Three repetitions of experimental investigations upon assimilatory pigments were carried out for all experimental variant samples. Average values, standard deviations and *t-test* have been applied for statistical analysis. The statistic significance of the differences between exposed samples and control ones was assessed by means of the Student *t-test*.

3. RESULTS AND DISCUSSIONS

The experimental data – average values and standard deviation – are presented in the figures below.

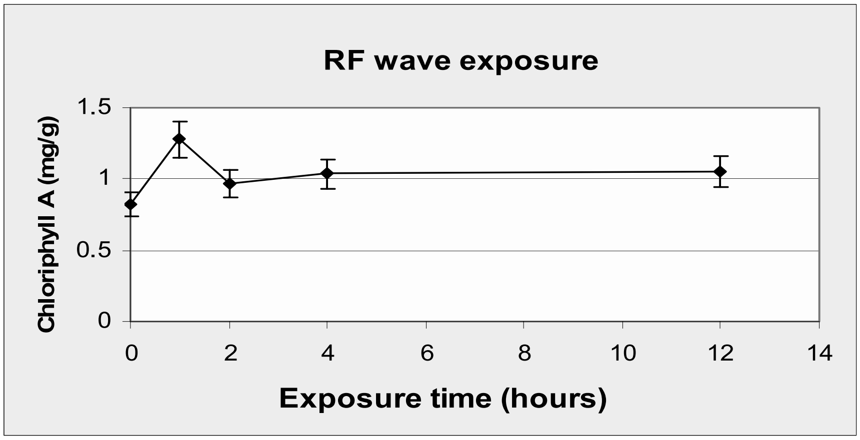

Fig. 1. – a. Chlorophyll A content after plant exposure to RF radiation.

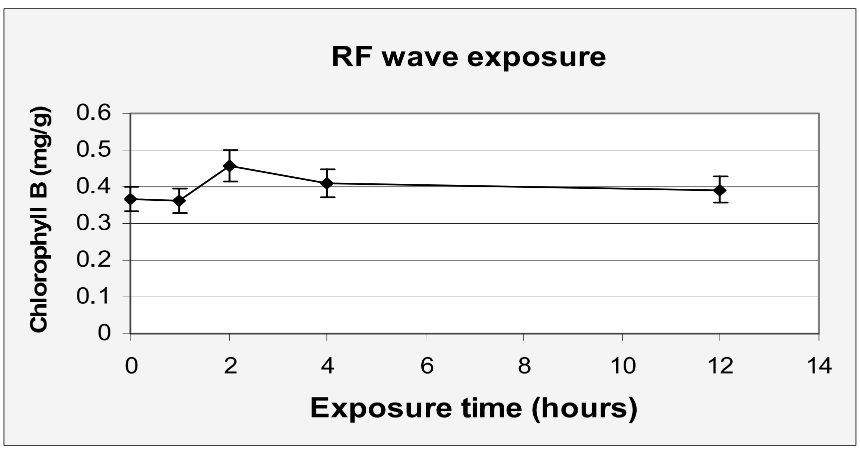

Fig. 1. – b. Chlorophyll B content after plant exposure to RF radiation.

As presented in the graphs from Fig. 1 a-b-c, slight stimulation of chlorophylls and carotenes biosynthesis was suggested by the data representing plant response to RF exposure. The values of all three pigment contents in the samples exposed to RF radiation were found higher than in the control – non exposed plants, but the statistic significance – relatively to the statistic threshold of 0.05 – was assured mainly in the case of short time exposures. So, the chlorophyll A content is considerably higher (with 60% in comparison to the control) for 1 h, while the chlorophyll B and carotene contents are the highest for 2 h (with up to 35% in comparison to the control).

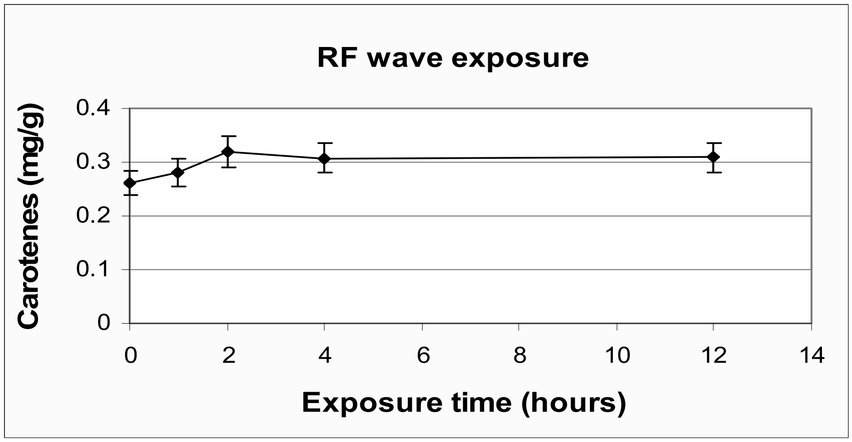

Fig. 1. – c. Carotenes content after plant exposure to RF radiation.

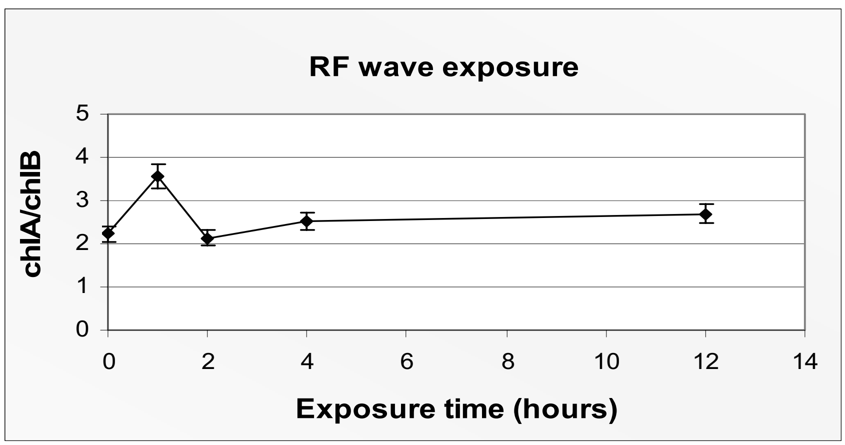

Fig. 1. – d. The chlorophyll ratio in plants exposed to RF radiation.

Chlorophyll ratio was significantly increased – with about 70% – for the exposure time of 1 h. The chlorophyll ratio is known as an indirect indicator of the energetic activity of LHC II system (light harvesting complex II, belonging to photosystem II from plant chloroplasts) that is controlling the first stage of solar energy conversion into its chemical form. It seems that, though thermal effects are not supposed to be involved – due to the low power density (up to 1 mW/cm^2, which is considered the thermal threshold) characterizing both types of exposure, the photosynthetic processes are positively influenced in the case of this plant species cultivated in controlled laboratory conditions. Following the irradiation the biosynthesis of chlorophylls and carotenes seems to be stimulated but there was no proportional increase of the pigment levels to the increase of the exposure time.

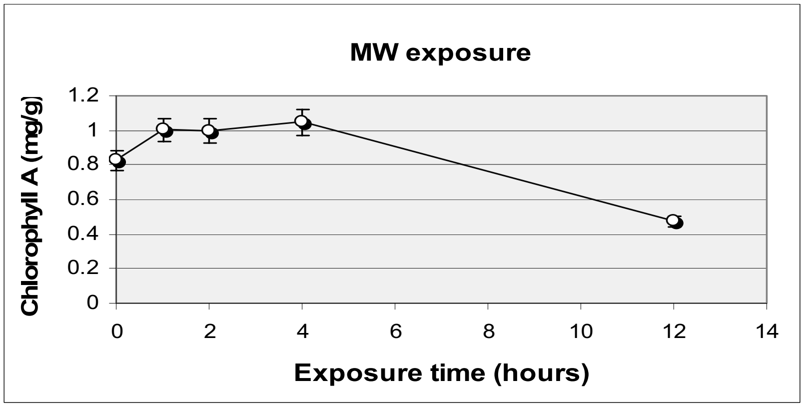

Fig. 2. – a. Chlorophyll A content after plant exposure to MW radiation.

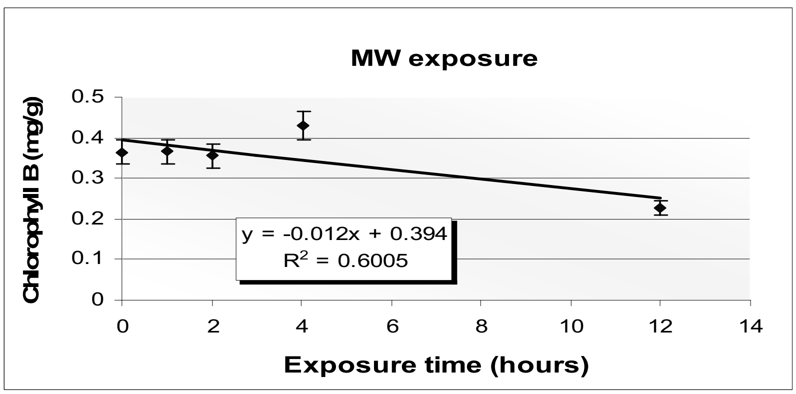

Fig. 2. – b. Chlorophyll B content after plant exposure to MW radiation.

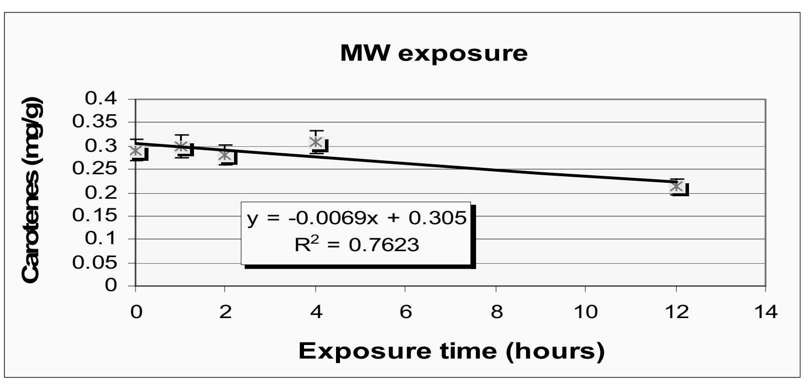

Fig. 2. – c. Carotenes content after plant exposure to MW radiation.

The considerable stimulatory effect corresponding to 1–2 h of irradiation is further attenuated for relatively long exposure time (12 h) – the plant organism response exhibiting accommodation abilities during time.

As can be seen in Fig. 2 a-b-c, chlorophyll and carotene biosynthesis were slightly stimulated for short exposure times to MW while for longer exposure times the pigment biosynthesis seems to be seriously affected.

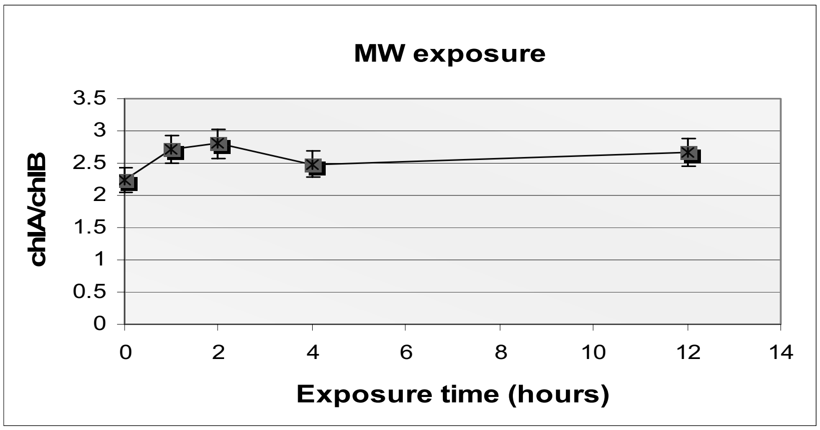

Fig. 2. – d. The chlorophyll ratio in plants exposed to MW radiation.

But the amplitude of the stimulatory effect experienced by the plants exposed for short times to microwaves was obviously lower if compared with that of plant exposure to RF waves of similar low power density: only 30% in chlorophyll A and, respectively, up to 15% in chlorophyll B and carotenes (for 4 hours) – with statistic significance relatively to the statistic threshold of 0.05. The diminution recorded in the photosynthetic pigment contents at 12 h of microwave exposure represents the most evident difference in the plant response to the two exposure ways, suggesting that the increased frequency (10.75 GHz comparatively to 418 MHz) may be the main cause of the biosynthesis decay for relatively long exposure time.

However, the chlorophyll ratio was generally increased in the exposed plants confirming the positive influence of electromagnetic exposure to MW on the photosynthesis efficiency, except the 12 h exposure where very slight damaging effect is suggested by the small diminution of the ratio chlorophyll A/chlorophyll B in comparison to the control non exposed plants (statistically non-significant as compared to the controls). Accordingly to the data presented in Fig. 3 a-b as well as to the statistic analysis, the biomass accumulation – though present some slight variations for short exposure times – seems to be not significantly influenced following the plant exposure to radiofrequency waves. The dry substance presents also slight variations versus the exposure time, but without statistic significance.

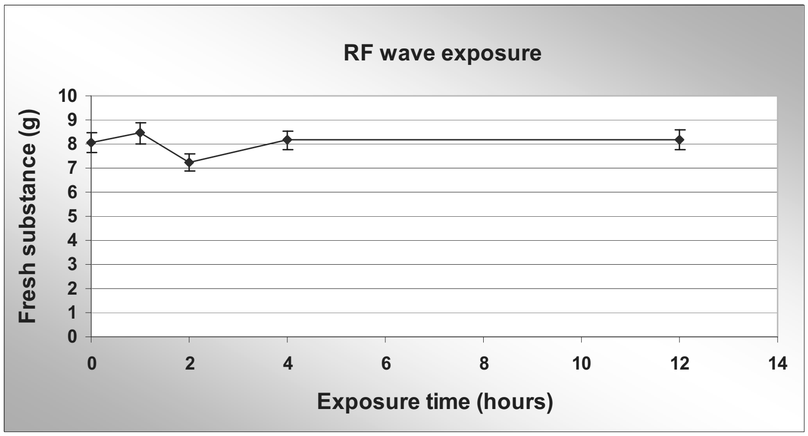

Fig. 3. – a. The fresh substance mass in the plants exposed to RF radiation.

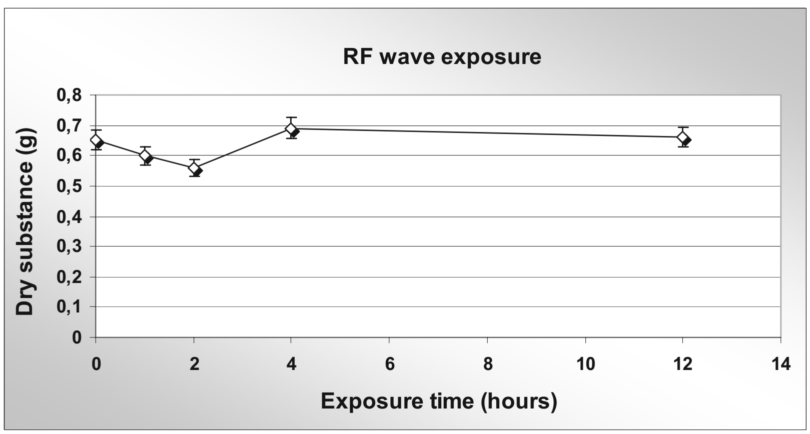

Fig. 3. – b. The dry substance in the plants exposed to RF radiation.

However, the chlorophyll ratio was generally increased in the exposed plants confirming the positive influence of electromagnetic exposure to MW on the photosynthesis efficiency, except the 12 h exposure where very slight damaging effect is suggested by the small diminution of the ratio chlorophyll A/chlorophyll B in comparison to the control non exposed plants (statistically non-significant as compared to the controls). Accordingly to the data presented in Fig. 3 a-b as well as to the statistic analysis, the biomass accumulation – though present some slight variations for short exposure times – seems to be not significantly influenced following the plant exposure to radiofrequency waves. The dry substance presents also slight variations versus the exposure time, but without statistic significance.

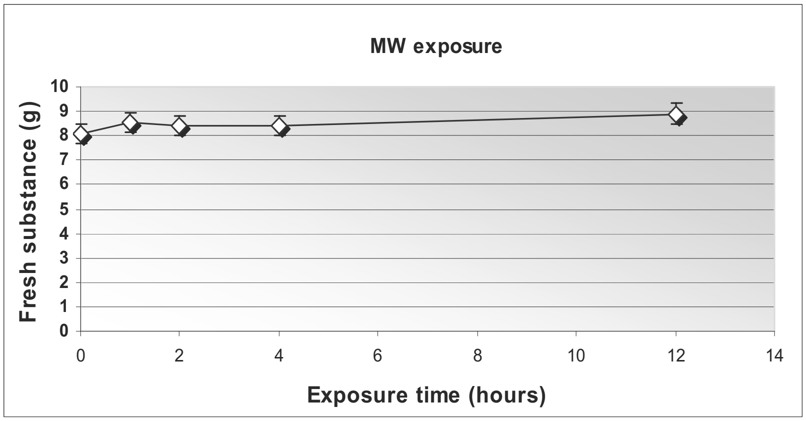

Fig. 4. – a. The fresh substance mass in the plants exposed to MW radiation.

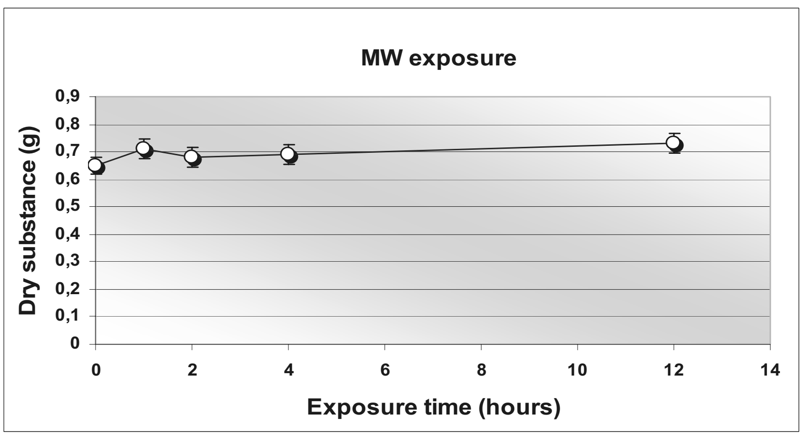

Fig. 4. – b. The dry substance mass in the plants exposed to MW radiation.

Slight stimulatory influence of MW exposure on the biomass accumulation seems to characterize the response of young maize plants to low power density microwaves: both the fresh substance mass and the dry substance were coherently slightly increased in the exposed plants in comparison to the control non-exposed ones (with statistic significance for 1; 4 and 12 hours)**.** Similar experiment is reported in literature by Hamada (2007): the author studied the effect of 10 GHz microwave exposure on the chlorophyll ratio in wheat seedlings. The increase of the chlorophyll ratio was noticed at 7th day but inhibitory one at 14th day of growth and in the same time the accumulation of proteins was found slightly increased following microwave exposure for no more than 1.25 h while Alexander *et al.* (1995) [4] have also found the increased accumulation of plant weight following the seed exposure for 12 hours to low electromagnetic field. The dependence of vegetal organism development on the frequency of electromagnetic waves was reported by Murajii *et al.* (1998) [1] following exposure experiments with low frequency electromagnetic waves (10 to 240 Hz) carried out on corn plants, the root growth being stimulated for lower frequencies but inhibited for higher ones.

There are several hypotheses formulated in order to explain from the physical viewpoint the cellular changes experienced by the exposed biological organisms but these ones consider mainly the low frequency or static magnetic fields: ion cyclotron resonance, ion parametric resonance and others. But for high frequencies the attempt to explain the cellular impact of the electromagnetic exposure remains at the biophysical and biochemical level of interpretation. So, it has become clear over the years that many magnetic effects in biology are probably due to alterations in membrane-associated calcium flux (Galland and Pazur, 2005, [22]), probably the deformation of membrane channels and modifications of the calcium ion functions. Na^+-channels seem to be affected to a lesser degree than Ca^2-channels (Rosen 2003, [23]). Less substantial seems to be the idea of water state changes under the impact of electromagnetic fields of both low and high frequency [24–25]. The most general phenomenological interpretation of high frequency biological effects assumes that RF or MW photon absorption is triggering a cascade of complex synergic cellular processes that can result in molecular damages leading either to DNA fragmentation or to free radical production. Or, free radicals as very reactive chemical species may interfere with various molecular processes involved in the growth of photosynthetic plants; this could be also the case regarding the changes evidenced in the frame of our experiment.

4. CONCLUSIONS

The exposure to RF and MW electromagnetic waves of low power density has elicited detectable responses in the young maize plants cultivated in controlled environmental conditions. Direct measurements of the levels of chlorophylls and carotenes showed stimulatory effect on the biosynthesis processes from vegetal green tissues when the exposure duration is relatively short (couple of hours). Indirect information on the photosynthesis efficiency provided by chlorophyll ratio suggested also positive effects of RF and MW treatment. The electromagnetic exposure of relatively long duration (over 10 hours) to ultra high frequency waves (MW) resulted in obvious inhibitory effects on the same biochemical parameters. The coherent influence on the biomass accumulation (both fresh mass substance and dry substance) was noticed in the case of microwave exposure for all exposure times. Non-thermal effects of RF/MW impact on the young plants seems to be responsible for the biological changes revealed in the frame of this experiment.

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