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Effects of Ultrasonic Treatment on Germination of Synthetic Sunflower Seeds

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***Abstract*—**One problem of synthetic sunflower cultivation is an erratic germination of the seeds. To improve the germination, pre-sowing seed treatment with an ultrasound was tested. All treatments were carried out at 40 kHz frequency with the intensities of 40, 60, 80 and 100% of the ultrasonic generator total power (250 W) for the durations of 5, 10, 15 and 20 minutes. Data on seed germination percentage, seed vigor index (SVI), root and shoot lengths of seedlings were collected. The results showed that germination, SVI, root and shoot lengths of ultrasonic treated seedlings were different from the control, depending on intensity of the ultrasound. The effects of ultrasonic treatment were significant on germination, resulting in a maximum increase of 43% at 40 and 60% intensities compared to that of the control seeds. In addition, seedlings of these 2 treatments had higher SVI and longer root and shoot lengths than that of the control seedlings. All treatment durations resulted in higher germination and SVI, longer root and higher shoot lenghts of seedlings than the control. Among the duration treatments, only SVI and seedling root length were significantly different.

***Keywords*—**Ultrasonic, germination, root length, shoot length, *Helianthus annuus* L.

I. Introduction

IN sunflower, using high quality seeds is a prerequisite of obtaining a high germination percentage. Synthetic sunflower seeds often lack of uniform germination and seedling growth contributing to poor plant growth at the later stage. In addition, the seeds are weak and do not germinate under a long storage condition. Several methods have been used for seed pre-sowing treatment in order to increase the germination and uniformity i.e. gibberellic acid [1], [2], sulphuric acid [2] and hot water treatment [3], [4]. However, seeds treated by these methods have to be rapidly grown and can not be storaged. Therefore, the methods that increase seed germination but do not affect their viability during the storage are much needed. Applications of ultrasound have been used to induce faster and greater seed germination of the Norway spruce [5], barley [6], orchids [7] and other crops [8]. It has been reported that the optimum condition of treated seeds lead to uniform and rapid germination.

In general, the ultrasonic wave acts as an alternative stress on cells or tissues and change the structure and function of biological molecules. The stimulating action of ultrasounds on the germination is produced by modifications of the cellulose membrane resulting in better nutrient's transportation and absorption of the useful elements around seeds [5]. Ultrasounds may cause stimulations or destructions, depending on plant genotypes and ultrasound parameters i.e. intensity, frequency and duration. Ultrasounds with frequencies between 1–42 kHz, intensities between 20 to 100 % and durations between 3 seconds to 60 minutes, were used depending on species. Recent studies have been shown that ultrasound with proper intensity and duration would increase the enzyme activities or promote the cell growth by stimulating physiological activities of cells compared to growth without ultrasound [9], [10], [11]. Goussous et al. [8] applied ultrasound at 40 kHz for duration of 5-60 minutes to enhance germination of vegetable seeds. They reported that the highest growth rate index of treated seeds was found at 45 minutes. Rista et al. [5] used ultrasound to promote germination of Norway spruce seeds. They reported increases of 40% in germination, 32% in root length and 5-8% in shoot length for the treatment duration of 50 and 60 seconds. Shin et al. [7] applied ultrasonic treatment at 42 kHz frequency of 135 W for 3-10 minutes. They found that seed germination and protocorm formation increased with the increasing ultrasonic duration (up to 10 min) without destroying embryos. Different researchers reported that the activity of enzymes increased under mild ultrasound, and in some case high intensities of ultrasound caused a decrease in the activity of alpha-amylase activity [6], [12]. According to those experiments, the proper intensity and duration of ultrasound are important and should be selected carefully, otherwise it would destroy the cell structures or deactivate the enzymes. For different species, the optimum ultrasonic intensity and duration appeared varied widely between species and cultivars. The aim of this experiment was to find the optimal duration and intensities of ultrasonic treatment that can stimulate a uniform and high germination of synthetic sunflower seeds.

II. Materials and Methods

A. Materials and Data Collection

Synthetic sunflower (variety S473) seeds were obtained from the SUT Sunflower Project on April 2012. The sample seeds were ultrasonic treated at a fixed frequency of 40 kHz using four different ultrasonic intensities (40, 60, 80 and 100% of the generator power setting) at the durtion of 5, 10, 15 and 20 minutes in a constant temperature (30 °C). All treatments were arranged in factorial experiment in a completely randomized design with three replications of 50 seeds per replication. The untreated (control) and treated seeds were

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germinated in towel paper and incubated in a growth chamber at 25˚C, 95% humidity with a 12 hours photoperiod. Germinated seeds were counted at 7 and 10 days after the incubation. After counting, 20 normal seedlings were randomly selected and measured for the root and shoot lengths. The seedling vigor index (SVI) was calculated following the ISTA [13] protocol.

B. Data Analysis

The data were analyzed by statistical package SPSS V.14 [14] software. Analysis of variance (ANOVA) was conducted and significant differences among treatment means were tested using Duncan's Multiple Range Test.

III. Results and Discussion

The effects of ultrasonic stimulation on the germination, SVI, root and shoot lengths of synthetic sunflower seeds were investigated. The results were as shown in Table I, Fig. 1 and 2. The ultrasonic intensities appeared to significantly affect the germination, SVI and root length, while the treatment durations affected only the SVI and root length.

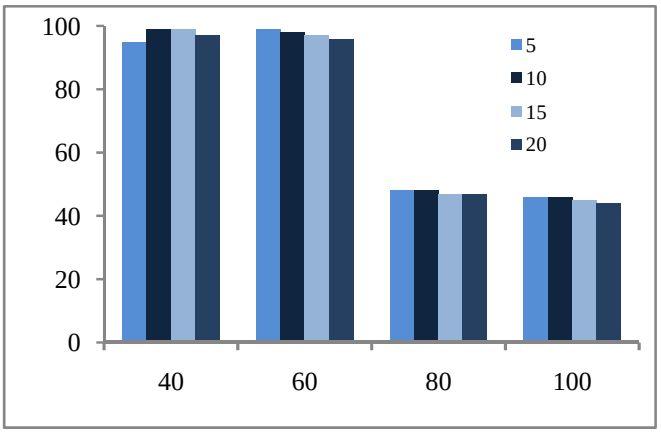

Fig. 1 Germination of synthetic sunflower seeds treated with different ultrasonic intensities for 5, 10, 15 and 20 minutes

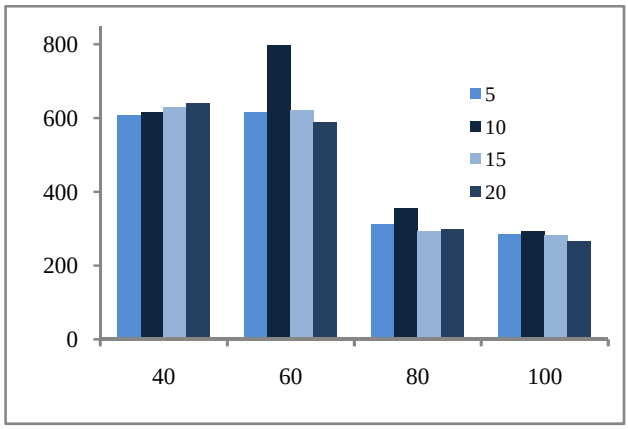

Fig. 2 Seed vigor index of synthetic sunflower seeds treated with different ultrasonic intensities for 5, 10, 15 and 20 minutes

TABLE I
Effects of Ultrasonic Intensity and Exposure Time on Germination, SVI, Root and Shoot Lengths of Synthetic Sunflower Seeds

Treatment	Germination (%)	Increase (%)	Seed vigor index	Root length (mm)	Shoot length (mm)
Intensity (%)					
40	98 a	43.38	623 a	12.60 a	6.40
60	98 a	43.38	656 a	12.35 b	6.72
80	48 b	-	316 b	11.72 b	6.64
100	45 b	-	282 b	11.56 b	6.23
Exposure time (min)					
5	72	5.88	455 b	13.11 a	6.33
10	73	7.35	516 a	12.67 ab	6.29
15	73	6.99	457 b	12.39 b	6.76
20	73	6.99	448 b	12.53 ab	6.99
control	68	-	273	6.56	4.02

*Means within each column followed by different letters are significantly different based on DMRT at 5% level

Germination: Analysis of variance revealed significant differences among treatment intensities on germination. Means of the germination percentage are presented in Table I. The results showed that seed germination percentage increased with the increasing intensity. However, increasing intensity more than 60% tended to decrease seed germination. The efficacy of ultrasonic treatment on sunflower seed stimulation was demonstrated at the intensity levels between 40 and 60% of the output power with the exposure times between 5 to 20 minutes. As can be seen from Table I that the highest germination percentages of sunflower seeds were 95 and 99% when treated with ultrasound at 40 and 60% intensity for 5-20 minutes compare to that of the control which was only 68 %. Similar results were obtained in orchid seeds with ultrasonic pre-sowing treatment [7]. However, in this study, the germination percentages were 44-48% lower than that of the control when treated with ultrasonic at 80-100%. Prolonged treatments of ultrasonic at higher intensity possibly impose injury on the embryo. The results of this study revealed that ultrasonic treatment duration (5-20 minutes) resulted in higher germination than the control but had little effects on seed germination.

Seed vigor index (SVI): Seeds of higher SVI are considered to be more vigorous. As can be seen from Fig. 2, it is obvious that increasing ultrasonic intensity increased SVI. The SVI of treated seeds were higher than that of the control seeds, whereas the SVI at 40 and 60% intensities were higher than those of SVI at 80 and 100% intensities. The duration of seed exposure to ultrasound resulted in SVI variations. The highest SVI of seedling was found at 10 minutes of ultrasonic treatment.

Root and Shoot Lengths: The variation of duration of seed exposure to ultrasound has resulted in the variation in the root and shoot lengths. From the data, mean root length of the treated seedlings was 11.56-13.11 mm, compared to 6.56 mm of the control. A similar effect was observed in shoot lengths of the treated seedlings which were 6.23-6.99 mm compared to 4.02 mm of the control. At the intensities of 40- 60% and durations of 5-20 minutes, the treatments produced maximum root and shoot lengths.

From the results, pre-sown ultrasonic treatment

considerably increased the germination percentage and seedling growth of the synthetic sunflower seeds (Table I, Fig. 1 and 2). Yaldagard et al. [6] reported that ultrasonic treatment could fragment the seed shell and produce larger porosity on the surface of barley grains by captivation of ultrasound (shock waves). Shell fragmentation and enlargement of the pore size of seeds lead to more water retention capacity in dry grains and result in better hydration. Moreover it is possible that endosperm modification, including starch degradation by means of ultrasound, may lead to increase in the rate of enzyme-catalyzed hydrolysis reactions within the seeds. More hydrolysis enzyme activity may consequently lead to quicker germination and faster embryo growth after the sonication. However, only ultrasound with proper intensity and duration would increase the enzyme activities or promote the cell growth by stimulating physiological activities of cells, while at high intensity there will be more damage to the cells or enzyme structure [9], [10], [11], [12]. In this study, the optimal ultrasonic intensities were between 40-60%, while the ultrasonic intensity more than 60% resulted in the reduction in all parameters recorded.

IV. Conclusion

It can be concluded that the use of ultrasound to stimulate germination of synthetic sunflower seeds is feasible. All recorded parameters positively responded to ultrasonic treatments at appropriate intensity and exposure duration. The optimal conditions for ultrasonic treatment at 40 kHz frequency for synthetic sunflower seeds were established at the intensities of 40 to 60% with the exposure durations of 5-10 minutes. However, this experiment was only preliminary, more varieties and seed storing duration after pretreatment should be further investigated in order to establish regularities with practical applicability.

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