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ARAŞTIRMA MAKALESİ **RESEARCH ARTICLE**

Impact of Ultrasonic Pre-Treatments on Germination and Initial Seedling Growth of Common Vetch (*Vicia sativa* L.)

Ultrasonik Ön Uygulamaların Adi Fiğin (*Vicia sativa* L.) Çimlenme ve İlk Fide Gelişimi Üzerine Etkisi

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Abstract

Although common vetch (*Vicia sativa* L.) is a very important forage legume plant, some varieties have a dormancy problem due to the hardness of the seed coat. So far, studies on breaking this dormancy seen in common vetch seeds have been quite limited. This research was carried out to determine the effects of ultrasonic pre-treatments applied to common vetch seeds under laboratory conditions on germination and initial seedling growth. Ultrasonic pre-treatment (US) was applied to common vetch seeds at a frequency of 47 kH for 0 -control-, 10, 20, 30 and 40 minutes, respectively. The experiment was designed according to a completely randomized plot trial design with 4 replications. In the research, germination percentage (GP-%-), mean germination time (MGT-day-), germination rate index (GRI-%-), shoot and root lengths (SL and RL-cm-), root-shoot ratio (R/S -%-), shoot and root fresh weights (SFW and RFW-mg per plant-), shoot and root dry weights (SDW and RDW-mg per plant-), shoot and root dry matter (SDM and RDM-% -), root/shoot dry matter ratio (R/S DM-%-), shoot and root water contents (SWC and RWC-%-) and seedling vigor index (SVI) parameters were measured. As a result of the research, it was determined that ultrasonic pre-treatments had statistically significant ($P \leq 0.05$) effects on germination parameters (GP, MGT and GRI). It has been determined that US_{10} and US_{40} treatments increase the GP by 11.11%. However, it was determined that US_{30} treatment shortened the MGT by 15.81% and increased the GRI and SL by 100.00% and 34.65%, respectively. In addition, it was determined that ultrasonic pre-treatments had statistically significant effects on growth parameters (RL, SL, R/S ratio, SFW, RFW, SDW, RDW, SDM, SDM and SVI). Compared to the control group, an increase of 42.95%, 51.35%, 17.75% and 21.00% in the RL, R/S ratio, SFW and RFW parameter values, respectively, was obtained from the US_{10} treatment. Additionally, US_{40} treatment caused an increase in RDW, SDM, RDM values by 51.51%, 33.83% and 33.87%, respectively. The highest increase in SVI was recorded in the US_{30} treatment with 30.56%. In conclusion, the data of this study showed that ultrasonic technology has a positive effect on breaking dormancy in common vetch seeds and thus on the initial seedling growth.

Keywords: Common vetch, Ultrasonic treatment, Dormancy, Seed germination, Initial seedling growth

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Öz

Adi fiğ (*Vicia sativa* L.), çok önemli bir baklagil yem bitkisi olmakla birlikte tohum kabuğunda karşılaşılan sertlik nedeniyle dormansi problemi görülmektedir. Şimdiye kadar, adi fiğ tohumlarında görülen bu dormansinin kırılmasına yönelik çalışmalar oldukça sınırlı kalmıştır. Bu araştırmada, laboratuvar koşulları altında yaygın fiğ tohumlarına uygulanan ultrasonik ön uygulamalarının çimlenme ve bununla birlikte ilk fide gelişimi üzerine etkilerinin belirlenmesi amacıyla yürütülmüştür. Ultrasonik ön uygulaması (UÖU) adi fiğ tohumlarına sırasıyla 0 -kontrol-, 10, 20, 30 ve 40 dakika buyunca 47 kH frekansında uygulanmıştır. Deneme 4 tekerrürlü olarak tamamen tesadüf blokları deneme desenine göre dizayn edilmiştir. Araştırmada, çimlenme yüzdesi (ÇY-%-), ortalama çimlenme süresi (OÇS-gün-), çimlenme hızı indeksi (ÇHİ-%-), sürgün ve kök uzunlukları (SU ve KU-cm-), kök-sürgün oranı (K/S-%-), sürgün ve kök taze ağırlıkları (STA ve KTA-mg bitki başına-), sürgün ve kök kuru ağırlıkları (SKA ve KKA-mg bitki başına-), sürgün ve kök kuru maddeleri (SKM ve KKM-%-), kök/gövde kuru madde oranı (K/S KM-%-), sürgün ve kök su içerikleri (SSİ ve KSİ-%-) ve fide güç indeksi (FGİ) parametreleri ölçülmüştür. Araştırma sonucunda, ultrasonik ön uygulamalarının çimlenme parametreleri (ÇY, OÇS ve ÇHİ) üzerine istatistiki açıdan önemli ($P \leq 0.05$) etkilerinin olduğu belirlenmiştir. $UÖU_{10}$ ve $UÖU_{40}$ uygulamalarının ÇY'yi %11.11 oranında artırdığı tespit edilmiştir. Bununla birlikte, $UÖU_{30}$ uygulaması OÇS'yi %15.81 kısalttığı, ÇHİ'yi ve SU'yu sırasıyla %100,00 ve %34,65 oranlarında artırdığı belirlenmiştir. Ayrıca, ultrasonik ön uygulamalarının büyüme parametreleri (KU, SU, K/S oranı, STA, KTA, SKA, KKA, SKM, KKM ve FGİ) üzerine de istatistiki açıdan önemli etkilerinin olduğu belirlenmiştir. Kontrol grubu ile kıyaslandığında, KU, K/S oranı, STA ve KTA parametre değerlerinde sırasıyla %42.95, %51,35, %17,75 ve %21,00 oranlarında bir artış $UÖU_{10}$ uygulamasından elde edilmiştir. İlave olarak $UÖU_{40}$ uygulaması KKA, SKM, KKM değerlerinde sırasıyla %51.51, %33.83 ve %33.87 oranlarında artışa neden olmuştur. FGİ'de ise en yüsek artış %30.56 ile $UÖU_{30}$ uygulamasında kayıt edilmiştir. Sonuç olarak bu araştırmanın verileri, ultrasonik teknolojisinin *adi fiğin* tohumlarındaki dormasinin kırılmasında ve bununla birlikte ilk fide gelişimi üzerine olumlu yönde etkisinin olduğunu göstermiştir.

Anahtar Kelimeler: Adi fiğ, Ultrasonik uygulama, Dormansi, Tohum çimlenmesi, İlk fide büyümesi

1. Introduction

Common vetch *(Vicia sativa* L.) is a grain legume crop that is highly valued for its protein level, fatty acid profile, and mineral composition. These nutritional characteristics make it an excellent choice for enhancing animal feed. Furthermore, significant pharmacological characteristics have been documented in human subjects. The common vetch, like other legumes, has the ability to perform nitrogen fixation, which is a vital characteristic for the development of sustainable agricultural systems. The aforementioned characteristics augment the use of vetch as a protective crop and its implementation in intercropping schemes (Nizam et al., 2022; Ramírez-Parra and De la Rosa, 2023). Despite all these important features of vetch, there is a problem of dormancy due to the hard seed coat (Büyükkartal et al., 2013). 'Hard seededness', or 'physical dormancy', is present in a few plant families, of which the *Leguminosae* are the largest (Paulsen et al*.,* 2013).

Seed dormancy is a characteristic of plants that prevents seed germination in favorable conditions. This property has developed to increase the chances of seedling survival by preventing germination in unfavorable climatic conditions (Iwasaki et al*.,* 2022). The reason of dormancy is seed coat and embryo (Yildiz et al*.,* 2017). Although the hardseededness (physiological dormancy) increases the survival rate of seeds in the soil, especially in adverse environmental conditions, and helps prevent the extinction of species in nature, it prevents rapid germination and healthy seedling development in plant species, and may prevent the use of plant varieties or wild species relatives for agricultural or breeding purposes (Tiryaki and Topu, 2014; Yildiz et al*.,* 2017; Altındal and Altındal, 2018). Physical dormancy, referred to as hardseededness or seed coat impermeability, is a situation wherein a seed's coat becomes impermeable, obstructing the ingress of water, gasses, and other external elements. This impermeability functions as a protective mechanism, postponing germination until optimal conditions are achieved. Factors affecting hard seed development are categorized into internal and external influences. Internal variables relate to plant-specific characteristics, including species and seed shape. Genetic differences and seed coat attributes influence the development of hard seeds. Environmental conditions also affect seed growth. Soil nutrient availability, water supply, humidity, temperature, and light conditions influence seed coat permeability and germination. The timing of seed maturity, drying, and storage conditions can influence the production of hard seeds. The interaction of these elements influences a plant's propensity to generate hard seeds (Ermis et al., 2024). There are some methods used for breaking seed coat dormancy such as scarification of seed coat by mechanical methods, temperature treatments, acid treatments, chemical treatments (Kinetin, GA3). Furthermore, a range of physical and extraterrestrial environmental elements, such as electromagnetic waves (EWs), magnetic fields (MFs), and ultrasonic sounds (US), have been documented as a potentially effective method for disrupting dormancy. The electromagnetic spectrum encompasses both ionizing and non-ionizing radiations. The spectrum of ionizing radiation is composed of X-rays and gamma rays, whereas non-ionizing radiation includes ultrasonic waves, magnetic fields, microwaves, and infrared light, among others (Alsuvaid and Demir, 2022; Bera et al*.,* 2022).

In recent years, ultrasonic waves have been widely used as an effective method for disrupting seed dormancy and enhancing germination properties (Nazari and Eteghadipour, 2017). The application of ultrasonic waves to seeds is an environmentally friendly technology that shows potential for revolutionizing the food business. It improves the process of seed germination and seedling growth in various species by stimulating the absorption of water and oxygen, as well as boosting seed metabolism (Nogueira et al*.,* 2023). The ultrasonic waving technique appears to be more convenient, safer, and healthier, while also being more time-efficient compared to conventional chemical and physical procedures (Sharififar et al., 2015). This therapy utilizes ultrasonic frequencies (20-100 kHz) to interact with material or seeds in either air or water (Memiş et al*.,* 2022). It has been reported by many researchers so far that ultrasonic treatments break seed dormancy and positively affect germination and seedling growth in many plant species (Goussous et al., 2010; Nazari et al., 2014; Lahijanian and Nazari, 2017; Sharififar et al*.,* 2015; Andriamparany and Buerkert, 2019; Babaei-Ghaghelestany et al*.,* 2020; Wang et al*.,* 2020; Alfalahi et al*.,* 2022; Memiş et al*.,* 2022; Babaei et al*.,* 2023; Foschi et al*.,* 2023). However, a thorough examination of previous studies revealed a lack of information regarding the assessment of ultrasound technology's efficacy in breaking the seed dormancy of common vetch. There are limited reports on breaking seed dormancy of common vetch in the literature (Samarah et al*.,* 2004; Samarah, 2005; Uzun, 2012). Hence, the objective of this research was to employ ultrasound waves as a noninvasive, uncomplicated, and economical method to induce dormancy release and promote early seedling development in common vetch.

2. Materials and Methods

2.1. Seed Materials

The study utilized common vetch seeds, gathered in 2023, received from the Department of Field crops at Kırşehir Ahi Evran University in Türkiye. An *in vitro* experiment was conducted in 2023 at the Department of Agricultural Biotechnology, Faculty of Agriculture, Kırşehir Ahi Evran University in Türkiye.

2.2. Ultrasonic irradiation treatments

100 seeds for each treatment were treated with 47 kHz ultrasonication in 200 ml of distilled water for 0, 10, 20, 30, and 40 mininutes using a completely randomized design. During ultrasonic treatment, water temperature was kept at 21±2°C by adding ice from time to time. Ultrasonication treatments are labeled US_{10}, US_{20}, US_{30}, and US_{40}, respectively. During the ultrasonication process, the seeds are in contact with distilled. Therefore, in order to determine the effect of the soaking factor on germination and initial seedling growth paramaters, the seeds were soaked in water for 0, 10, 20, 30, and 40 minutes, just like in the ultrasonication process, and these treatments were labeled as -water soaking- WS_{10}, WS_{20}, WS_{30}, and WS_{40}, respectively. Soaking the seeds in water is considered hydro-priming. Seeds that were not exposed to water or ultrasonic treatments were used as controls.

2.2. Germination Assay and Growth Measurement

Test solution were prepared with distilled water. For each treatment, 25 seeds were sown between three rolls of filter paper in four replicates. At the beginning of the experiment, each subject was irrigated with 10 ml of test solution. Before planting, seeds were given a fungicide treatment (Thiram 80%). To prevent moisture loss, the rolled paper with seeds was placed in sealed, clear plastic bags. For 14 days, seeds were allowed to germinate at 20 ± 1ºC degrees in the dark in incubator (Memmert-In110). The radicles were deemed to have germinated when they reached a length of ~ 2 mm. For two days, the germination percentage was tracked every 24 hours (ISTA, 2003).

Germination Percentage(GP) = (Number of germinating seeds/ Total number of seeds) × 100

(Al-Enezi et al., 2012) (Eq. 1)

Equation 1 was used to calculate the proportion of seeds that germinated after being subjected to ultrasonic treatments. To assess the rate of germination, the mean germination time (MGT) was determined (Ellis and Roberts, 1980).

$$MGT=\sum Dn/\sum D \quad \text{(Eq. 2)}$$

where n is the number of freshly germinated seeds on day D, and D is the number of days since the start of the experiment.

The germination rate index (speed of germination) was calculated using the formula according to Maguire (1962).

GRI = Σ No of Germinated Seeds/ Σ No of Days (Eq. 3)

Seedlings with stunted primary roots and short, thick, spiral-shaped hypocotyls were deemed to have aberrant germination. Initial seedling growth paramaters (shoot and root length, shoot and root fresh weights, shoot and root dry weights, shoot and root dry matter, shoot and root water content, and seedling vigor index) were measured after the 14th day (*Figure 1*). Samples were dried in an oven at 70°C for 48 hours before dry weights were calculated (Beyaz et al., 2011). The following parameters were used to calculate the growth parameters.

Water content (WC) = (fresh weight-dry weight)/fresh weight×100 (Zheng et al., 2008) (Eq. 4)

Dry matter (DM)= (dry weight/fresh weight) × 100 (Bres et al., 2022) (Eq. 5)

Seedling vigor index (SVI)= (average root length +average hypocotyl length) x germination percentage

(Abdul-Baki and Anderson, 1973) (Eq. 6)

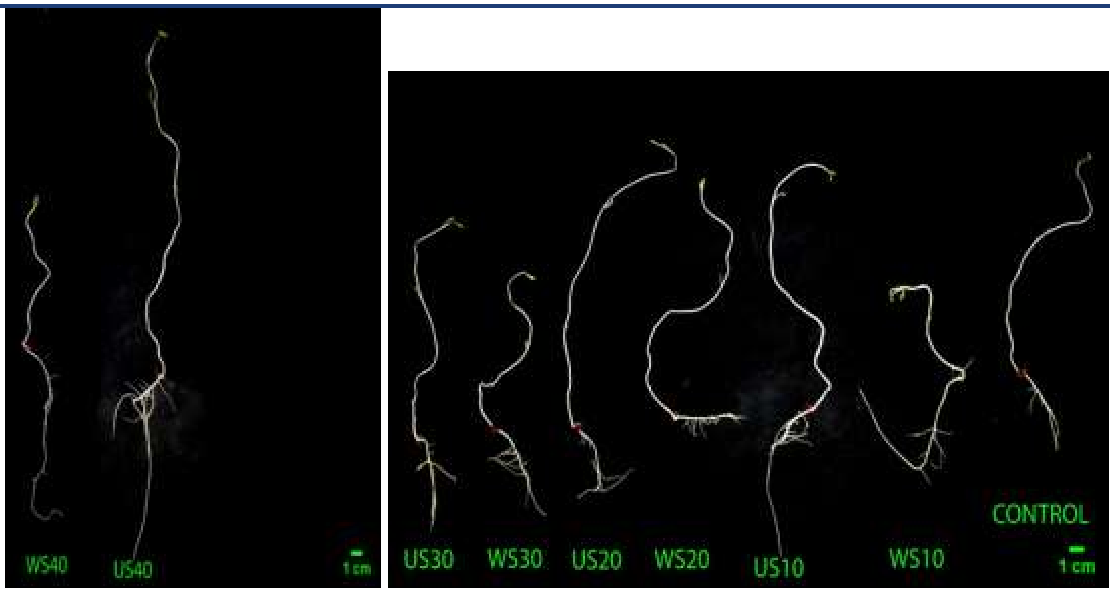

Figure 1. The morphology of common vetch seedlings after water soaking (WS) and ultrasound (US) treatments (after 14 days of germination)

Treatments*:

Control: Seeds that were neither soaked in distilled water nor exposed to ultrasonic treatments.

US_{10}, US_{20}, US_{30}, and US_{40}: Seeds exposed to ultrasonic (ultrasound) treatments for 10, 20, 30, and 40 minutes, respectively.

WS_{10}, WS_{20}, WS_{30}, and WS_{40}: Seeds soaked in distilled water for 10, 20, 30 and 40 minutes respectively. It is considered a hydropriming treatment.

2.2. Statistical Analysis

The gathered data were analyzed using a one-way Analysis of Variance (ANOVA) in the statistical software package SPSS 22. This analysis aimed to evaluate whether the treatments had a significant impact on the observed parameters. The statistical significance of the means was assessed using the Duncan Multiple Range Test (DMRT) at a significance level of $P \leq 0.05$. Before statistical analysis, the data in percentages were transformed using Arcsine transformation (Snedecor and Cochran, 1967).

3. Results and Discussion

3.1. Effects of ultrasonic pre-treatmens on germination of Common Vetch Seeds

The effect of different ultrasound (US) and water soaking (WS) treatments on the germination percentage (GP), germination rate index (GRI) and mean germination time (MGT) is given in *Table 1* and it was determined that there was a statistical difference between the ultrasound (US) and water (W) treatments in terms of GP ($P \leq 0.05$), GRI ($P \leq 0.05$), and MGT ($P \leq 0.01$). Water and US treatments caused an increase in GP compared to the control group. However, when water soaking (WS) and US treatments were compared, it was observed that the GP increase was greater in US treatments. The highest GP values of 100% were obtained from US_{10} and US_{40} treatments. The lowest GP value (90.00%) was recorded in the control group. The observed increase in GP due to increased ultrasound coincides with the findings of various researchers in other plants (Aladjadjiyan, 2002; Yaldagard et al., 2008; Goussous et al., 2010; Andriamparany and Buerkert, 2019; Babaei-Ghaghelestany et al., 2020; Alfalahi et al., 2022). The surface and architecture of the seed coat may potentially have an impact on the ultrasonic effect (Memiş et al., 2022). The primary effects of ultrasonic waves are mostly attributed to mechanical phenomena, such as acoustic cavitation and the disruption of plant cell walls. These processes ultimately lead to an enhanced uptake of water (Abd El-Sattar and Tawfk, 2023). The occurrence of cavitation during the sonication process was postulated as an additional potential factor that caused micro fissures on the seed coat. These fissures may have facilitated the absorption of moisture, leading to an accelerated germination rate (Yaldagard et al., 2008;

Table 1. Effects of ultrasound (US) and water soaking (WS) treatments on germination percentange (GP), mean germination time (MGT), germination rate index (GRI), shoot lenght (SL), root lenght (RL), root to shoot ratio (R/S) of 14 days-old common vetch seedling

Treatments	GP (%)	MGT (day)	GRI (%)	SL (cm)	RL (cm)	R/S (%)
0-control-	90.00 ± 0.0^{c}	1.96 ± 0.03^{a}	4.83 ± 0.44^{d}	14.60 ± 1.1^{b}	5.75 ± 0.5^{bc}	0.37 ± 0.04^{bc}
WS$_{10}$	96.66 ± 3.3^{ab}	1.74 ± 0.04^{c}	8.16 ± 0.60^{ab}	13.70 ± 0.3^{b}	6.13 ± 0.4^{bc}	0.44 ± 0.02^{ab}
US$_{10}$	100.00 ± 0.0^{a}	1.73 ± 0.04^{c}	8.66 ± 0.88^{ab}	14.81 ± 1.3^{b}	8.22 ± 0.3^{a}	0.56 ± 0.03^{a}
WS$_{20}$	93.33 ± 3.3^{ab}	1.78 ± 0.04^{ab}	7.33 ± 0.60^{abc}	14.63 ± 0.3^{b}	4.90 ± 0.3^{c}	0.34 ± 0.00^{bc}
US$_{20}$	96.66 ± 3.3^{ab}	1.79 ± 0.05^{ab}	7.50 ± 0.86^{abc}	14.55 ± 0.6^{b}	6.54 ± 0.5^{abc}	0.44 ± 0.02^{ab}
WS$_{30}$	93.33 ± 3.3^{ab}	1.83 ± 0.08^{ab}	6.66 ± 1.01^{bcd}	14.33 ± 1.1^{b}	4.92 ± 0.4^{c}	0.35 ± 0.06^{bc}
US$_{30}$	93.33 ± 1.6^{c}	1.65 ± 0.05^{c}	9.66 ± 0.83^{a}	19.66 ± 2.0^{a}	6.31 ± 0.8^{abc}	0.32 ± 0.01^{c}
WS$_{40}$	90.00 ± 0.0^{c}	1.93 ± 0.06^{a}	5.16 ± 0.72^{cd}	16.56 ± 0.4^{ab}	7.06 ± 0.5^{ab}	0.42 ± 0.04^{bc}
US$_{40}$	100.00 ± 0.0^{a}	1.71 ± 0.07^{c}	9.33 ± 1.45^{ab}	16.32 ± 1.4^{ab}	7.75 ± 1.0^{ab}	0.47 ± 0.03^{ab}
Summary of ANOVA						
Treatment	*	*	**	*	**	**

*significant at $P\leq0.05$, **significant at $P\leq0.01$. Different letters at the same column show significant differences at 0.05 level. ±: Standard errors.
Treatments*:
Different durations of two treatments: Seeds soaked in water (WS) or soaked in water in ultrasonic device (US).
Control: Seeds that were neither soaked in distilled water nor exposed to ultrasonic treatments.
US$_{10}$, US$_{20}$, US$_{30}$, and US$_{40}$: Seeds exposed to ultrasonic (ultrasound) treatments for 10, 20, 30, and 40 minutes, respectively.
WS$_{10}$, WS$_{20}$, WS$_{30}$, and WS$_{40}$: Seeds soaked in distilled water for 10, 20, 30 and 40 minutes respectively. It is considered a hydropriming treatment.

Abd El-Sattar and Tawfk, 2023). According to Bai et al. (2023), The ultrasound-germination therapy across the entire germination phase led to a notable increase in water content compared to the germination treatment alone in barley.

When compared to the control group, water soaking (WS) and US treatments caused a shortening of MGT. This shortening was recorded at higher values in ultrasound treatments (*Table1*). Considering the control group, the highest shortening was obtained from US_{30} and US_{40} treatments, with 15.81% and 14.61%, respectively. Memiş et al. (2022) reported that ultrasound treatment positively affects MGT and shorten its duration in vegetable species such as watermelon, melon, leek, pepper, carrot, tomato and aubergine. GRI increased in both water and US treatments when control was considered (*Table 1*). However, this increase was greater in US treatments. The highest increase was obtained from US_{40} treatment with 93.16 % compared to the control. Similar studies have reported that ultrasound treatments increase GRI in other plants (Goussous et al., 2010; Andriamparany and Buerkert, 2019; Namjoo et al., 2022; Parlak and Erken, 2023). The high germination index is considered to be a reliable sign of strong seed viability (Büyükyıldız et al., 2023). Consequently, based on the research findings, it can be concluded that US treatments enhance the seed vigor of vetch. Ultrasonic waves can accelerate the germination process of common bean seeds by producing tiny openings on the seed coat and reducing the stiffness of the cotyledon cell walls. This allows the seeds to absorb water more efficiently, leading to faster and more successful water uptake (Lahijanian and Nazari, 2017). Ultrasound improves the germination of specific seeds by stimulating the activity of different enzymes (Potolea et al., 2019).

3.2. Effects of ultrasonic pre-treatmens on morphological characters of Vicia sativa L. Seedlings

The effect of WS and US treatments on shoot lenght (SL), root length (RL), and root to shoot ratio are seen in *Table 1*. Statistically, the effect of both WS and US treatments on these parameters was found to be significant. For SL, statistically 0-control-, WS_{10}, US_{10}, WS_{20}, US_{20} remained in the same group. The highest SL (19.66cm) was obtained from the US_{30} treatment. It was calculated that there was a 34.65% increase in SL compared to the control group. However, it has been observed that WS (except, WS_{20} and WS_{30}) and US treatments increase RL. The highest RL value was recorded in US_{10} treatment with 8.22 cm. There was an increase of 42.95% between US_{10} and the 0-control- group. Considering R/S, the highest value was again obtained from US_{10} treatment with 0.56%. The increase here between the control group and US_{10} treatment was 51.35%. WS and US treatments affected SL, RL, and R/T parameters differently. In general, the results of the study showed that both treatments provided a general increase in these parameters, and this increase was greater in US treatments. Alfalahi et al. (2022) reported that ultrasonic exposure increased root and shoot lengths of the three soybean varieties. Zeng et al. (2023) reported that the application of ultrasonic treatment had a substantial impact on the growth of sugarcane shoots in all three cultivars. According to Huang et al. (2020), using lower levels of ultrasound over a prolonged period can promote cell division and maintain high-quality seedlings.

The results of WS and US treatments effects on shoot fresh weight (SFW), root fresh weight (SFW), shoot dry weight (SDW), and root dry weight (RDW) are shown in *Table 2*. SFW, RFW, SDW, and RDW were significantly affected by WS and US treatments. In general, SFW (except, WS_{20}, WS_{30}, US_{40}) and RDW (except, W_{30}) caused an increase in both applications. For SFW, the highest result (0.126 mg per plant) obtained from both US_{10} and WS_{10} treatments. However, for RFW, the highest value (0.144 mg per plant) was recorded at US_{10}. Considering RDW and RDW, the highest values were obtained from WS_{30} treatments with 0.0144 mg per plant and US_{40} treatments with 0.0300 mg per plant, respectively. The effects of ultrasonic assisted seeds priming (0 -cotnrol-,10, and 20 min) with a frequency of 40 kHz on seed enhancement of Fenugreek seeds were investigated by (Abd El-Sattar and Tawfk, 2023) who showed that SFW, RFW, SDW and RDW were signifcantly improved by using ultrasonic treatments.

Table 2 shows effects of the different durations of US treatment compared to WS on the shoot and root dry matter (SDM and RDM), root to shoot dry matter (R/S DM), shoot and root water contents (SWC and RWC), and seedling vigor index (SVI). For SDM, RDM, R/S DM, SWC, RWC and SVI, the means were statistically significant between the control, WS, and US treatment groups. Compared to the control, it was determined that WS and US treatments generally caused a decrease in SDM (except, WS_{40} and US_{40}) and an increase in RDM. However, it also caused an increase in R/SDM. The highest SDM and RDM values were obtained from US_{40} treatment with 14.36% and 22.05%, respectively. However, the highest value for R/S DM was obtained from WS_{30}

Table 2. Effects of ultrasound (US) and water soaking (WS) treatments on shoot fresh weight (SFW), root fresh weight (RFW), shoot dry weight (SDW), root dry weight (RDW), shoot dry matter (SDM), root dry matter (RDM) of 14 days-old common vetch seedling.

Treatments	SFW (mg/plant)	RFW (mg/plant)	SDW (mg/plant)	RDW (mg/plant)	SDM (%)	RDM (%)
0-control-	0.107±0.006^{b}	0.119±0.007^{bc}	0.0115±0.0008^{ab}	0.0198±0.005^{b}	10.73±0.15^{ab}	16.47±0.97^{b}
WS_{10}	0.126±0.003^{a}	0.137±0.007^{ab}	0.0110±0.0007^{ab}	0.0228±0.001^{b}	8.83±0.06^{bc}	16.77±0.98 ^{b}
US_{10}	0.126±0.006^{a}	0.144±0.007^{a}	0.0110±0.0004^{ab}	0.0250±0.006^{ab}	8.95±0.88^{bc}	17.16±1.63 ^{b}
WS_{20}	0.084±0.009^{c}	0.120±0.004^{bc}	0.0085±0.0009^{bc}	0.0213±0.002^{b}	10.45±0.15^{abc}	17.68±0.48 ^{b}
US_{20}	0.116±0.001^{ab}	0.128±0.003^{abc}	0.0114±0.0009^{ab}	0.0230±0.001^{b}	9.82±0.18^{abc}	18.01±0.55 ^{b}
WS_{30}	0.082±0.001^{c}	0.107±0.007^{c}	0.0067±0.0009^{c}	0.0197±0.002^{b}	8.16±0.13^{c}	18.34±0.31 ^{b}
US_{30}	0.109±0.006^{ab}	0.127±0.005^{abc}	0.0087±0.0004^{bc}	0.0220±0.003^{b}	8.04±0.65^{c}	17.28±0.80 ^{b}
WS_{40}	0.119±0.003^{ab}	0.123±0.002^{abc}	0.0144±0.0040^{a}	0.0231±0.002^{b}	12.19±2.25^{a}	18.71±0.45 ^{b}
US_{40}	0.107±0.004^{b}	0.137±0.010^{ab}	0.0137±0.0032^{a}	0.0300±0.001^{a}	14.36±0.14^{a}	22.05±1.87^{a}
Summary of ANOVA						
Treatment	**	*	**	*	**	*

*significant at $P \leq 0.05$, **significant at $P \leq 0.01$. Different letters at the same column show significant differences at 0.05 level. ±: Standard errors.
Treatments*:
Different durations of two treatments: Seeds soaked in water (WS) or soaked in water in ultrasonic device (US).
Control: Seeds that were neither soaked in distilled water nor exposed to ultrasonic treatments.
US_{10}, US_{20}, US_{30}, and US_{40}: Seeds exposed to ultrasonic (ultrasound) treatments for 10, 20, 30, and 40 minutes, respectively.
WS_{10}, WS_{20}, WS_{30}, and WS_{40}: Seeds soaked in distilled water for 10, 20, 30 and 40 minutes respectively. It is considered a hydropriming treatment.

Table 3. Effects of ultrasound (US) and water soaking (WS) treatments on shoot dry matter (R/S DM), shoot water content (SWC), root water cantent (RWC), seedling vigor index (SVI) of 14 days-old common vetch seedling.

Treatments	**R/S DM (%)**	**SWC (%)**	**RWC (%)**	**SVI**
0-control-	1.55±0.30^{b}	89.26±0.54^{ab}	83.52±0.97^{a}	1845±213^{c}
WS_{10}	1.91±0.04^{ab}	91.16±0.96^{a}	83.22±0.98^{a}	1912±20^{ab}
US_{10}	1.93±0.12^{ab}	91.04±0.88^{a}	82.83±1.63^{a}	2303±153^{ab}
WS_{20}	1.80±0.54^{ab}	89.54±1.86^{ab}	82.31±0.48^{a}	1826±114^{c}
US_{20}	1.83±0.03^{ab}	90.17±0.18^{ab}	81.98±0.55^{a}	2036±106^{ab}
WS_{30}	2.24±0.11^{a}	91.83±0.13^{a}	81.65±0.31^{a}	1791±21^{c}
US_{30}	2.16±0.20^{a}	91.95±0.65^{a}	82.71±0.80^{a}	2409±200^{a}
WS_{40}	1.62±0.06^{b}	87.80±0.63^{bc}	81.28±0.45^{a}	2124±118^{ab}
US_{40}	1.61±0.04^{b}	85.63±1.06^{c}	77.94±1.87^{b}	2407±248^{a}
Summary of ANOVA				
Treatment	*	**	*	*

*significant at $P \leq 0.05$, **significant at $P \leq 0.01$. Different letters at the same column show significant differences at 0.05 level. ±: Standard errors.
Treatments*:
Different durations of two treatments: Seeds soaked in water (WS) or soaked in water in ultrasonic device (US).
Control: Seeds that were neither soaked in distilled water nor exposed to ultrasonic treatments.
US_{10}, US_{20}, US_{30}, and US_{40}: Seeds exposed to ultrasonic (ultrasound) treatments for 10, 20, 30, and 40 minutes, respectively.
WS_{10}, WS_{20}, WS_{30}, and WS_{40}: Seeds soaked in distilled water for 10, 20, 30 and 40 minutes respectively. It is considered a hydropriming treatment.

treatment with 2.24%. Abd El-Sattar and Tawfk (2023) reported that US treatments (40 kHz, 10 and 20 min) increased seedling dry matter of fenugreek. Alfalahi et al. (2022) stated that ultrasonic exposure increased seedling dry matter of the three soybean varieties. In addition, Shekari et al. (2015) mentioned that sesame seedling dry matter was significantly affected by the priming sonication technique.

As shown in *Table 3*, the effects of WS and US treatments on shoot water content (SWC) and root water content (RWC) were different. While WS and US treatments generally caused an increase in SWC (except, WS_{40} and US_{40}), RWC caused a decrease. In terms of RWC, the root was found to be more negatively affected by US treatments. In terms of SWC, it was determined that the shoot was slightly positively affected by WS and US treatments, but it was observed that there was no statistical difference between WS and US treatments.

However, the 40-minute application period, which can be considered as a high WS and US treatment, has been observed to have a negative effect on SWC. Researchers report that US treatments can have a positive effect in the short term as well as a negative effect in the long term. Ran et al. (2015) reported that ultrasound increase wheat seedling leaf relative water content.

The vigor index is a numerical measure that represents the energy and efficiency of seeds during the process of germination and growth of seedlings (Uslu and Gedik, 2020; Abd El-Sattar and Tawfk, 2023). The result of both WS and US treatment affects on seedling vigor index (SVI) are given in *Table 3*. According to these results, both WS and US treatments increased SVI values. It has been observed that this increase is greater in US treatments. The highest SVI value (2409) was recorded in US_{30} treatment. SVI germination percentage was calculated along with shoot and root growth. It has been determined that US_{30} treatment significantly increases the values in terms of germination percentage, shoot and root development. Nazari and Eteghadipour (2017) found that quick germination and the appearance of seedlings are crucial for the effective establishment of plants and commodities. They also discovered that gentle sonication can enhance seedling development. The results given in this study align with the conclusions reached by other researchers (Machikowa et al., 2013; Risca et al., 2007; Yaldagard et al., 2008; Kouchebagh et al., 2014; Shekari et al., 2015; Alfalahi et al., 2022; Memiş et al., 2022; Abd El-Sattar and Tawfk, 2023).

4. Conclusions

The data collected study suggest that the hypothesis, which proposed that exposure to ultrasound pre-treatments would results in breaking dormacy and a increase initial seedling growth, was supported by the experimental results. In summary, the present study found that using an appropriate ultrasonic therapy (namely, a mixed frequency of 47 kHz for a duration of both 10 and 30 minutes) enhances the germination and growth of common vetch. In addition, it has been observed that water soaking (WS) treatments, also considered hydropriming, have a positive effect on germination and growth parameters in general. However, when WS and US treatments were compared, it was found that US treatments had more positive effects. It has been observed that the root is more positively affected by US treatment than the shoot. Overall, the research results show that ultrasound/ultrasonic pre-treatment can have positive effects on the cultivation of common vetch, but it also reveals the need for further studies on whether these effects are economical.

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Ethical Statement

There is no need to obtain permission from the ethics committee for this study.

Conflicts of Interest

The authors declare that there are no conflicts of interest with respect to the research, authorship, and/or publication of this article.

Authorship Contribution Statement

Concept: Beyaz, R., Beyaz, A.; Design: Beyaz, R., Beyaz, A.; Data Collection or Processing: Beyaz, R.; Statistical Analyses: Beyaz, R.; Literature Search: Beyaz, R., Beyaz, A.; Writing, Review and Editing: Beyaz, R., Beyaz, A.

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