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Effects of Different Types of Music on the Germination and Seedling Growth of Alfalfa and Lettuce Plants

You-Ning Lai and How-Chiun Wu[*)]

Department of Natural Biotechnology, Nanhua University, No. 55, Nanhua Road, Sec. 1, Dalin, Chiayi County, 62249, Taiwan

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[*)] Corresponding author:
E-mail: hcwu@nhu.edu.tw

ABSTRACT

The aim of this study was to investigate the effects of music on lettuce and alfalfa seed germination and seedling growth. Nine music treatments were used: Control (no music); Gregorian Chant; Baroque; Classical; Jazz; Rock; Nature sound; New-age; Waltz. For the duration of the study, music was played for 12 hours each day for seven days. Results showed that a significantly lower number of alfalfa seeds germinated in the rock music treatment. In contrast, the highest percentage of alfalfa seeds germinated in treatments with music was those in the classical, nature sound, and waltz treatments. In lettuce seedlings, significantly longer radicles were observed in the Gregorian chant, new-age, and waltz treatments compared to the control, whereas no such effects were evident in alfalfa seedlings. Significantly longer lettuce hypocotyls were found in Gregorian chant, jazz, nature sound, new-age, and waltz music treatments. Of these, although exposure to jazz and nature sound significantly promoted the growth of lettuce hypocotyls, the growth of alfalfa hypocotyls was inhibited under identical conditions. Lettuce seedlings grown in Gregorian chant, new-age, and waltz music produced both significantly longer radicles and hypocotyls, and are suitable to be used as an alternative method to improve seedling growth and development.

INTRODUCTION

Music can be described as a harmonious and coherent blend of various frequencies and vibrations (Chowdhury & Gupta, 2015). There have been many studies on the effects of musical sound on plant growth, some of which attempted to refute the view that music influences plant growth (Gagliano, 2013; Mauck, De Moraes, & Mescher, 2014). In the past, scientists believed that plants cannot hear and process sound waves because plants do not possess structural organs that allow them to detect sound. Nevertheless, it is now known that plants can detect the vibration generated from sounds, and that these vibrations act as a stimulus to plants (Jung, Kim, Kim, Jeong, & Ryu, 2018). Plants are multi-cellular organisms that may not 'hear' sounds, but several current studies have shown that plants can respond to music similar to how humans do. Sound waves can be transmitted by medium materials, and plants' response to music can cause changes in plant metabolism, which can affect plant growth (Creath & Schwartz, 2004; Ramekar & Gurjar, 2016).

Research by Gagliano, Mancuso, & Robert (2012) demonstrated the ability of plants' roots to detect vibrations caused by sound. Furthermore, in addition to detecting vibrations, plant roots also exhibit a frequency-selective sensitivity that generates behavioral modifications. The young roots of corn that were used in their study, which generated structured, spike-like, acoustic emissions, illustrated this point. Moreover, it is also known that plants continuously sense and respond to their dynamic and complex surroundings, which involves identifying important environmental cues and reacting with

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appropriate responses (Mishra, Ghosh, & Bae, 2016). Studies have also established that plants modulate their growth and development in response to environmental factors such as sound waves and other mechanical perturbations. Studies have demonstrated that roots are able to locate a water source by sensing the vibrations generated by the water moving inside pipes (Gagliano, Grimonprez, Depczynski, & Renton, 2017). Plants have also been shown to be able to communicate similarly through acoustic vibration (Gagliano, Renton, Duvdevani, Timmins, & Mancuso, 2012; Mishra, Ghosh, & Bae, 2016). However, the presence of noise affects the root's ability to respond correctly to the surrounding soundscapes.

According to Haswell, Phillips, & Rees (2011), plant membranes comprise a large number of mechano-sensitive channels that are believed to be responsive to mechanical vibrations. There is an increasing number of studies that suggest sound vibrations of certain frequencies can positively influence seed germination, root elongation, callus growth, cell cycling, and other plant processes (Chowdhury, Lim, & Bae, 2014; Hendrawan, Rizky, Susilo, Prasetyo, & Damayanti, 2020; Teixeira da Silva & Dobránszki, 2014). In a study on the germination of *Cicer arietinum* (chickpea) seeds exposed to light Indian music, results showed that it promoted the growth and development of these plants (Chowdhury & Gupta, 2015). However, when noise was used during germination, seed growth was hindered. Findings from another study showed that protein content in plants such as soya, spinach, and wheat was positively affected by Indian classical ragas (Creath & Schwartz, 2004; Reddy, Geetha, & Ragavan, 2013). Furthermore, it was also found that the germination of okra and zucchini seeds were stimulated by musical vibrations.

A study by Vanol & Vaidya (2014) exposed guar plants to classical music, rhythmic rock music, and non-rhythmic traffic noise with varying frequencies. Data on seed germination percentage, plant height, and the number of leaves demonstrated that, compared to untreated plants, classical music and rhythmic rock music showed positive effects, whereas non-rhythmic traffic noise negatively affected the plants. However, results of other studies reported that, in comparison to silence treatment, the growth of bean plants was promoted when exposed to any kind of sound (Singh, Jalan, & Chatterjee, 2013; Vanol & Vaidya, 2014). Few studies have investigated how various types of music affect radicle and hypocotyl growth during the germination stage in lettuce and alfalfa, thus, the aim of this study was to determine the effects of different types of music on their germination percentage and seedling growth.

MATERIALS AND METHODS

Plant Material and Growth Conditions

This research was conducted from November 2018 to April 2019 at the Department of Natural Biotechnology, Nanhua University, Taiwan. Lettuce (*Lactuca sativa*) and alfalfa (*Medicago sativa*) seeds were germinated in 90-mm sterile Petri-dishes lined with two pieces of 90-mm filter paper. Six milliliters of reverse osmosis (RO) water was added to each Petri-dish. Each Petri-dish contained 12 seeds, which were arranged in a 3 x 4 layout. The Petri-dishes were sealed with two layers of Parafilm and placed in a box (33 x 33 x 33 cm). Each box, which represented a single treatment, had five Petri-dishes inside. The Petri-dishes were evenly spaced out with a cellphone placed horizontally facing the Petri-dishes. Each box was fitted with 23-mm thick sound-absorbing Nitrile Butadiene Rubber (NBR) on all 6 sides. The seeds were germinated in total darkness. All the boxes with different music treatments were placed far apart at different areas inside a growth room with the temperature adjusted to 25±2ºC (daytime) and 20±2ºC (nighttime). The relative humidity inside each box was 60-70% throughout the study.

Music Treatments and Playback Conditions

In total, nine music treatments were used, namely: 1) Control (no music); 2) Gregorian Chant; 3) Baroque; 4) Classical; 5) Jazz; 6) Rock; 7) Nature sound; 8) New-age; 9) Waltz (Table 1). A digital sound level meter (Smart Sensor AS804B, Sensor Instruments) was placed inside each box to record the decibel values of each music track. The volume of the music treatments was adjusted to within a similar range. In each music treatment, the track was played for 12 hours (7:00 am to 7:00 pm) for the duration of the study (7 days). Identical cellphones with two speakers were used for music playback in each music treatment. The cellphones were installed with the AutomateIt software program and set up to play music automatically at the set time period and duration.

Table 1. The category, track and decibel value of the music treatments

Music Treatment	Category	Track	Composer/Performer	dB value
Control Group	-	-	-	-
Gregorian Chant	Religious	Ave Mundi Spes Maria (Sequenza (Modos VII Y VIII))	Choir of the Benedictine Abbey of Santo Domingo de Silos	79±9
Baroque	Class	The Four Seasons, Spring (La Primavera)	A.L.Vivaldi	91±9
Classical	Class	Duettino Sull'aria Le nozze di Figaro	W.A.Mozart	84±7
Jazz	Modern	In the Swing	Upbeat Brass	93±4
Rock	Modern	All Shall Fall	Immortal	87±8
Nature Sounds	-	Water and Bird Sounds	None	90±6
New Age	Modern	Self-composed Track	Self-composed by author	87±4
Waltz	Modern	Self-composed Track	Self-composed by author	85±4

Statistical Analysis

A total of 60 replicates per treatment was used. The seeds were germinated for 7 days, during which the following data were collected: Germination percentage was collected on the 3rd, 5th, and 7th day. On the 7th day, the radicle length and hypocotyl length of all germinated seedlings were measured. Each seedling was placed on a workbench and its radicle and hypocotyl were carefully straightened against a ruler to take their measurements. Data were analyzed using the Chi-square test and Duncan's Multiple Range Test to compare germination % and treatment means, respectively, using SPSS v. 17 software.

RESULTS AND DISCUSSION

Effect of Different Types of Music on Germination Percentage

In lettuce seeds, high germination percentages were evident throughout all music treatments. Results showed that no significant differences were found between the different types of music on the germination percentage of lettuce seeds after 3, 5, or 7 days (Table 2). After 7 days, the lowest and highest germination percentages were found in the classical (93.3%) and nature sound treatments (100%), respectively, while 98.3% of lettuce seeds germinated in the control treatment. On the contrary, significant differences were observed among music treatments in alfalfa seed germination after 5 days and 7 days, namely, classical, nature sound, and waltz music showed 100% germination, whereas rock music significantly reduced the germination percentage (91.7%) (Table 3). These findings are in agreement with those reported by Chivukula & Ramaswamy (2014) where rock music containing hardcore vibrations was found to inhibit plant growth. However, it is also important to note that other characteristics of rock music may also be an important factor in their effects on seed germination, which could be a factor in the present study as the inhibitory effects of the rock music treatment on alfalfa seed germination were not as profound as one would expect, with over 90% germination. Evidence of this nature has been shown in literature where, despite rock music often being referred to as 'noise' or 'unharmonious' music, there are studies that found positive influences. Specifically, regardless of the type of music played, it seems those that are considered rhythmic music or rhythmic frequencies tend to have a positive effect. This was demonstrated in the study by Vanol & Vaidya (2014) where rhythmic rock music showed a positive effect on the germination of seeds. Ekici, Dane, Mamedova, Metin, & Huseyinov (2007) also found rhythmic music to positively affect root elongation and mitotic division in onion root tips during germination. Thus, compelling cases demonstrating rhythmic differences in music that directly affect germination seem to be gaining recognition.

Table 2. The effects of different types of music on germination % of lettuce seeds

Music Treatment	Germination %		
	3 days	5 days	7 days
Control	98.3[a]	98.3[a]	98.3[a]
Gregorian Chant	95.0[a]	95.0[a]	95.0[a]
Baroque	98.3[a]	98.3[a]	98.3[a]
Classical	93.3[a]	93.3[a]	93.3[a]
Jazz	96.7[a]	96.7[a]	98.3[a]
Rock	95.0[a]	95.0[a]	98.3[a]
Nature sound	100[a]	100[a]	100[a]
New age	98.3[a]	98.3[a]	98.3[a]
Waltz	98.3[a]	98.3[a]	98.3[a]

Remarks: Percentages in the same column with different letters are significantly different (Chi-square at $P \leq 0.05$)

Table 3. The effects of different types of music on germination % of alfalfa seeds

Music Treatment	Germination %		
	3 days	5 days	7 days
Control	98.3[a]	100[a]	100[a]
Gregorian Chant	96.7[a]	96.7[a]	96.7[a]
Baroque	93.3[a]	95.0[a]	96.7[a]
Classical	100[a]	100[a]	100[a]
Jazz	96.7[a]	96.7[a]	96.7[a]
Rock	91.7[a]	91.7[b]	91.7[b]
Nature sound	98.3[a]	100[a]	100[a]
New age	93.3[a]	93.3[a]	93.3[a]
Waltz	98.3[a]	100[a]	100[a]

Remarks: Percentages in the same column with different letters are significantly different (Chi-square at $P \leq 0.05$)

Nevertheless, the results of the present study suggest that the rhythmic characteristics of music are not solely responsible for affecting seed germination, given that the unharmonious, non-rhythmic hard rock music used in this study significantly inhibited alfalfa seed germination but did not cause significant adverse effects in lettuce seed germination. This finding is indicative of the importance of taking into consideration the different facets that makeup music, such as rhythm, melody, harmony, repetition or variation, and instruments, and how each component individually or in combination can affect seed germination or plant growth. Furthermore, it is particularly noteworthy that both lettuce and alfalfa seeds exposed to nature sound and classical music, respectively, were the only treatments that had 100% germination by day 3 (Table 2; Table 3). These results agree with those reported by Vanol & Vaidya (2014), where classical music was found to have a positive effect on guar seed germination.

Effect of Different Types of Music on Radicle Growth

In lettuce seedlings, results showed that after 7 days, significantly higher radicle lengths were observed in the Gregorian chant, new-age, and waltz treatments compared to the control (Table 4). Of these three treatments, seedlings in the Gregorian chant and waltz treatments produced the longest radicles, which were significantly longer than those exposed to new-age music. With regard to the radicle growth of alfalfa seedlings, no significant differences were found between all treatments (Table 4). Nevertheless, results indicated that alfalfa seeds grown with jazz music tend to have shorter radicles, whereas those germinated in Gregorian chant and classical music tend to produce longer radicles.

Metabolism in plants can be influenced by music, including its frequency (Hz) (Chowdhury & Gupta, 2015). According to Coghlan (1994), the production of protein increases when music at the appropriate frequency is played, which stimulates overall plant growth. Similarly, it has been found that audible sound at 125 Hz and 250 Hz frequencies, plant genes are more active in DNA code translation, and as a result, growth and development are increased (Ekici, Dane, Mamedova, Metin, & Huseyinov, 2007). The results of the present study agree with this finding as shown by the radicles of lettuce seedlings that were exposed to Gregorian chant, which falls close to within the abovementioned frequencies, being significantly longer than most of the other music treatments (Table 4). Nonetheless, these results also clearly showed that the effects of music on radicle growth are highly dependent on plant type as illustrated by the non-significant responses of alfalfa seedlings in all the music treatments. Several reports have shown that music with string instruments such as violins tend to have a positive impact on plant growth (Chivukula & Ramaswamy, 2014; Laad & Viswanathan, 2010; Petrescu, Mustâţea, & Nicorini, 2017; Reddy, Geetha, & Ragavan, 2013). However, in the present study, no significant differences were found in the growth of radicles and hypocotyls of lettuce and alfalfa seedlings between the control treatment and the baroque music treatment, which was primarily performed with violins.

Table 4. The effects of different types of music on radicle growth of lettuce and alfalfa seedlings after 7 days of germination

Treatment	Radicle Length (mm)		Frequency (Hz)
	Lettuce	Alfalfa	
Control	32.49±8.88[ab]	39.75±15.89[a]	-
Gregorian Chant	43.77±9.38[d]	43.36±17.15[a]	131 ~ 349
Baroque	32.44±8.80[ab]	41.62±13.59[a]	123 ~ 1661
Classical	29.26±10.23[a]	43.93±15.87[a]	466 ~ 1397
Jazz	31.48±8.86[ab]	38.32±12.88[a]	207 ~ 557
Rock	34.53±11.01[b]	40.33±14.37[a]	196 ~ 294
Nature Sound	34.22±10.83[b]	39.17±16.59[a]	Pink Noise
New-age	39.76±9.95[c]	41.76±15.00[a]	73 ~ 1760
Waltz	43.58±12.71[d]	41.02±16.59[a]	147 ~ 1319

Remarks: Different letters in the same column indicate values differ significantly according to Duncan's Multiple Range test ($P \leq 0.05$)

Effect of Different Types of Music on Hypocotyl Growth

In lettuce seedlings, except for those in the baroque, classical, and rock music treatments, which had similar hypocotyl lengths to those in the control treatment, the hypocotyls of seedlings grown in all the other music treatments were significantly longer than the control (Table 5). Overall, the highest hypocotyl length was produced in jazz and waltz music treatments, whereas those germinated in the control treatment produced the shortest of all treatments. In contrast to lettuce seedlings, the response of the growth of alfalfa hypocotyls to music was less pronounced. Except for jazz and nature sound, which had significantly shorter hypocotyls, the hypocotyl length of alfalfa seedlings grown without any music was comparable to the other music treatments (Table 5). In general, when lettuce seedlings were exposed to music, there seemed to be a stimulatory effect on hypocotyl growth regardless of the type of music used. The response of lettuce in our study is similar to those reported by Singh, Jalan, & Chatterjee (2013) and Vanol & Vaidya (2014) who found that, in comparison to no sounds, growth of bean plants was promoted when exposed to any type of music treatment. Overall, the results of the present study clearly showed that the growth of both the radicle and hypocotyl of alfalfa seedlings was less responsive to music treatments while the growth of lettuce seedlings was more easily affected by the presence of music.

With regard to the response of plants to green music, which comprises natural sounds such as birds, insects, and water, studies have shown seed germination to be significantly affected when germinated under these treatments (Creath & Schwartz, 2004). In the present study, the hypocotyl growth of lettuce plants was significantly promoted by nature sound compared to the control treatment (Table 5). However, the opposite effect was true in alfalfa seedlings where significantly shorter hypocotyls were observed in nature sound than those in the control treatment. Similar effects on hypocotyl growth were also evident in the jazz treatment (Table 5). This is further evidence that positive or negative responses to a particular type of music are plant-specific, and in the case of this study, only present in certain organs of the seedling as these effects were not observed in the radicle treatments.

Table 5. The effects of different types of music on hypocotyl growth of lettuce and alfalfa seedlings after 7 days of germination

Treatment	Hypocotyl Length (mm)		Frequency (Hz)
	Lettuce	Alfalfa	
Control	33.93±5.57^{a}	50.11±15.08^{cde}	-
Gregorian Chant	38.83±6.71^{cd}	49.66±11.13^{cde}	131 ~ 349
Baroque	36.20±6.08^{abc}	47.89±13.27^{bcd}	123 ~ 1661
Classical	35.82±7.36^{ab}	50.94±10.93^{de}	466 ~ 1397
Jazz	39.98±9.64^{d}	44.00±9.76^{ab}	207 ~ 557
Rock	36.65±8.01^{abc}	45.66±11.50^{bc}	196 ~ 294
Nature Sound	38.34±7.26^{bcd}	39.53±10.35^{a}	Pink Noise
New-age	38.95±6.18^{cd}	48.17±11.29^{bcd}	73 ~ 1760
Waltz	39.84±6.74^{d}	53.25±13.15^{e}	147 ~ 1319

Remarks: Different letters in the same column indicate values differ significantly according to Duncan's Multiple Range test ($P \leq 0.05$)

CONCLUSION

Rock music was found to significantly inhibit the germination of alfalfa seeds. Results also showed that Gregorian chant, new age, and waltz music were able to induce lettuce seedlings to produced significantly longer radicles and hypocotyls. Compared to seedlings grown without any music, exposure to nature sound or jazz music resulted in inverse effects in the growth of hypocotyls of lettuce and alfalfa. Further studies are needed to determine the specific factors within each music type that causes these effects.

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