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## Süs Bitkilerinin Tohum Çimlenmesi, Fide Büyümesi ve Gelişiminde Kullanılan Ultrasonik Ses Uygulamaları

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**ÖZET:** Ultrasonik ses, insan kulağının duyamayacağı (20-100 kHz) frekanslarda üretilen, maddelerle etkileşime giren akustik dalgalar, tarım endüstrisinde yaygın olarak kullanılmaktadır. Son yıllarda ultrasonik ses, tohumlarla ilgili literatürde bildirilen birçok örnekle çimlenmeyi teşvik eden bir teknoloji olarak büyük ilgi görmüştür. Bu derlemede ses ve mekanizması, ultrasonik ses uygulamalarının tohum ve bitki büyüme ve gelişimine etkileri kısaca sunulmuştur. Derlemenin temel amacı, süs bitkisi türlerinin tohum çimlenmesi üzerine ultrasonik ses uygulamalarının etkilerini detaylı olarak incelemek ve süs bitkilerinde ultrasonik ses uygulamalarının kullanım ve potansiyelini ortaya koymaktır. Ultra ses dalgası teknolojisi uzun bir geçmişe sahip olmasına rağmen, kullanılan teknolojinin sürekli gelişimi, modifikasyonu ve genişlemesi ile güncelliğini korumaktadır. Bu derleme, süs bitkisi türlerinin üretimine bu güncel teknolojinin dahil edilmesine dikkat çekilmesine katkıda bulunacaktır.

**Anahtar Kelimeler:** Çimlenmenin iyileştirilmesi, süs bitkisi, çiçekçilik, tohum çimlenmesi, ultrasonik ses

## Ultra-Sonic Sound Applications Used in Seed Viability, Seedling Growth and Plant Development of Ornamentals

**ABSTRACT:** Ultra-sonic sound, acoustic waves generated from frequencies in the ranges (20-100 kHz) that cannot be heard by the human ear, which interact with substances, are extensively used in agricultural industry. In recent years, ultra-sonic sound has gained great attention as a technology to stimulate germination with many examples reported in literature on seeds. In this review, sound and its mechanism, the effects of ultra-sonic sound applications on seed and plant growth and development are briefly presented. The main purpose of the review is to examine the effects of ultra-sonic sound applications on seed germination of ornamental plant species in detail and to present the use and potential of ultra-sonic sound applications in ornamental plants. Although ultra sound wave technology has a long history, it remains up-to-date with the continuous development, modification and expansion of the technology used. This review would help to contribute drawing attention to the inclusion of this current technology in the production of ornamental plant species.

**Keywords:** Improving germination, ornamental plant, floricultural, seed germination, ultrasound

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## INTRODUCTION

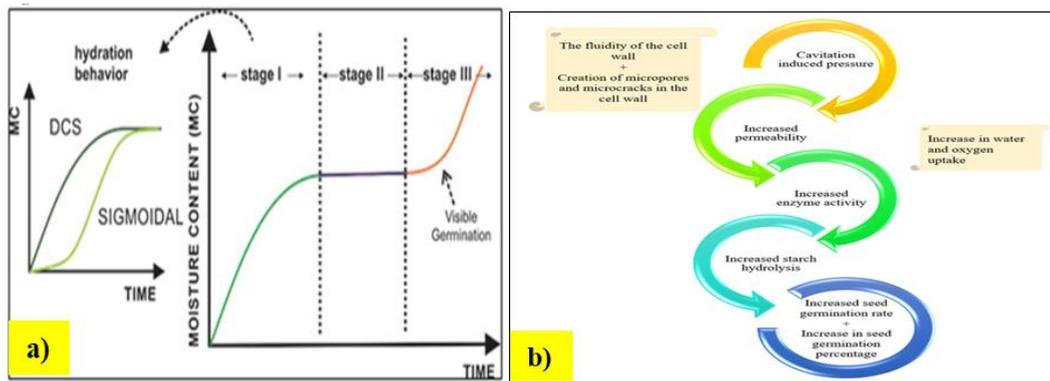
Seed is the basis for annual and biennial ornamental plants and an important starting material for the propagation of these plants. Germination of the seed is an event controlled by many biotic and abiotic factors. The seed must first emerge from embryonal dormancy and be in an environment with the optimum temperature and humidity required for germination in order to germinate the seed. In addition, the seed coat must allow water and gas (oxygen and carbon dioxide) permeability for germination.

In plant production, it is desired to complete the germination of the seeds sown in a short time and to complete the seedling emergence in a homogeneously. The seed used must be of high quality order for this request to be realized in the best way. The term seed quality describing the potential performance of a seed lot means seed in high genetic and physical purity, vigor, high yield potential, disease and pest infestation free, weed-free, not aged, undamaged, with optimum moisture content (usually 8-12%), high germination percentage (>80%), fast and uniform germination traits (Memiş, 2020). In general, seed quality is affected by many factors that occur before harvest, during harvest and after harvest during drying, processing and storage (Kibinza et al., 2006).

In order to improve seed quality, seed coating applications, germination promoting chemical applications, pre-germination (osmotic conditioning, hydropriming, priming, etc.) applications, which are called post-harvest applications, are widely used today. One of the applications for improving germination and eliminating dormancy in seeds (Miano et al., 2015) is Ultra-sonic sound applications. Successful results have been obtained from the studies on the effect of ultra-sonic sound applications on seed germination and plant growth. It has been reported that Ultra-sonic sound application induces seed germination (Creath and Schwartz 2004; Wang et al., 2012; Miano et al., 2015; Shekari et al., 2015; Liu et al., 2016; López-Ribera and Vicient, 2017). It has been stated that Ultra-sonic sound vibrations have effects such as washing the germination inhibitory substances in the seed coat, increasing in the embryonic development of the vibrations, and thinning the impermeable layer on the outer part of the seed coat (Gaba et al., 2008). The micro-holes opened from the seed coat can eliminate the negative effects caused by the seed coat and thus seed germination can be improved by increasing in the oxygen uptake and the contact of water with the embryo during the application process (Baker et al., 2001; Moussatov et al., 2005; Yaldagard et al., 2008). It also increases in germination percentage, root and stem length of seedlings, and seedling emergence index. It promotes enzyme activity and initiation of other biological processes (Baker et al., 2001; Teixeira Da Silva and Dobránszki, 2014; Shekari et al., 2015). In addition, this technique provides seed application by modifying the seed coat without damaging the seed, with natural drying allowing storage after application (Porto et al., 2018).

The mechanism of action of ultra-sonic sound waves on the seed is shown in Figure 1.b. Ultra-sonic sound applications provide the water and oxygen required for seed germination. Cavitation works by creating micro bubbles in water. It is led to the formation of the cell wall micro-pores, micro-cracks and cell wall fluidity by applying mechanical pressure on the seed. This increases in water and oxygen uptake (Figure 1.a). Enzyme activity increases as the seeds absorb water (Baker et al., 2001; Moussatov et al., 2005; Yaldagard et al., 2008; Wu et al., 2013).

Ultra-sonic sound water bath applications are short-term applications and seeds are applied for a short time with a simple Ultra-sonic sound water bath and can be used for sowing immediately after application. Ultra-sonic sound applications in seeds are advantageous as they are low cost, water saving, low energy use, easy and safe (Nazari and Eteghadipour 2017; Memiş, 2020).



**Figure 1. a);** Stages of the seeds germination as function of the moisture content. Adapted from Bewley and Black (1978) and Ibarz and Augusto (2015); DCS: the downward concave shape (Miano et al., 2016). **b);** the mechanism of action of ultra-sonic sound waves on the seed (Memiş, 2020)

The effects of ultra-sonic sound applications on organisms have been discussed in detail in terms of the preservation of agricultural products and plant growth, and the positive effects of the frequency and intensity of the sound wave on the products have been evaluated in various studies (Awad et al., 2012; Bilek and Turantaş, 2013; Hassanien et al., 2014; Teixeira Da Silva and Dobránszki, 2014; Dikilitaş et al., 2016; Leon et al., 2018). While some studies have focused on the positive effects of frequency and power on organisms, some of studies have examined the negative (Yiyao et al., 2002; Chivukula and Ramaswamy, 2014) effects of sound waves. It has been reported that when sound waves are applied to various plants, they contribute to the increase in their aromatic components (Shao et al., 2008; Li et al., 2008) and to the change of hormone levels in tissues (Bochu et al., 2001). Ultra-sonic sound applications have also been found to be effective in increasing in resistance to diseases (Zhang, 2012) and pests (Hou et al., 2009) and thus reducing the need for chemical fertilizers and biocides (Zhang, 2012; Carlson 2013; Hassanien et al. 2014; Chen et al., 2016).

In recent years, there have been many examples reported in the literature of seeds exposed to high-energy ultra-sonic sound using an ultrasonic wave water bath. Moreover, Ultra-sonic sound wave applications have also attracted attention as a technology to promote plant germination (Wang et al., 2012; Ghafoor et al., 2014; Miano et al., 2015; Tabaru et al., 2015; Yang et al., 2015; López-Ribera and Vicient, 2017). In previous studies, the application of Ultra-sonic sound has been investigated to promote germination in carrot, radish, okra, squash, tomato, corn, barley, soybean, bean, chickpea, wheat, pepper, watermelon, rice and sunflower seed species (Miyoshi and Mii, 1988; Hebling and da Silva, 1995; Shimomura, 1998; Shors et al., 1999; Carbonell et al., 2000; Aladjadjian, 2002; Florez ' et al., 2007; Yaldagard et al., 2008). The results of these studies have determined that the effects of ultra-sonic sound application on seed germination depend on the frequency, exposure time, distance and density from the organism, the physiological state of the environment, the temperature of the environment, and show great differences between different species and cultivars.

The literature on the use of ultra-sonic sound to enhance seed germination of in horticultural crops, is scarce. The literature on the use of ultra-sonic sound to enhance seed germination of in vegetable crops, is rather scarce. A study was conducted to determine its effectiveness on germination rate and time of different exposure times (10, 20, 30 minutes) of 50 kHz sound wave at 25 C on seed lots of 10 varieties of 8 species (watermelon, cucumber, melon, carrot, leek, tomato, pepper, eggplant) (Memiş 2020). The effects of the applications varied according to the time and plant species. Dönmez (2018) reported that ultrasound applications can be used as a pre-germination application in spinach seeds. Processing of spinach with agri-wave technology stimulated growth rate and increased yield (22.7%)

(Hou and Mooneyham, 1999). It has been reported that the sound wave-mediated growth of the mushroom mycelium increased by 15% and accelerated the fruiting of the edible mushroom (Jiang et al., 2011). The effects of the applications on both the germination rate and seedling emergence rate were positive (Memiş 2020).

The literature on the use of ultra-sonic sound on seed germination of ornamental plants is much more limited. Ultra-sonic sound applications on some ornamental plant species were performed in vitro and their effects on root growth and callus development in general were investigated [(*Gerbera jamesonii*) (Wang et al. 2003), (*Aloe arborescens* Mill.) (Liu et al. 2003), (*Dendrobium officinale*) (Wei et al. 2012), (*Dahlia x pinnata*) (Otani et al. 2013)].

In this review, it is aimed to examine the effects of Ultra-sonic sound applications on seed germination of ornamental plant species in detail and to present the use and potential of Ultra-sonic sound applications in ornamental plants. Although ultra sound wave technology has a long history, it remains up-to-date with the continuous development, modification and expansion of the technology used. This review would help to strengthen our knowledge to apply Ultra-sonic sound in horticultural practice and contribute to the development and drawing attention to the inclusion of this current technology in the production of ornamental plant species.

### Sound Wave and Mechanism of Action

Mechanical waves are physical factors that we are commonly exposed to as sound, light and water waves in our environment. A sound wave is a mechanical vibration that oscillates in frequency within the audible range of the ear (Middlebrooks and Green 1991). The unit of sound is measured in decibels and is a logarithmic unit. The lowest sound that can be heard with normal hearing is zero decibels (0 dB), and a person with good hearing can even hear a weaker sound level of minus five (-5) dB (Takeuchi et al., 2014; Mohanta, 2018).

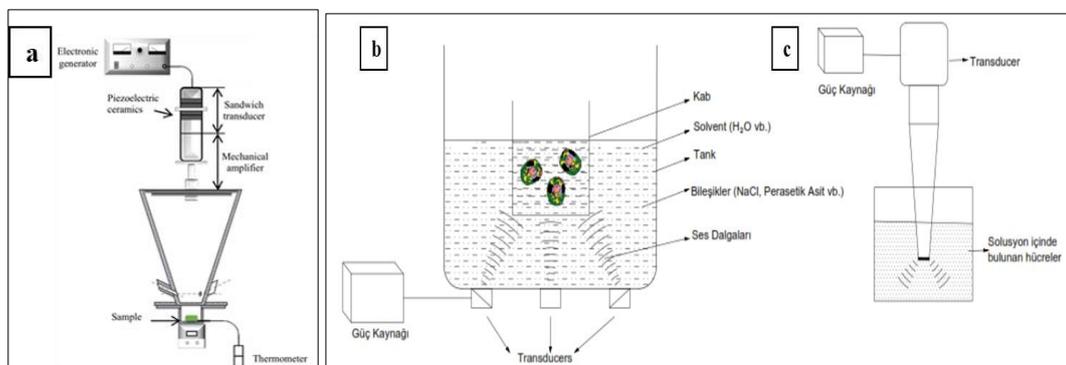
Sound waves are examined in three main spectra: “infrasound” with a frequency (vibration) less than 20 Hz, “audible sound” between 20 Hz and 20 kHz and “Ultra-sonic sound” with a frequency greater than 20 kHz. Ultra-sonic sound is a sound wave that includes frequencies that cannot be heard by the human ear, such as infrasound (Dolatowski et al., 2007; Dikilitaş et al., 2018). The human hearing limit is 15-20 kHz, and the frequency of ultra sound waves is above 50 kHz (Knorr et al., 2004). Accordingly, Ultra-sonic sound applications are divided into two as low energy and high energy (Kentish and Ashokkumar 2011). The low energy group has a frequency between 100-1000 kHz and a sound intensity lower than  $1 \text{ W m}^{-2}$ , the high energy group has a frequency between 20-100 kHz and a sound intensity higher than  $1 \text{ W m}^{-2}$ . Low-energy sound waves are waves that do not change the physical and chemical structure of the material they pass through. They do not have a destructing effect on foods, they are used to illuminate the structure and physicochemistry of foods (Leadley and Williams, 2006). They are also used for imaging purposes in the medical and industrial areas (Kentish and Ashokkumar, 2011). High-energy Ultra-sonic sound waves, on the other hand, not only increase in physical deterioration by leaving mechanical and chemical effects on living and non-living organisms, but also increase in chemical reactions (Golmohamadi et al., 2013). High-energy Ultra-sonic sound waves are the most widely used sound waves in biological studie (Piyasena et al., 2003).

When the ultra-sonic sound wave is applied to liquids (sonication), it creates waves deep in the liquid, and when these waves hit a solid object, they create a compression and expansion zone around them, creating a structure called a vortex or cavitation. These structures, called cavitation, are formed as a result of the formation, growth and collapse of air bubbles. Cavitation in the liquid is divided into stable and non-stable. Stable cavitation is achieved by low-intensity ultra-sonic sound waves, and non-

stable cavitation is achieved by high-intensity ultra-sonic sound waves. The gas formed in micro-air bubbles creates a shock wave with high temperature and pressure by exploding. During the explosion, a heat of 50-70 °C is released as theoretical and a pressure of 100 MPa, that is, 1000 atm, is formed, which allows the formation of free radicals in the liquid inside and outside the cell (Fellows, 2000). This situation, which occurs in a chain manner, creates negative pressure and facilitates the inactivation of microorganisms (Valero et al., 2007). These bubbles are very successful in breaking off particles on foods such as fruits and vegetables and disrupting their membrane structure. As a mechanism, the ultra-sonic sound wave also has the power to break up the film layer formed by microorganisms. Apart from the physical mechanism mentioned above, the most important mechanism is the chemical mechanism of action. This mechanism is explained by thinning of cell membranes, local heating and formation of free radicals (Fellows, 2000; Kadkhodae and Povey, 2008; Weiss et al., 2011). Again, hydrogen atoms by combining with hydroxyl radicals during cavitation generate H<sub>2</sub>O<sub>2</sub>; this formation causes an extra damage to the cell wall (Lee and Feng, 2011; Dikilitaş et al., 2016).

### Generation of Ultra-Sonic Sound Wave

To generate an Ultra-sonic sound wave, an electrical source, transducer and coupler/emitter are needed firstly (Mothibe et al., 2011). While the electrical source produces the energy required for the Ultra-sonic sound wave, the transducers convert the electrical energy into mechanical vibration at the desired frequency and create pressure (Bermudez-Aguirre et al., 2011). Coupler/emitter, also called reactor or ultra-sonic sound cell, is involved in transferring to the liquid medium from the transducer of the ultra-sonic sound wave (Leadley and Williams, 2006). Such devices are used for mixing and homogenization process. The mechanism of the sound wave and the method used in this area are presented in Figure 2.



**Figure 2.** (a) Scheme of the Airborne Acoustic Ultra-sonic sound generator (Porto et al., 2018); (b) Application of Ultra-sonic sound on cell suspension in a cap; (c) Application of soundwave via sonicator (modified from the work of Sao Jose et al., 2014) (Dikilitaş et al., 2016)

### Ultra-Sonic Sound Applications Used in Seed Germination, Seedling Growth and Development of Ornamental Plants

Ultra-sonic sound applications used in seed germination, seedling development, plant growth and development of ornamental plants were generally carried out in vitro conditions, and root growth and callus development were emphasized especially as growth parameters. The reviewed research results on the subject are presented below.

In an attempt to develop a suitable protocol for the asymbiotic germination of *Calanthe* hybrid seeds (“Hyesung”x“Jeongmong”, “Hwagung”x“Hesung”), modified medium supplemented with different concentrations of activated charcoal (0, 0.01, 0.1 g l<sup>-1</sup>), naphthaleneacetic acid (NAA) and 6-benzylaminopurine (BA) (0.1, 0.5, 1.0 mg l<sup>-1</sup>) were tested. In addition, the effects of ultra-sonic sound

pretreatment time (at 42 kHz frequency, 0 (control), 3, 5, 7 and 10 minutes) were also investigated. Ultra-sonic sound applications significantly increased in the number of germinated seeds at exposure times of up to 10 minutes (Shin et al., 2011).

Mature seeds of *Calanthe discolor* were sterilized with 1% sodium hypochlorite solution and sowed in liquid medium after ultra-sonic sound application. The seed coat was removed from almost all seeds after 4 minutes of sonication. However, prolonged administration of more than 8 minutes increased in the percentage of destroyed embryos. Seed germination rate increased significantly with sonication application. Less than 10% of the seeds were germinated in the control cultures while 60% of the seeds were germinated in the sonicated cultures (Miyoshi and Mii 1988).

*Pinus banksiana* Lamb., *P. resinosa* Ait., *Larix europaea* L. and *Picea glauca* (Moench) Voss seeds were subjected to 30 minutes to 1 MHz Ultra-sonic sound at one of three Ultra-sonic sound intensities ranging from 0.5 to 6.0 W cm<sup>-2</sup>. Only *Pinus banksiana* Lamb. responded to ultrasonic treatments with higher mean daily germination and germination values. Seeds were stimulated equally by all intensities used. Germination rates and percentage of other tree species were not affected by any of the sonication treatments. *Pinus banksiana* Lamb. seeds were also sonicated at 25, 50, 100, 250 and 750 kHz at an intensity in the range of 0.5–1.0 W cm<sup>-2</sup>. In addition, the highest seedling height and the highest number of seedlings were detected in *Pinus banksiana* Lamb. among plant species that were sonicated at 1 MHz (Weinberger and Burton (1981).

Due to the poor germination of *Picea abies* (L.) Karsten seeds (the germination rate is less than 50% in some years), it is aimed to stimulate the germination of seeds by exposing them to Ultra-sonic sound at certain doses. Ultra-sonic sound was applied to the collected seeds with piezoelectric generator at two different frequencies (0 kHz, 500 kHz and 1 MHz), generating a density of 1 W/cm<sup>2</sup>, at different exposure times (3, 6, 9, 12, 15, 18, 20, 30, 40, 50 and 60 seconds). Significant increases were determined in terms of germination values and root elongation. It has been determined that they generally have an inhibitory effect, especially at the 1 MHz frequency, at exposure times above 30 seconds. It was concluded that if optimal parameters (frequency, intensity and exposure times) were established, its use could be a useful tool for stimulating the germination of *Picea abies* (L.) Karsten seeds (Rîșca et al., 2007).

In the research in which the relationship between embryo development and seed germination of *Cypripedium formosanum* in vitro was investigated, ultrasonic application was performed for 15, 30, 45 and 60 minutes. Ultrasonic application for 30 minutes increased in germination when compared to control. However, ultrasonic applications for 45 and 60 minutes caused to the extension of the germination period. It was determined that the Ultra-sonic sound application applied to the seeds of *Cypripedium formosanum* for 30 minutes did not damage the seed coat; It was emphasized that this result may be an important factor in mechanical abrasion (Lee et al., 2005).

The effects of Ultra-sonic sound application on grass seed germination and seedling were investigated. 5, 15, 25, 35 minutes application time, 25, 35, 45, 55 °C application temperature and 200, 300, 400, 500 W ultrasonic power were used in the study. As a result, ultrasonic power had the greatest effect on seedling growth, while temperature was the most influential factor for germination. Ultra-sonic sound treatment at 39.7 °C with an ultrasonic power of 348 W for 22.5 minutes provided the greatest germination percentage and the best seedling growth. The electrical conductivity of the seed leaks during the ultra-sonic sound application was significantly higher than the control. It have showed that this ultra-sonic sound application had positive effects on the grass seed (Wang et al., 2012).

Pre-moistened *Achillea millefolium* seeds were subjected to ultra-sonic sound for 5 and 10 minutes. The highest seed germination rate was measured at 5 minutes application, and the highest seedling length

was measured at 5 and 10 minutes application times. The highest seedling dry weight was obtained from the 5 minutes application. Ultra-sonic sound for 5 minutes gave the best results in all criteria (Mirshekari et al., 2013).

The effects of media and seed pretreatments on seed germination and seedling growth of *Paphiopedilum armeniacum* S. C. orchid were investigated. The study was carried out at 40 kHz for 2, 4, 6, 8 and 10 minutes at room temperature. Ultra-sonic sound treated seeds were stained with 2,3,5-triphenyl tetrazolium chloride (TTC). The percentage of TTC staining and germination gradually were increased when ultra-sonic sound was applied to the seeds for 2 to 8 minutes. When the ultrasonication time exceeded 10 minutes, the percentage of TTC staining and germination were decreased. However, the highest TTC staining (19.7%) and germination percentage (25.4%) were observed after 8 minutes of Ultra-sonic sound (Zhang et al., 2015).

In another similar study, the effect of ultra-sonic sound wave application on TTC staining on in vitro *Paphiopedilum* SCBG Red Jewel seed germination was investigated and it was determined whether these priming applications could promote seed germination. Ultra-sonic sound application was carried out at 40 kHz frequency at room temperature for 2, 3, 4, 6, 8 and 10 minutes. The germination percentage increased when ultra-sonic sound was applied to the seeds for 2-8 minutes. The highest TTC staining (18.3% and 20.0%) and germination percentage (53.1% and 54.03%) were seen in 6 and 8 minute ultra-sonic sound applications. Prolonged Ultra-sonic sound (10 minutes) caused adverse effects (Jiang et al., 2016).

Ultra-sonic sound was effective in revitalizing aged seeds by increasing in superoxide dismutase (SOD) and peroxidase (POD) activities and decreasing malondialdehyde (MDA) content. In addition, Liu et al. (2018) have reported where ultra-sonic sound temperature is the most important factor for germination of aged seeds.

Yi et al. (2003a) have determined that the sound wave (1 kHz, 100 dB) accelerated their development, and increased in the activity of soluble sugar, protein and amylase by stimulating the roots of chrysanthemum plants. The chrysanthemum seedling cuttings were irradiated with a sound wave of 100 dB at 1,000 Hz per day for 60 minutes and for 3-15 days (Yi et al., 2003b). The relative permeability of root membranes, as measured by the conductivity of the exosmosis solution, did not change under sound stimulation, although there were significant differences in soluble protein content after 6 and 9 days of sound stimulation. After the sound stimulation, the soluble sugar of the roots increased in about 30% and the total amylase activity increased. Based on these results, the authors have hypothesized that robust irradiation accelerates metabolism and growth in chrysanthemum roots. However, these claims were not supported by any growth data.

Sound waves also contribute to the change of hormone levels in tissues. Bochu et al. (2001) have found that a 95 dB sound wave at a frequency of 1.4 kHz, when applied to chrysanthemum plants for 10 days, caused an increase in indole acetic acid (IAA) level and a decrease in abscisic acid (ABA) level. This proportional situation not only accelerated tissue formation but also led to different tissue formation. Yiyao et al. (2002) have determined that certain sound waves contributed to the development of the chrysanthemum plant. But they have stated that the opposite occurred when the energy level of the sound wave increases. It is known that the vibrations created by the low frequency sound wave make a positive contribution to the plant or seeds. At the same time, high frequency sound waves have a negative effect on plants or seeds (Chivukula and Ramaswamy, 2014). In fact, high-vibration sound waves can have a deadly effect on sensitive plants, even at low volume settings (Chivukula and Ramaswamy, 2014).

In another study supporting this, the best results obtained from impatiens seedlings (*Impatiens* sp.) at the same sound pressure level (91-94 dB) exposed to sound vibrations of different frequencies (500,

5.000, 6.000, 12.000, 14.000 Hz) were found in plants exposed to 12.000 Hz frequency, especially in terms of leaf sizes (Collins and Foreman, 2001).

Russowski et al. (2013) have said that the production yield of valepotriate, which is a secondary metabolite, and the growth of cultivated plants without any change could double when *Valeriana glechomifolia* (FGMey.) plants in liquid culture, sonicated with an ultra-sonic sound wave for 2.5-5 minutes (Ultrasonic with 40 kHz). However, the total phenolic content was not affected by ultra-sonic sound treatment, indicating that secondary metabolism was only partially affected by ultrasonication (Wang et al. 2002; Liu et al. 2003; Ananthakrishnan et al., 2007; Rokhina et al., 2009).

To evaluate the effect of NAA, BA and ultra-sonic sound waves on the in vitro rooting and callus induction of *Lilium ledebourii*, scale explants were exposed to an ultrasonic bath with a frequency of 35 kHz for 0, 5, 10, 20 and 30 seconds at different concentrations (0, 0.01, 0.1 and 1 mg l<sup>-1</sup>) NAA and BA alone or in combination with each other were cultured in MS medium. It was stated that plant growth regulators and ultra-sonic sound application did not have a significant effect on callus formation. In this study, the best treatment for increasing in rooting parameters in scale explants was 1 mg l<sup>-1</sup> NAA concentration (Azimzadeh et al., 2018a; Mohebodini et al., (2018) have applied the same treatments to the same plant species under the same conditions and showed that Ultra-sonic sound had a negative effect on root length, thus the highest root length was observed in explants that were not treated with Ultra-sonic sound. The result also showed that ultra-sonic sound had a positive effect on bulb production and root induction. A different effect of growth regulators on the percentage of bulb formation was observed in similar environments. The maximum average weight of the bulblet was within 30 seconds of the ultra-sonic sound treatment, which had a significant difference with the control treatment (without ultrasonic treatment). On the other hand, ultra-sonic sound increased in the number and weight of bulbs. The results showed that bulblet production in the early stages and little root formation in tissue culture are beneficial for rapid bulblet induction and subsequent rooting. Finally, ultra-sonic sound application with plant growth regulators has been reported to have the potential to produce the highest mean number of bulblets in the scale explant. In addition to the results of this research, as a result of the research conducted by Azimzadeh et al., (2018b), parallel results were obtained by applying the same applications to the same plant species under the same conditions.

An efficient micropropagation protocol was developed for *Dendrobium officinale* via stem-like protocorm (PLBs). A correlation has been described between enhanced differentiation of PLBs of *D. officinale* by ultra-sonic sound and changes in endogenous hormone levels and antioxidant enzyme activities. Ultra-sonic sound treatments improved the transformation of PLBs of *D. officinale* into shoots. The highest conversion frequency of PLBs to shoots was obtained from applying ultra-sonic sound at 300W for 5 minutes. When compared to control, the increase in the transformation of PLBs into shoots following ultra-sonic sound application was accompanied by an increase in the ratio of total cytokinins (CTKs) to indole-3-acetic acid (IAA). Ultra-sonic sound application was increased in the activities of superoxide dismutase, catalase and peroxidase during the transformation of PLBs into shoots (Wei et al., 2012).

It was conducted to document the basic anatomical features during development of *P. armeniacum* zygotic embryos and their ability to germinate asymbiotically in vitro. The effect of media and seed pretreatments on seed germination and subsequent seedling growth were also investigated in this study. Pretreatment of dry mature seeds (180 days after pollination) for 90 minutes or with 1.0% sodium hypochlorite solution for 40 kHz ultra-sonic sound with 8 minutes were increased in the germination percentage. Orchid seedlings at least 5 cm height were transplanted into special orchid substrate and 85.3% of the seedlings survived after 180 days from transplantation (Zhang et al., 2015).

## CONCLUSION

As a result of the general evaluation of the study results in this review, we can express that the effects of ultra-sonic sound application on seed germination depend on the frequency, exposure time, distance and density from the organism, the physiological state of the environment, and the temperature of the environment. It can be added that ultra-sonic sound application showed great differences between different plant species and cultivars. In this review, it was emphasized again that low-intensity ultra-sonic sound applications show a number of non-lethal biological effects with potential importance in seed germination of ornamental plants.

Low-energy ultra-sonic sound can alter cellular metabolisms or facilitate nutrient uptake and pass them easily through cell walls and membranes. Due to these characteristics of ultra-sonic sound applications, it has been made many positive contributions to the study results. It is a useful method for improving seed germination, to determine seed viability, germination of aged seeds, accelerating plant and root development by stimulating the roots of the plants, increasing in soluble sugar, protein and amylase activity, also contributing to the change of hormone levels in tissues.

In addition, it can be said that when used in bulb cultivation, it has a positive effect on flower bulb production and root induction, and it can contribute greatly to the quality and aesthetic properties of ornamental plants, especially by increasing in plant leaf size. It can be stated that the use of ultra-sonic sound applications as a priming method may be beneficial in species that have the potential to be many ornamental plants in nature and show low germination rate.

## Conflict of Interest

The article authors declare that there is no conflict of interest between them.

## Author's Contributions

The authors declare that they have contributed equally to the article.

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