

ISSN 1392-3196 / e-ISSN 2335-8947

Zemdirbyste-Agriculture, vol. 100, No. 2 (2013), p. 185–190

DOI 10.13080/z-a.2013.100.024

The effect of strong microwave electric field radiation on: (2) wheat (*Triticum aestivum* L.) seed germination and sanitation

Irena GAURILČIKIENĖ¹, Jūratė RAMANAUSKIENĖ¹, Mindaugas DAGYS²,
Rimantas SIMNIŠKIS², Zenonas DABKEVIČIUS¹, Skaidrė SUPRONIENĖ¹

¹Institute of Agriculture, Lithuanian Research Centre for Agriculture and Forestry
Instituto 1, Akademija, Kėdainiai distr., Lithuania
E-mail: irenag@lzi.lt

²Center for Physical Sciences and Technology
A. Goštauto 11, Vilnius, Lithuania

Abstract

The seeds of the winter and spring wheat (*Triticum aestivum* L.) were subjected to a strong microwave electric field (SMEF) generated in a high power magnetron generator, working in a pulsed mode. Seed samples with different moisture contents (12, 15 and 18 %) were treated at the band 2.6, 5.7 and 9.3 GHz at 5, 10 and 20-minute exposures. Seed germination energy (GE), germination (G), abnormally germinated seeds (AG), germination index (GI) and seedling vigour index (SVI) were tested. The effect of SMEF on *Tilletia caries* infestation, plant productivity and seed sanitation was also explored.

Seed treatment with SMEF did not sufficiently effect on winter wheat seed GE and G; however, the increase in AG and reduction in GI and SVI were observed. The negative effect on GI and SVI increased when seeds with higher humidity were treated. Treatment of winter wheat cv. 'Kovas' seeds, artificially inoculated with *Tilletia caries*, with SMEF showed a trend towards reduction of bunt infected ears. Seed treatment with SMEF stimulated grain yield increase and the bands 2.6 GHz for 20 min, 5.7 GHz for 10 min and 9.3 GHz for 5 and 10 min induced an increase in grain number and grain weight per ear. The efficacy of the chosen SMEF bands and exposition time on winter and spring wheat seed sanitation were contradictory. The SMEF treatment against wheat seed borne pathogens such as *Phaeosphaeria nodorum* (causal agent of Stagonospora glume blotch), *Pyrenophora tritici-repentis* (causal agent of tan spot) and *Fusarium* spp. (causal agents of seedling and adult plant foot and root rots and *Fusarium* head blight) gave conflicting and inconsistent efficacy results. The composition of most frequently isolated *Fusarium* species on SMEF treated winter wheat seeds was pertained to SMEF bands and exposition time.

Key words: microwave, seed borne pathogens, seed germination, *Triticum aestivum*.

Introduction

The organic and sustainable low-input farming systems with reduced chemical inputs have become increasingly widespread. The lack of chemical seed treatment has caused the increase of many seed borne diseases, including common bunt (*Tilletia caries* (DC.) Tul. & C. Tul.), that were previously controlled with chemicals. The problem of bunt and other seed borne diseases in Europe has been aggravated by the European Commission Regulation No. 1452/2003, which provides that from the beginning of January 2004 all plant materials used for organic agriculture have to be produced under organic production conditions. The organic seed lots frequently do not make the grade and are often discarded because of contamination with common bunt (Lammerts van Bueren et al., 2003). In the United Kingdom, organic seed lots are predominantly contaminated with common bunt spores (McNeil et al., 2004). A 4-year monitoring of bunt incidence in grain samples showed an increase of bunt spores in winter wheat grain from low-input and organic farms in various locations of Czech Republic (Vanova et al., 2006). In Lithuania-grown wheat the outspread of bunt resumed in 1994 and in the year 2001 bunt was spread in 28.3% of the total area of winter wheat (Observation pests..., 2002). The control of seed borne diseases is of vital significance for the organic production.

There is a need for developing new methods for their prevention, monitoring and control and more quantified thresholds are required. Removal of the smallest seeds in a seed lot may in some cases limit the content of seed borne diseases. Presumptively, it is possible to reduce the impact of the attack of diseases such as loose smut (*Ustilago nuda* (C.H. Jensen) Rostr.), glume blotch (*Phaeosphaeria nodorum* (E. Müll.) Hedjar.), leaf stripe (*Pyrenophora graminea* S. Ito & Kurib.) and *Fusarium* spp. by removing the smallest grains of particular seed lot (Borgen, 2004). Hot water treatment is being applied in organic farming systems as an efficient sanitation method. Soaking of barley seeds in 50°C water for 30 min significantly reduced the incidence of *Fusarium* spp., *Alternaria* spp. and *Penicillium* spp. (Farahani, Chaichi, 2012). A sufficient control of *Pyrenophora graminea* was achieved by hot water treatment at 55°C and that of *Ustilago nuda* by soaking the seeds at 45°C and subsequent treatment at 55°C for 2–4 min (Nielsen et al., 2000). However, the hot water method is costly and complicated especially with large quantities of seeds, which are needed to be dried afterwards.

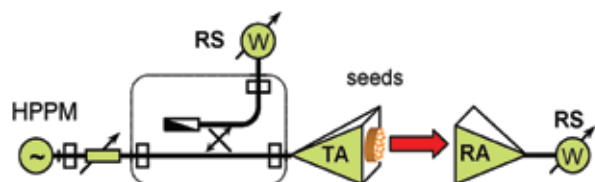
Different physical methods for seed microbial decontamination have been tested. Equipment for hot steam combined with ultrasound has been developed.

It eliminates *Tilletia caries* in wheat after four seconds and in spelt after eight seconds without germination vigour decrease (Borgen et al., 2005). As an alternative to chemical seed treatment in Lithuania, the use of ozonized air and electric field was investigated (Dabkevičius, Kreimeris, 1994). Thermotherapy and various physical methods such as seed treatment with water steam thermal impulses, electromagnetic and electrophysical factors were introduced later (Požėlienė et al., 2005; Požėlienė, Lynikienė, 2006; Dabkevičius et al., 2008). Seed treatment employing the above mentioned methods gave a reduction of some phytopathogenic fungi; however, in many cases thermal or physical seed treatment caused a reduction in seed germination. The innovative approach to eradicate seed borne pathogens is the use of microwave energy. Microwave seed treatment as one of the alternative chemical free ways to destroy seed borne microorganisms has been applied since the mid-twentieth century (Seaman, Wallen, 1967; Hankin, Sands, 1977). The contamination of *Cucumis melo* seeds with *Fusarium oxysporum*, the main causal agent of *Fusarium* wilt significantly declined after the treatment in microwave oven (Soriano-Martin et al., 2006); whereas *Phaseolus vulgaris* seed contamination with *Fusarium* spp. and *Alternaria* spp. fungi did not change, and that of *Penicillium* spp. significantly declined (Tylkowska et al., 2010). Application of microwave energy enabled successful control of seed phytopathogenic infection, improved sowing qualities of the processed seeds (the germination increased by 16–17%), and promoted intensification of growth processes (Vas'ko et al., 2004). The results of Reddy et al. (1998) indicated that eradication of *F. graminearum* in wheat seeds increased with the total microwave energy imparted, but the seed viability and seedling vigour decreased accordingly. Authors concluded that microwave irradiation can be used to significantly reduce seed borne *F. graminearum* in wheat. The percentage of seed infection could be reduced to below 7% (from 36.5% for the control), without reducing the seed quality below the commercial threshold of 85% seed germination.

The objectives of the present study were to elucidate the effect of radiation of high electric field non-heating microwave pulses on wheat germination and sanitation.

Materials and methods

Experimental set-up of microwave radiation, conditions and doses. In Centre for Physical Sciences and Technology on a basis of developed resistive sensors for high power microwave measurements, the experimental setup for microwave electric field monitoring and radiation of seeds was developed (Fig.). The sample with seeds was mounted in the aperture of radiating horn transmitting antenna (TA) as shown in Figure. To prevent



Note. HPPM – high power pulsed magnetron, RS – resistive sensor, applied as pulsed microwave power meters, W, TA and RA – transmitting and receiving horn antennas (set-up was developed at Centre for Physical Sciences and Technology).

Figure. Experimental set-up of microwave radiation on seed samples in semi-anechoic chamber

the microwave radiation to environment, the experiments were carried in a semi-anechoic chamber. The microwave source was high power magnetron (HPPM) generator, working in pulsed mode. The power transmitted to transmitting antenna was measured with resistive sensor (RS) connected to directed coupler inserted in transmitting waveguide, the radiated microwave power density was also controlled with RS attached to receiving antenna (RA).

As it is seen in Figure the container with grain was mounted in the centre of aperture of radiating antenna. The non-homogeneity of electric field in aperture can consist of up to 30%. It was supposed that the above mentioned non-homogeneity of electric field was acceptable for statistical evaluation of microwave effect on grain properties. Microwave power density D in the aperture of transmitting horn antenna was evaluated by dimensions of antenna aperture A and B and power and measured transmitted power P as follows: $D = \frac{P}{AB}$. Microwave electric field was calculated by the following formula: $E = \sqrt{DZ_0} = \sqrt{\frac{P}{AB}} Z_0$, where $Z_0 = 377 \Omega$ is vacuum impedance.

The dimensions and maximum radiated power levels on grain aperture of horn antenna are presented in Table 1. Wheat (*Triticum aestivum* L.) seeds were subjected to strong microwave electric field (SMEF) radiation. The main task while irradiating was to avoid the thermal heating of seeds by microwave energy. Seeds were treated in band S, C, X for 5, 10 and 20 min exposition, pulse duration 6 μ s, pulse repetition 50 Hz with electric field intensity (E) 42 (X), 38 (C) and 31 (S) kV m^{-1} (Table 1). The temperature of grain under exposition was measured. In all the treatments of experiments the rise of temperature did not exceed 8°C.

Table 1. Maximum microwave electric field level radiated on grain in aperture of horn antenna

Band	Frequency	Antenna aperture	Power	E
	GHz	A × B mm ²	kW	kV m ⁻¹
S	2.6	200 × 200	100	31
C	5.7	160 × 160	100	38
X	9.3	130 × 130	80	42

S, C and X – a strong microwave electric field band

Evaluation of seed germination and seedling vigour. The significance of seed moisture content before irradiation with SMEF for seed germination and seedling vigour was tested. Winter wheat cv. 'Kovas' seeds were dried with warm air flow (40°C), or moistened with sprayer to uniform 12, 15 and 18 % moisture content. Seed moisture content was measured with "Infratec 1241" ("Foss", Sweden). Seeds with 12, 15 and 18 % moisture content were subjected to a SMEF in high power magnetron at 2.6, 5.7 and 9.3 GHz band at exposure 5 and 10 min. The treated seeds were placed in "Phytotoxkit microbiotests" (Belgium) transparent plates (10 per plate) on wet sand, covered with black blotter. All treatments were replicated four times. The plates were put in an incubator at 20°C for 8 days with a 12-h light/12-h dark photoperiod. The plates were observed daily until day 8. The seeds were considered to be germinated when their root length reached the seed length and shoot length reached half of seed length. The percent of germinated seeds after 4 days was referred to as germination energy (GE), and after 8 days was referred to as final germination (G). After 4 days, the seeds showing only roots without shoot or only shoot without roots were referred to as abnormally germinated (AB). Germination index (GI)

and seedling vigour index (SVI) were calculated by the method of Cao et al. (2008):

$$GI = \sum \left(\frac{G_i}{t} \right), \text{ where } G_i \text{ is the number of germinated seeds on day } t;$$

$$SVI = GI \times H_s, \text{ where } H_s \text{ is shoot height cm.}$$

Evaluation of strong microwave electric field (SMEF) treatment on *Tilletia caries* infection and productivity of winter wheat. Field experiment. The seeds of winter wheat cv. 'Kovas' artificially inoculated with *T. caries* (5 g teliospores/1 kg seeds) were treated in SMEF at 2.6, 5.7 and 9.3 GHz band at exposure of 5, 10 and 20 min. Not treated in SMEF, *T. caries*-inoculated seeds were used as a control. The seeds were sown in the experimental field on 30 September, 2011 using a small plot drilling machine "Hege 90" (Austria) at a rate of 300 seeds per treatment in 3 m long single rows with four replicates. Germinated seedlings were counted two weeks after drilling and overwintered plants were quantified at the beginning of vegetation (17 March, 2012). At hard ripening stage (BBCH 87), *T. caries*-infected ears were counted in each row. Harvested sheaves from each row were thrashed with a "Wintersteiger LD 360" (Austria). A total of 25 ears randomly selected from each sheaf before thrashing were collected and thrashed with a portable thrasher "Wintersteiger LD 180" (Austria); the number of grains and grain weight per ear were calculated.

Evaluation of SMEF treatment on seed sanitation. Laboratory analysis. The effect of SMEF treatment at 2.6, 5.7 and 9.3 GHz band at 5 and 10 min exposures on winter wheat cv. 'Kovas' and spring wheat cv. 'Tibalt' seed borne infection with *Phaeosphaeria nodorum*, *Pyrenophora tritici-repentis* and *Fusarium* spp. was tested. Untreated with SMEF seeds were used as a control. Treated seeds were placed in Petri dishes 9 cm in diameter, 25 grains per dish. In total, 400 seeds per treatment (100 seeds per replicate) were tested for each pathogen. The grain infection level was evaluated in percent. To estimate seed borne infestation with *P. nodorum* the grains were not surface-disinfected prior to plating on SNAW agar. The dishes were incubated at 20°C for 72 hours, followed by 7–8 hours at –20°C to cause grain or seedling killing, and finally for 7 days at 20–23°C. After incubation, the grains were examined for the occurrence of *P. nodorum* by using ultraviolet (UV) light ("Cole Parmer", USA). Under UV light, fungus is detectable from typical green colour (Manandhar, Cunfer, 1991). Evaluation of grain infection by *P. tritici-repentis* was done by osmotic method. Filter paper was moistened

in sucrose solution (200 g of sucrose/1 l distilled water) prior to placing in dishes. Petri dishes were incubated under mix light using fluorescent day-light and NUV tubes. Incubation period was 6–7 days under 16 h day and 8 h night regime at 24 ± 1°C. Under these conditions, sucrose solution inhibits the germination of wheat grain, and favours pigment formation. Pigments of *P. tritici-repentis* fungi under NUV light shine in brick-red colour (Joelsson, 1983). For *Fusarium* spp. analysis, the seeds, surface-sterilized with potassium hypochlorite (1% Cl available) for 4 min, were placed on a potato dextrose agar and incubated at 26 ± 2°C in the dark. Microscopic studies of *Fusarium* fungi were carried out after 7–8 days. The effect of SMEF treatment on prevailing *Fusarium* species was done by the purified single spore cultures of *Fusarium* from infected grains of winter wheat cv. 'Kovas' and 'Mulan'. *Fusarium* species were identified on the basis of their cultural and morphological characteristics according to Leslie and Summerell, (2006). The frequency of occurrence (%) of prevailing *Fusarium* species was calculated.

Statistical analysis. Two-way analysis of variance was used for testing the effect of SMEF on germination of seed differing in humidity and seedling vigour. Significant differences among treatments were assessed by Fisher's least significant difference (LSD) test, ($P \leq 0.05$). Statistical analysis was done using the statistical data processing software package SELEKCIJA (software ANOVA, STAT) (Tarakanovas, Raudonius, 2003).

Results and discussion

The effect of strong microwave electric field (SMEF) on seed germination and seedling vigour. Seed treatment with a SMEF at 2.6, 5.7 and 9.3 GHz band at 5 and 10 min exposures did not exert any significant effect on winter wheat cv. 'Kovas' seed germination energy (GE) and germination (G); however, a significant increase in the quantity of abnormally germinated seeds (AG) and reduction of germination index (GI) and seedling vigour index (SVI) were observed (Table 2). The increase of seed moisture content from 12 to 15 and to 18% before treatment with SMEF exhibited a significant negative influence on seed GE, G, GI and SVI, while in the grain with 18% moisture content an increase occurred in the percent of AG seed. The interactions between the two factors, SMEF treatment and seed moisture content before treatment, for GE and G were nonessential but they were significant for AG ($P \geq 0.01$) and for GI and SV ($P \geq 0.05$). Comparison of SMEF bands 2.6, 5.7 and 9.3 GHz showed that bands 5.7 GHz exerted

Table 2. The effect of seed moisture content before irradiation (factor A) and strong microwave electric field (SMEF) irradiation at 2.6, 5.7 and 9.3 GHz band for 5 and 10 min (factor B) on seed germination energy (GE), germination (G), quantity of abnormally germinated seeds (AG), germination index (GI) and seedling vigour index (SVI)

Treatment	GE %	G %	AG %	GI	SVI
Seed moisture content (factor A)					
12%	83.6	83.2	25.7	4.32	30.7
15%	72.9	75.0	23.2	3.57	25.7
18%	75.0	76.1	34.6	3.36	21.4
LSD ₀₅ A	3.80	3.35	4.11	0.227	2.29
Sd _A	3.29	2.90	3.56	0.20	1.98
F-act.	5.95**	4.75*	5.69**	3.01**	11.02**
SMEF / time (factor B)					
Untreated	81.7	84.2	18.3	4.00	30.2
2.6 GHz 5 min	75.0	78.3	35.0	3.75	24.0
2.6 GHz 10 min	80.8	77.5	26.7	4.08	29.8
5.7 GHz 5 min	73.3	75.8	35.0	3.42	21.8
5.7 GHz 10 min	73.3	76.7	27.5	3.42	22.5
9.3 GHz 5 min	79.2	79.2	33.3	3.83	25.1
9.3 GHz 10 min	76.7	75.0	19.7	3.75	28.4
LSD ₀₅ B	6.58	5.80	7.12	0.393	3.96
Sd _B	5.02	4.43	5.44	0.30	3.02
F-act.	0.95	0.94	3.38**	1.49	2.61*

* – $P < 0.05$, ** – $P < 0.01$; Sd_A – standard deviation factor A, Sd_B – standard deviation factor B, F-act. – interaction of treatment and tested parameter

a negative effect on seed GE, G, GI and SV. The negative effect of all SMEF treatments on GI and SVI increased with increasing grain moisture ($P < 0.01$). The percent of AG seeds was found to be higher at 5 min exposure to SMEF all tested bands compared with 10 min exposure. These results are in contrast with the research evidence obtained by Aladjadjiyan (2010). His study on the influence of microwave irradiation treatment with the frequency 2.45 GHz on the germination of lentil seeds ascertained that longer exposure time had an inhibitory effect on plant development as well as higher output power of microwave irradiation. Shorter exposure time (30 s) demonstrated a higher stimulating effect than longer one. The results of research done by Nikulin et al. (2009) indicated that the effect of low intensity microwave radiation (6.74–7.44 GHz) on wheat germination and development could exert both negative (overwhelming germination and intensity of development) and positive (accelerating germination and development of growth) influence. The data obtained by Ayrapetyan (2004) strongly suggest the existence of non-thermal biological effect of microwave extremely high power pulses (EHPP) on seed metabolic dependent water uptake and its germination potential. Because water molecules are polar, they vibrate when subjected to microwave energy, causing considerable friction between molecules. The author suggested that EHPP-induced water structure changes must be different from traditional heated one and such differences could underlay in the ground of generation of non-thermal biological effect of EHPP.

The effect of SMEF treatment on *Tilletia caries* infection and productivity of winter wheat. The SMEF

treatment did not affect inoculated seed germination and winter survival; however, the trend towards reduction of bunt infected ears was observed (Table 3). Despite the nonessential SMEF effect on seed germination and only moderate reduction of bunt-infected ears, the grain yield increase was significant in all treatments ($P < 0.05$). The SMEF seed treatment at the band 2.6 GHz for 20 min, 5.7 GHz for 10 min and 9.3 GHz for 5 and 10 min was given the best response by the increase in grain number and weight per ear. According to these results, the SMEF treatment might produce some stimulating effects on winter wheat plant development and productivity. The stimulating effect of microwave treatment on increasing root fresh weight of bean and fresh weight and length of shoots of some ornamental perennials is presented in the study by Aladjadjiyan (2007), who has concluded that microwave radiation might have a promising future as a plant stimulation factor. Our results showed only a slight reduction of *T. caries* infestation, but subsequent studies of microwave treatment might yield some promising results. *T. caries* is listed as a plant quarantine pest in some regions of China, for whose control microwave treatment has been introduced to replace a highly flammable method of fumigation with ethylene oxide. Microwave treatment under 800, 640, 480, 320 and 160 W at exposition 40, 40, 50, 80, and 180 s respectively, completely killed *T. caries* teliospores on wheat grains. However, microwave treatment reduced germination rate, respiratory rate, accelerated the rate of deterioration of wheat. The treatment under 160 W for 180 s completely killed *T. caries* and ensured the best combination of wheat quality (Study..., 2012).

Table 3. The effect of strong microwave electric field (SMEF) irradiation at 2.6, 5.7 and 9.3 GHz band for 5, 10 and 20 min expositions on *Tilletia caries* infestation and winter wheat cv. 'Kovas' productivity

Treatment	Number of germinated seedlings	Overwintered plants %	Bunt infected ears (row)	Yield kg (row)	Grain number per ear	Grain weight per ear g
Untreated	190	88.8	52	0.499	32.7	1.20
2.6 GHz 5 min	185	92.9	35	0.657	35.9	1.28
2.6 GHz 10 min	189	93.6	41	0.616	34.6	1.26
2.6 GHz 20 min	184	90.3	47	0.669	40.0	1.46
5.7 GHz 5 min	171	91.8	28	0.653	36.7	1.28
5.7 GHz 10 min	192	87.9	40	0.664	37.2	1.34
5.7 GHz 20 min	194	90.1	31	0.661	35.0	1.26
9.3 GHz 5 min	165	94.6	39	0.805	36.7	1.38
9.3 GHz 10 min	161	94.8	59	0.794	38.2	1.45
9.3 GHz 20 min	187	87.5	35	0.650	33.0	1.20
LSD ⁰⁵	26.7	11.38	18.9	0.091	2.18	0.097
Sd	12.9	5.54	9.18	0.044	1.06	0.046
<i>F-act.</i>	1.64	0.48	2.17	7.59**	9.07**	7.19**

** – $P < 0.01$; Sd – standard deviation, *F-act.* – interaction of treatment and tested parameter

The effect of SMEF treatment on seed sanitation. The results of winter wheat seed treatment with SMEF against *Phaeosphaeria nodorum* (causal agent of *Stagonospora glume* blotch) infestation were conflicting: in winter wheat seed treatments with the band 2.6 GHz for 10 min and 5.7 for 5 and 10 min the trend towards pathogen stimulation was noted; however, with the band 9.3 GHz for 5 min the trend towards reduction of pathogen infection was observed (Table 4). The level of spring wheat seed infestation with *P. nodorum* was lower; however, the trend towards pathogen stimulation in all treatments was also observed, except for the band 9.3 for 10 min. The efficacy of SMEF irradiation against seed borne *P. tritici-repentis* (causal agents of tan spot) and *Fusarium* spp. (causal agents of seedlings and adult plants foot and root rots and *Fusarium* head blight) was inconsistent.

The most frequent *Fusarium* species on winter wheat seeds were *F. sporotrichioides*, *F. culmorum*, *F. poae* and *F. avenaceum* (Table 5). On the seeds of cv. 'Kovas', irradiated with SMEF at the bands 2.6 and 5.7 GHz, the ratio of these species changed: in longer (10 min) exposition

time treatments a clear reduction of *F. sporotrichioides* and an increase of *F. culmorum* were observed and conversely, a shorter exposition (5 min) of SMEF resulted in a reduction of *F. culmorum* and an increase of *F. sporotrichioides*. Conversely, on the seeds of cv. 'Mulan' the reduction of *F. culmorum* in *Fusarium* composition in longer exposition of irradiation was noted. Supposedly, the variation of *Fusarium* species composition was depended upon sensitivity to microwave irradiation of all complex of seed pathogenic and non-pathogenic mycoflora; loss of one species caused an increase in other.

Our findings indicated a significant dependence ($P < 0.01$) of seed humidity before treatment with SMEF on seed germination and seedling vigour. According to our results, the SMEF treatment might produce some stimulating effects on winter wheat plant development and productivity. However, we obtained contradictory results of SMEF efficacy on winter wheat seed sanitation. The trend towards reduction of bunt infected ears was observed. Supposedly, SMEF bands and exposition times used were insufficiently well-suited for reducing

Table 4. The effect of strong microwave electric field (SMEF) irradiation at 2.6, 5.7 and 9.3 GHz band for 5 and 10 min expositions on winter wheat cv. 'Kovas' and spring wheat cv. 'Tibalt' seed borne pathogens

Treatment	Infected grain %					
	'Kovas'			'Tibalt'		
	<i>Phaeosphaeria nodorum</i>	<i>Pyrenophora tritici-repentis</i>	<i>Fusarium</i> spp.	<i>Phaeosphaeria nodorum</i>	<i>Pyrenophora tritici-repentis</i>	<i>Fusarium</i> spp.
Untreated	9	21	40	1	20	34
2.6 GHz 5 min	8	24	45	4	24	40
2.6 GHz 10 min	13	21	47	6	24	32
5.7 GHz 5 min	12	15	49	5	19	17
5.7 GHz 10 min	12	19	57	4	21	34
9.3 GHz 5 min	4	17	25	5	28	17
9.3 GHz 10 min	9	24	44	0	11	25
LSD ⁰⁵	4.1	5.4	19.3	1.6	3.7	22.6
Sd	1.96	2.58	9.33	0.75	1.44	10.72
<i>F-act.</i>	10.79**	3.41*	2.21	17.33**	18.06**	1.42

* – $P < 0.05$, ** – $P < 0.01$; Sd – standard deviation, *F-act.* – interaction of treatment and tested parameter

Table 5. The effect of strong microwave electric field (SMEF) irradiation at 2.6, 5.7 and 9.3 GHz band for 5 and 10 min expositions on the frequency of occurrence (%) of *Fusarium* species on winter wheat grain (cv. 'Kovas' and 'Mulan')

Treatment	Frequency of occurrence %				
	'Kovas'		'Mulan'		
	<i>F. sporotrichioides</i>	<i>F. culmorum</i>	<i>F. poae</i>	<i>F. avenaceum</i>	other <i>Fusarium</i> spp.
Untreated	22.2	44.4	11.1	–	22.2
2.6 GHz 5 min	46.7	13.3	13.3	6.7	20.0
2.6 GHz 10 min	13.3	60.0	6.7	–	20.0
5.7 GHz 5 min	46.7	33.3	–	6.7	13.4
5.7 GHz 10 min	13.3	40.0	6.7	–	40.0
9.3 GHz 5 min	50.0	25.0	–	6.2	18.8
9.3 GHz 10 min	26.7	6.7	13.3	6.7	46.6
Untreated	–	20.0	6.7	13.3	33.3
2.6 GHz 5 min	3.0	46.7	6.7	–	26.7
2.6 GHz 10 min	30.0	20.0	30.0	–	30.0
5.7 GHz 5 min	16.7	41.7	8.3	–	33.3
5.7 GHz 10 min	33.3	8.3	16.7	–	41.6
9.3 GHz 5 min	–	16.7	33.3	–	50.0
9.3 GHz 10 min	55.6	22.2	–	11.1	11.1

other seed borne pathogenic fungi. The research into microwave technology for seed irradiation against seed borne phytopathogenic fungi done in different countries is promising since this technology is a non-polluting, efficient pre-sowing treatment of seeds. The study done by Vas'ko et al. (2004) in Belarus indicated that application of microwave energy allowed successful control of seed phytopathogenic infection, improved sowing qualities of the processed seeds promoting an intensification of growth processes. Under super high frequency irradiation, a considerable reduction of *Fusarium* spp. on the seeds of spring wheat and perennial forage grasses was achieved. Research done in Canada by Reddy et al. (1998) proved that microwave irradiation at 750 W with the variable power at 2.45 GHz can be used to significantly reduce infection by seed borne *F. graminearum* in wheat. The percentage of seed infection could be reduced to below 7% (from 36% for the control), without reducing the seed quality below the commercial threshold of 85% seed germination (Reddy et al., 1998).

Conclusions

1. Seed irradiation with a strong microwave electric field (SMEF) at 2.6, 5.7 and 9.3 GHz band for 5 and 10-minute exposures did not have any effect on winter wheat cv. 'Kovas' germinating power and germination; however, an increase in the quantity of abnormally germinated seeds and reduction in germination index (GI) and seedling vigour index (SVI) were observed. The negative effect of SMEF on GI and SVI increased with increased grain moisture before treatment ($P < 0.01$).

2. Treatment of seeds, artificially inoculated with *Tilletia caries*, in a SMEF showed a trend towards reduction of bunt infected ears. Seed treatment by SMEF stimulated plant productivity and the band 2.6 GHz for 20 min, 5.7 GHz for 10 min and 9.3 GHz for 5 and 10 min received a good response with an increase in grain number and weight per ear ($P < 0.05$).

3. Winter and spring wheat seed treatment with SMEF against infestation with *Phaeosphaeria nodorum*, *Pyrenophora tritici-repentis* and *Fusarium* spp. gave conflicting and inconsistent efficacy results. The composition of most frequently isolated *Fusarium* species on SMEF-treated winter wheat seeds was pertained to SMEF bands and exposition time.

Acknowledgements

The present study was supported by grant No. MIP-040/2011 provided by the Research Council of Lithuania.

Received 22 10 2012

Accepted 25 01 2013

References

- Aladjadjyan A. 2007. The use of Physical methods for plant growing stimulation in Bulgaria. Journal of Central European Agriculture, 8 (3): 369–380
- Aladjadjyan A. 2010. Effect of microwave irradiation on seeds of lentils (*Lens culinaris* Med.). Romanian Journal of Biophysics, 20 (3): 213–221
- Ayrapetyan S. N. 2004. Molecular and cellular mechanisms of possible non-thermal biological effect of extremely high-power microwave pulses (EHPP). Project Technical Report of ISTC A-803P. <http://www.dtic.mil/cgi-bin/GetTRDoc?Location=U2&doc=GetTRDoc.pdf&AD=ADA428135>
- Borgen A. 2004. Strategies for regulation of seed borne diseases in organic farming. Seed Testing International (ISTA News Bulletin), 127: 19–21
- Borgen A., Krebs N., Langkjær C. 2005. Novel development of heat treatment techniques for seed surface sterilization. 5th SHC Seed Health Symposium. Angers, France, p. 28
- Cao D., Hu J., Huang X., Wang X., Guan Y., Wang Z. 2008. Relationships between changes of kernel nutritive components and seed vigor during development stages of F1 seeds of sh2 sweet corn. Journal of Zhejiang University: Science B, 12: 964–968 <http://dx.doi.org/10.1631/jzus.B0820084>
- Dabkevičius Z., Kreimeris J. 1994. The comparison of winter wheat seed treating measures before sowing. Plant protection: a collection of research articles, 75: 23–33 (in Lithuanian)

- Dabkevičius Z., Sirvydas P. A., Sinkevičienė J., Šaluchaitė A., Vasiliauskaitė I. 2008. The effect of physical factors and chemical seed treatment means on spring barley diseases. *Žemės ūkio mokslai*, 15 (4): 28–34 (in Lithuanian)
- Farahani S. M., Chaichi M. R. 2012. Effect of field treatments on barley seed sensitivity to organic seed sanitation (hot water). *Research Journal of Seed Science*, 5 (3): 96–105 <http://dx.doi.org/10.3923/rjss.2012.96.105>
- Hankin L., Sands D. C. 1977. Microwave treatment of tobacco seed to eliminate bacteria on the seed surface. *Phytopathology*, 67: 794–795 <http://dx.doi.org/10.1094/Phyto-67-794>
- Joelsson G. 1983. The osmotic method – a method for rapid determination of seed borne fungi. 20th International Seed Testing Association (ISTA) Congress. Ottawa, Canada, p. 5
- Lammerts van Bueren E. T., Struik P. C., Jakobsen E. 2003. Organic propagation and planting material: an overview of problems and challenges for research. *NJAS-Wageningen Journal of Life Sciences*, 51: 263–277 [http://dx.doi.org/10.1016/S1573-5214\(03\)80019-2](http://dx.doi.org/10.1016/S1573-5214(03)80019-2)
- Leslie J. F., Summerell B. A. 2006. The *Fusarium* laboratory manual. Ames, USA, 388 p. <http://dx.doi.org/10.1002/9780470278376>
- Manandhar J. B., Cunfer B. M. 1991. An improved selective medium for the assay of *Septoria nodorum* from wheat seed. *Phytopathology*, 81: 771–773 <http://dx.doi.org/10.1094/Phyto-81-771>
- McNeil M., Roberts A. M. I., Cockerell V., Mulholand V. 2004. Real-time PCR assay for quantification of *Tilletia caries* contamination of UK wheat seed. *Plant Pathology*, 53: 741–750 <http://dx.doi.org/10.1111/j.1365-3059.2004.01094.x>
- Nielsen B. J., Borgen A., Kristensen L. 2000. Control of seed borne diseases in production of organic cereals. BCPC conference Pest and Diseases: proceedings of an international conference. Brighton, UK, 1: 171–176
- Nikulina R. N., Kovalev I. A., Chang L. H. 2009. The effect of low-intensity microwave radiation on germinating and growth intensity of wheat grains. 19th international Crimean conference Microwave and Telecommunication Technology. Sevastopol, Russia, p. 887–888
- Observation pests and disease of agricultural crops. 2002. *Plant Protection. Review* 2001, p. 139–149
- Požėlienė A., Lynikienė S., Sinkevičienė J., Dabkevičius Z. 2005. Influence of physical factors on seed impurity by micromicets. *Žemės ūkio inžinerija*, 37 (1): 60–68 (in Lithuanian)
- Požėlienė A., Lynikienė S. 2006. Establishment of effective electromagnetic origin factors, which reduces seed impurity, caused by microbiote. *Žemės ūkio inžinerija*, 38 (1): 105–117 (in Lithuanian)
- Reddy M. V. B., Raghavan G. S. V., Kushalappa A. C., Paulitz T. C. 1998. Effect of microwave treatment on quality of wheat seeds infected with *Fusarium graminearum*. *Journal of Agricultural Engineering Research*, 71 (2): 113–117 <http://dx.doi.org/10.1006/jaer.1998.0305>
- Seaman W. L., Wallen V. R. 1967. Effect of exposure to radio-frequency electric fields on seed borne microorganisms. *Canadian Journal of Plant Science*, 47 (1): 39–49 <http://dx.doi.org/10.4141/cjps67-006>
- Soriano-Martin M. L., Porras-Piedra A., Porras-Soriano A. 2006. Use of microwaves in the prevention of *Fusarium oxysporum* f. sp. *melonis* infection during the commercial production of melon plantlets. *Crop Protection*, 25 (1): 52–57 <http://dx.doi.org/10.1016/j.cropro.2005.03.016>
- Study on quarantine treatments against *Tilletia caries* (DC.) Tul. (TCT). Posted by Agricultural science on 2012. <<http://www.agrpaper.com/study-on-quarantine-treatments-against-tilletia-carries-dc-tul>> [accessed 01 08 2012]
- Tarakanovas P., Raudonius S. 2003. Statistical analysis of agronomic research data using software ANOVA, STAT from the package SELEKCIJA and IRRISTAT. Lithuanian University of Agriculture (in Lithuanian)
- Tylkowska K., Turek M., Blanco Prieto R. 2010. Health germination and vigour of common bean seeds in relation to microwave irradiation. *Phytopathologia*, 55: 5–12
- Vanova M., Matysinsky P., Benada J. 2006. Survey of incidence of bunts (*Tilletia caries* and *Tilletia contraversa*) in the Czech Republic and susceptibility of winter wheat cultivars. *Plant Protection Science*, 42: 21–25
- Vas'ko P., Ermolovich A., Karpovich V., Mikhalenko E. 2004. Low intensive influence effect of the electromagnetic waves on the seed germination of winter crops, spring corn and forage grasses. 5th international symposium Physics and Engineering of Microwaves, Millimeter, and Submillimeter Waves. Kharkov, Ukraine, 2: 832–834

ISSN 1392-3196 / e-ISSN 2335-8947

Zemdirbyste-Agriculture, vol. 100, No. 2 (2013), p. 185–190

DOI 10.13080/z-a.2013.100.024

Didelės galios mikrobangų impulsų poveikis: (2) paprastojo kviečio (*Triticum aestivum* L.) sėklų daigumui ir sanavimui

I. Gaurilčikienė¹, J. Ramanauskienė¹, M. Dagys², R. Simniškis², Z. Dabkevičius¹, S. Supronienė¹

¹Lietuvos agrarinių ir miškų mokslų centro Žemdirbystės institutas

²Fizinių ir technologijos mokslų centras

Santrauka

Paprastojo kviečio (*Triticum aestivum* L.) sėklos buvo apšvitintos didelės galios mikrobangų impulsais, generuotais aukšto dažnio magnetrone. Skirtingo drėgnio (12, 15 ir 18 %) sėklos švitintos 5, 10 ir 20 min 2, 6, 5,7 ir 9,3 GHz elektromagnetinių bangų dažnių ruožuose. Tirta didelės galios mikrobangų impulsų įtaka sėklų daigumui, dygimo energijai, nenormaliai sudygsių sėklų kiekiui, daigumo indeksui ir daigų augimo indeksui. Taip pat tirtas jų poveikis *Tilletia caries* infekcijai, augalų produktyvumui ir sėklų užterštumui patogeniniais mikromicetais. Neapšvitintos sėklos naudotos kaip kontrolinis variantas.

Sėklų apšvitinimas didelės galios mikrobangų impulsais iš esmės neturėjo įtakos dygimo energijai ir sėklų daigumui, tačiau padaugėjo nenormaliai sudygsių sėklų, taip pat nustatytas ir daigumo bei daigų augimo indeksų sumažėjimas. Apšvitinus didesnio drėgnio sėklas, didėjo ir didelės galios mikrobangų impulsų neigiamas poveikis daigumo bei daigų augimo indeksams ($P < 0,01$). Apšvitinus dirbtinai *Tilletia caries* inokuliuotas žieminių kviečių sėklas, pastebėta kūlėtų varpų sumažėjimo tendencija. Didelės galios mikrobangų impulsais apdorotomis sėklomis sėtų augalų derlingumas padidėjo iš esmės, o 20 min apdorotų 2,6 GHz, 10 min – 5,7 GHz ir 5 bei 10 min – 9,3 GHz bangų ruožuose ir grūdų skaičius varpose bei vienos varpos grūdų svoris padidėjo iš esmės ($P < 0,05$). Švitinimo didelės galios mikrobangų impulsais poveikis su kviečių sėklomis pernešamai grybinei infekcijai (*Phaeosphaeria nodorum*, *Pyrenophora tritici-repentis* ir *Fusarium* spp.) buvo nepakankamo efektyvumo ir nenuoseklus. *Fusarium* genties dažniausiai aptiktų rūšių kompozicija ant apšvitintų sėklų kito priklausomai nuo didelės galios mikrobangų impulsų dažnių ir ekspozicijos trukmės.

Reikšminiai žodžiai: mikrobangos, sėklos daigumas, sėklos infekcija, *Triticum aestivum*.