INFLUENCE OF MAGNETIC FIELDS OF VARIABLE INTENSITY ON BEHAVIOUR OF SOME MEDICINAL PLANTS

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Manuscript received: May 24, 2006; Reviewed: December 20, 2006; Accepted for publication: January 8, 2007

ABSTRACT

The present paper contains a study on the influence of some magnetic fields of variable intensity on two species of medicinal plants: Mentha sp. and Calendula officinalis. We had in view the effect of the magnetic field on the growth dynamic and the percentage in which the plants regenerated from callus.

Keywords: Mentha, Calendula officinalis, cuttings, micropropagation material, variable magnetic field

REZUMAT

In lucrare este prezentat un studiu despre efectele câmpurilor magnetice de intensitate variabilă asupra dinamicii de dezvoltare a unor specii de plante medicinale ca Mentha sp. şi Calendula officinalis.

REZUMAT DETAILAT

Cercetările realizate au urmărit influența și efectele ce pot apărea la menținerea unor plante medicinale în câmpuri magnetice de intensități diferite. S-au utilizat speciile de mentă: Mentha x piperita, Mentha spicata, Mentha villosa și specia de Calendula officinalis. Pentru inducerea culturilor in vitro s-au utilizat trei surse diferite de explante: minibutași de tulpină, fragmente de frunze (pentru speciile de Mentha) și semințe (pentru specia Calendula officinalis).

Culturile in vitro obținute au fost expuse timpi diferți în câmp magnetic de intensități de $0.4x10^{-4}$T, 3 X 10^{-4}Tși 0T. Obiectivele propuse au fost studierea dinamicii de creștere a materialelor obținute și procentajul de regenerare a plantelor din calus în câmp magnetic variabil.

În urma cercetărilor efectuate s-a constatat existența unei corelații între dinamica de germinare, creștere și regenerarea celor patru specii studiate din punct de vedere al influenței câmpului magnetic.

Diferențele înregistrate indică receptivitatea speciilor la stimulările sau inhibările produse de câmpul magnetic.

Keywords: Mentha, Calendula officinalis, cuttings, micropropagation material, variable magnetic field

INTRODUCTION

Domain literature presents experimental data regarding the influence of some physical environmental factors on various agricultural biosystems. The dynamic of in vivo germination has been taken into consideration, as well as the usage of some physical parameters in modulating and perfecting the treatment against the main diseases in agricultural and garden plants [1], [4].

Our research was centered on studying the modification of the genome in species of Mentha and Calendula officinalis under the influence of variable intensity magnetic fields [2].

Thus, the experiments had in view the choice of the explant type for the in vitro induction of medicinal plants followed by the exposure of the plants obtained in vitro in magnetic fields of variable intensity [5],[6].

We studied as well the effect of the magnetic field on the growth dynamic of the material obtained in vitro. We stimulated the callus genesis in Mentha and Calendula officinalis followed by the exposure of the callus for various periods of time in magnetic fields of variable intensity.

MATERIAL AND METHODS

For accomplishing these objectives, the following species of Mentha have been used: Mentha x piperita, Metha spicata, Mentha vilosa and Calendula officinalis.

For the initiation of a sterile culture, three different sources of plants have been used: mini-cuttings of stem, fragments of leaves and seeds [3].

In the case of Mentha sp. we used mini-cuttings of stem and fragments of leaves as initial material for micropropagation. The mini-cuttings were obtained from mother-plants cultivated in flowerpots.

The disinfection of the explants was done with a solution of NaClO 5% and 2-3 drops of TWEEN 80. The duration of the process was 15 minutes, and afterwards the material was washed in distilled water in order to eliminate the action of the disinfecting substance, the two types of mediums were tested:

- Medium I – Murashige-Skoog (1962) (table 1) to which we added 0.5 mg/l indolil acetic acid (AIA) and 1 mg/l Kinetin (Kin);
- Medium II – Murashige- Skoog (1962) (MS) to which we added 1.5 mg/l indolil acetic acid.

We noticed that the best results were obtained by using the second medium, with 1.5 mg/l AIA added. The result was the growth of normal developed plants only when using as explant mini-cuttings of stem.

All species were maintained in vitro by passing the culture on the same medium approximately every 6 weeks.

For this purpose, every 5 mini-cuttings were inoculated together on the second medium, in phials. All the seeds were inoculated on the same medium. Ten seeds were inoculated in each phial.

For the initiation of the callus culture we use the following explants: mini-cuttings of stem, leaf fragments sampled from the plants that have been maintained in the in vitro culture.

The mediums used to induce the genesis of the callus in all the four analyzed species are:

- MS1 – containing micro and macroelements in concordance with the standard recipe Murashige-Skoog, vitamins as in Morel and Wetmore prescription, saccharose 10g/l, manitol 50g/l, indolil butyric acid (IBA), 0,5 mg/l benzyl aminopurina, agar 0.8%.
- MS2 – the standard Murashige-Skoog medium enriched with 2.4-D, 20g/l saccharose, 0.8% agar

The pH of the two medium variants was adjusted to 5.8.

The culture recipients were maintained at a temperature of 24°C and a photoperiod of 14 hours light.

The callus genesis was observed at approximately 6 weeks from the moment of inoculation.

The leaf explants belonging to all the four species analyzed, inoculated in MS2 medium, turned brown after

Table 1 The prediction of the germination capacity in *Calendula officinalis*, depending on the period of exposure, under the influence of the magnetic field

Exposure time (hours)	**Calendula officinalis (%)**		
	0.4 x10^{-4}T	**0.0T**	**3.0 x10^{-4}T**
22	4.3	3.5	7.8
47	20.2	25.2	35.6
69	40.4	53.4	56.7
96	52.5	58.3	72.8

Table 2 The influence of the magnetic field on the growth dynamic

Exposure time (hours)	***Mentha x piperita* (mm)**			***Mentha spicata* (mm)**			***Mentha villosa* (mm)**			***Calendula officinalis* (mm)**		
96	0.4	0.0	3.0	0.4	0.0	3.0	0.4	0.0	3.0	0.4	0.0	3.0
	x10^{-4}T			x10^{-4}T			x10^{-4}T			x10^{-4}T		
	2	10	3.5	2.2	12	4.6	2.5	8	5.0	3	14	4

Table 3 The influence of the magnetic field on the regeneration and growth dynamic of the studied species

Species	**The intensity of the magnetic field**		
	0.4 x10^{-4}T	3 x10^{-4}T	0 T
Mentha x piperita (cm)	1.3	2.3	0.75
Mentha spicata	1.2	2.5	0.60
Mentha villosa	1.3	2.1	0.72
Calendula officinalis	1.5	1.2	2.6

4 weeks of in vitro culture.

Good results were obtained on MS1 medium, where the genesis of the callus took place at approximately 4 weeks from the inoculation.

This medium was used for repeated passages in order to maintain the callus in culture and afterwards for its passage on regeneration mediums.

The regeneration medium used to induce the genesis of small plants was the basic MS medium enriched with growth modulators.

The buds obtained from the callus were inoculated on fresh culture mediums after approximately 6 weeks.

In this case, the best combination of the growth modulators was the mixture of 1 mg/l indolil acetic acid (AIA) and 1 mg/l benzilaminopurina (BAP).

With this combination, the percentage of bud genesis from the callus was 26.6% if using 1 mg/l AIA and 1 mg/l BAP in Mentha x piperita, 25.5% Mentha spicata, 28.8% Mentha villosa and 45% for Calendula officinalis.

If using the combination of growth modulators 1 mg/l AIA and 1 mg/l BAP, the percentage of regeneration in the four species analyzed ranged between 11.5% and 13.9%.

The recipients were exposed to experimental varieties of the magnetic field, which consisted in:

- 0.4x10^{-4}T, present geomagnetic field, changeable, of low frequency, representing the standard sample;
- 3x10^{-4}T, high intensity static magnetic field, ancestral type, representing the maximum of the Terrestrial field;
- 0T, null magnetic field or the future terrestrial geomagnetic field.

The installation used for the type of field that cancels the permanent constituent of the Terrestrial magnetic field contains a Helmholtz type coil with a diameter of 1.2 meters. The coil was fixed on a wooden holder, with

no metallic component and it was orientated north-south with an angle between the axis of the coil and the vertical line as close to the geomagnetic field's orientation vector, as possible. The charging of the coil was made so that the total magnetic field inside it could be optimized at a low level.

In order to obtain a magnetic field of $3x10^{-4}$T, stronger than the natural one, we used a Helmholtz coil, identical with the one described, but containing a different number of whirls and using charging current of higher intensity. The wooden holder was also identical with the one described.

The recipients containing the inoculated material were placed approximately 2 meters away from the coil, at the same distance from the ground. The rooms that shelter the magnetic field installations are thermostats. The samples were studied for 5 days during which the results were collected after certain periods of time considered to be the exposure time of the samples in the magnetic field.

The recipients containing seeds were exposed for 22 hours, 47 hours, 69 hours and 96 hours. The biometric measurement of the growth dynamic for the phials containing mini-cuttings were performed 96 hours after the exposure in all the three magnetic fields. The results were processed.

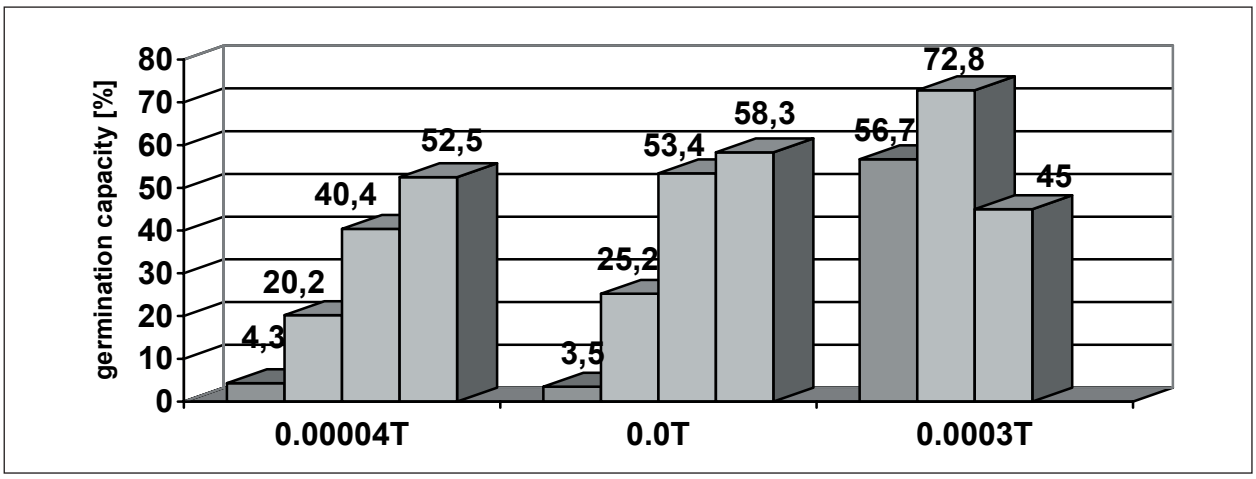

Fig. 1. The germination capacity

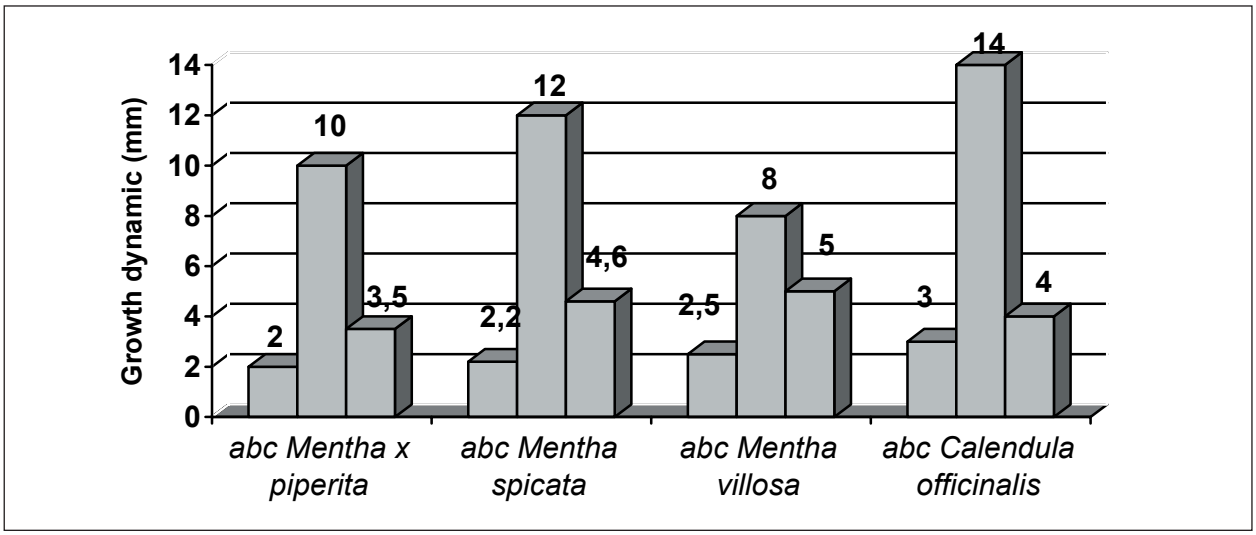

Fig.2.The growth dynamic in 94h exposure time;
a.in 0.4x 10-4T field, b.in 0T field; c.in 3.0x 10-4T field

RESULTS AND DISCUSSIONS

Data analysis revealed a correlation between the germination dynamic and the growth dynamic for the species studied in electromagnetic field.

The research on the germination degree for Calendula officinalis seeds, cultivated in vitro, have revealed various influences, depending on the time of exposure and the type of magnetic field tested.

The best results were obtained for the germination of seeds in a magnetic field of $3x10^{-4}$T, with a time of exposure of 96 hours, resulting in a germination percentage of 72.8%, while the lowest germination percentage for the same period of exposure was obtained in a $0.4x10^{-4}$T magnetic field. (Table 1, fig, 1).

Regarding the growth dynamic of the plants in vivo, we observed that from all the species of Mentha studied, Mentha spicata had the best evolution, reaching a length of 12 mm in 96 hours of exposure, in a 0T magnetic field. Analyzing Calendula officinalis, a better growth can also be observed in a 0 T magnetic field, reaching a length of 14 mm after 96 hours of exposure. (Table 2, fig.2).

The research on the germination degree in Calendula officinalis, in vitro, revealed various influences, depending on the time of exposure and the variant of the magnetic field.

The differences observed in the evolution of the growth dynamic in all the four studied species describe the receptivity of each species to the stimulation or the inhibition produced by the magnetic field.

Considering the influence of the magnetic field, there is a correlation between the regeneration dynamic and the growth dynamic of the plants, in all the four studied species.

Mentha had a better evolution in a $3x10^{-4}$T magnetic field, while in the 0T field it ceased its growth.

In what Calendula officinalis is concerned, the best results were observed in the 0T magnetic field (table 3).

Studying plant behavior in extreme conditions of the magnetic field, with 10 times higher intensity than the present one, simulating the conditions of the archaic geomagnetic field, which existed approximately five milleniums ago, represents a tempting subject.

The domain literature appreciates these conditions to be extremely favorable for the evolution of the vegetation and for the human being.

The analyses underlined the existence of a correlation between the germination and the growth dynamic in the studied species, regarding the influence of the magnetic field.

The research on the germination degree of the Calendula officinalis seeds, in vitro, showed various influences, depending on the exposure time and the tested variant of the magnetic field.

The best results were obtained for seed germination in $3x10^{-4}$T magnetic field, with a period of exposure of 96 hours, while the lowest percentage of germination at the same time of exposure was obtained in $0.4x10^{-4}$Tfield.

The differences in the evolution of he growth dynamic in the four studied species come from the different receptivity of each species at the stimulation or inhibition produced by the magnetic field.

In what growth dynamic is concerned, we observed that out of the studied species of Mentha, Mentha spicata had the best evolution in a 0T magnetic field, as well as Calendula officinalis.

CONCLUSIONS

Regarding the influence of the magnetic field, there is a correlation between the regeneration and the growth dynamic of the plants, in all the four studied species.

The research made concerning the level of Calendula oficinallis seed's germination in condition of vitro, showed different influence depending on the exposure time and tested field magnetic variants.

The best results concerning the seed's germination were obtained in the case of $3x10^{-4}$T magnetic field, at an exposure time of 96 hours, and the smallest percentage of germination at the same time of exposure was in the case of $0.4x10^{-4}$T magnetic field.

The differences registered in the evolution of growing dynamic at the four species taken into study shows the receptivity of the specie at the stimulation or inhibition produced by the magnetic field.

Regarding the growing dynamic of the seedlings in vitro condition, among the species of Mentha taken into study, the best evolution noticed was in the case of Mentha spicata, at 0T magnetic field, as Calendula oficinallis.

Concerning the magnetic field influence it is noticed the presence of an interaction between seedlings` regeneration and growing dynamic at the four species taken into study.

Peppermint had a better evolution in $3x10^{-4}$T magnetic field while in the condition of zero magnetic field it was observed a seedlings growing stagnation.

Regarding Calendula oficinallis specie, the best results were observed in condition of 0T magnetic field (table 3).

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