

Deciphering the Airborne Sounds of Plants Under Drought Stress

1. Abstract

Plant xylem vascular bundles are crucial for transporting water to the leaves. Research indicates that the tension within the vascular bundle's water-conducting system arises from the strong adherence of tiny air bubbles to the hydrophobic lignin domains of the xylem vessel wall. This adhesion, coupled with the sudden reorganization of these bubbles, is believed to generate the plant's acoustic emissions. Drawing from this understanding, it was hypothesized that the ultrasonic characteristics of plant roots would differ depending on the moisture levels within the roots. To explore this hypothesis, experiments were conducted using an ultrasonic analyzer and software, revealing significant variations in ultrasonic emissions correlated with the plant's moisture content.

The findings are particularly revealing: with adequate moisture, plants emit fewer sharp waves, characterized by a short wave width (approximately 7ms) and a high dBmax frequency (over 50KHz). In contrast, lower moisture levels lead to an increase in sharp waves, often containing 2-3 smaller spikes within a single wave, resulting in a longer spike width (about 16 ms) and a lower dBmax frequency (below 30 KHz). These insights are not just academically intriguing; they hold substantial practical applications, especially in developing precision irrigation systems.

Precision irrigation is a transformative tool that can reduce water waste by delivering water according to the specific needs of plants. This not only optimizes plant growth and

agricultural yield, but also contributes to sustainable water management in water-limited regions. This research advances our understanding of plant physiology and addresses pressing environmental issues in water-limited regions.

2. Research Motivation

The motivation behind this research is multifaceted, aiming to explore the intricate ways in which plants communicate and interact with their environment, particularly through sound waves. A significant aspect of this study is to understand how plants can emit ultrasonic waves in response to various stimuli, including the need for water. This understanding could revolutionize agricultural practices, especially in regions where precise irrigation is crucial due to water scarcity.

By deciphering the ultrasonic signals emitted by plants, I hope to develop technologies that allow for more efficient water use in agriculture. This is particularly relevant for arid and semi-arid regions where water resources are limited, and farming practices must be optimized to conserve every drop of water. The potential to use plant-emitted ultrasonic waves as a natural indicator of water needs could lead to the development of smart irrigation systems that only water crops when necessary, reducing waste and ensuring that plants receive the right amount of water at the right time.

3. Literature Review

The ability of organisms to communicate is a fundamental aspect of their survival and interaction with the environment. Animals, for instance, utilize their senses—such as hearing, sight, smell, taste, and touch—to receive external messages and employ various methods like chirping, vibrating, touching, flashing, emitting smells, and even using electrical currents to

transmit messages (Yen, 2013). In contrast, plants have developed their unique communication systems. They communicate with other plants through volatile organic compounds (Baldwin et al., 2006) and exchange messages via mycorrhizae, which form symbiotic relationships with fungi (Gilbert & Johnson, 2017).

Recent studies have shed light on the intriguing capability of plants to produce ultrasonic signals under stress conditions (Hadany, 2019). Further research indicates that plant cells possess mechanosensitive channels capable of detecting vibrations (Haswell et al., 2011), leading to physiological changes (Gagliano et al., 2012; Oda et al., 2021) or affecting gene expression (Ghosh et al., 2016). This raises a compelling question: Can plants use ultrasonic signals as a means of communication?

Exploring this phenomenon, it has been observed that plants under adverse conditions, such as drought stress or physical damage, emit ultrasound in the range of 20kHz to 150kHz. For example, tomatoes and tobacco have been recorded emitting 35 and 11 ultrasound waves per hour under drought conditions, respectively, and about 25 and 15 ultrasound waves per hour when physically damaged (Hadany, 2019). However, given that plants lack vocal organs like vocal cords or tubes, the mechanism behind their sound production is intriguing.

A significant breakthrough came from Ponomarenko et al. (2014), who discovered that the ultrasound emitted by plants during water deficit conditions is associated with air bubbles forming in the wood. They conducted experiments using fresh Scotch pine (*Pinus sylvestris* L.) wood samples placed in a hydrogel. As the water evaporated from the hydrogel, creating a negative pressure, air bubbles began to form in the xylem, captured by synchronized image analysis and ultrasound recordings. This correlation between ultrasound wave generation and air

bubble formation in the wood during cavitation—where air bubbles expand and collapse—suggests that this process could be a source of the vibrations producing these ultrasonic signals.

4. Hypothesis

The hypothesis suggests that ultrasonic emissions from plant roots display unique characteristics under different water availability conditions. It is predicted that the number of ultrasonic spikes, their width, and the maximum dB frequency emitted by potted plants will show significant differences between well-watered and water-deficient scenarios. These ultrasonic signatures could form the basis for designing an innovative irrigation system tailored to the specific water needs of plants, thereby improving water efficiency in agricultural practices.

5. Methodology

5.1 Instruments for Environmental Measurements

Fig.1



Fig. 1 Potting Soil Moisture

Fig. 2



Fig. 2 Wind Speed Gauge for Outdoor Velocity Measurement

Initially, a wind speed gauge was used to measure outdoor velocity before acquiring the acrylic cover.

5.2 Detection and Analysis of Ultrasonic Features



Fig. 3 SONAPHONE



Fig. 4 Acrylic cover

In this section, the SONAPHONE ultrasonic testing device and self-designed acrylic cover are included.

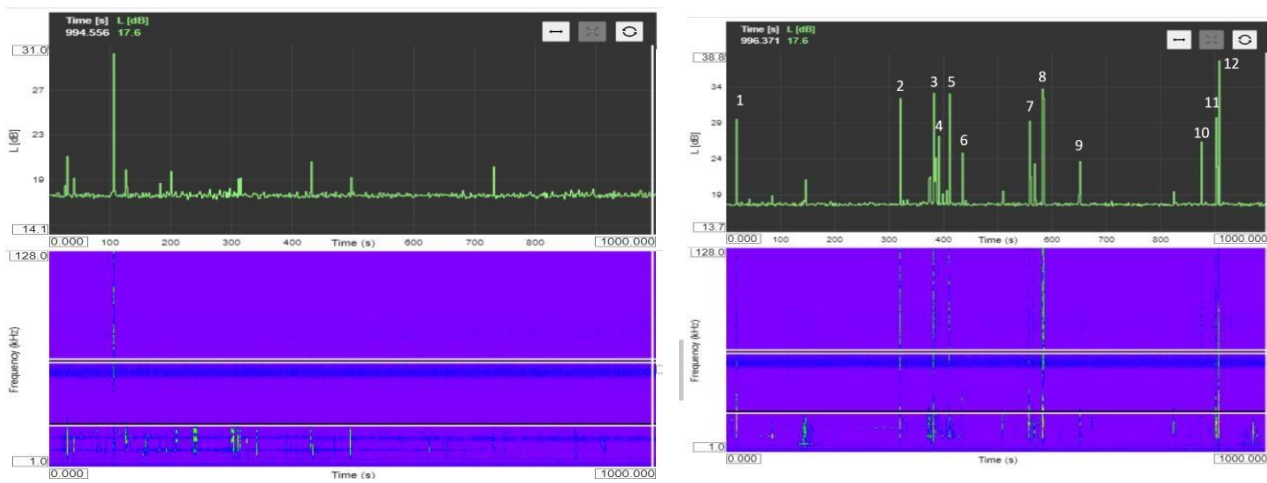


Fig. 5 Analyze waveform and frequency changes to identify spikes with Sonaphone

Using Sonaphone to analyze waveform and frequency changes over 1000 seconds to identify peaks. When plants are exposed to drought, air bubbles form in the xylem where they expand and collapse. This produces vibrations and the whole process is called cavitation.

6. Experimental Framework and Methodology

Experimental Design

Analysis of Ultrasonic Signal Patterns Based on Regular Watering and Drought Conditions. Procedure for Minimizing Interference During Signal Recording and Analysis

The experimental procedure for the plant species *Ixora* is as follows.

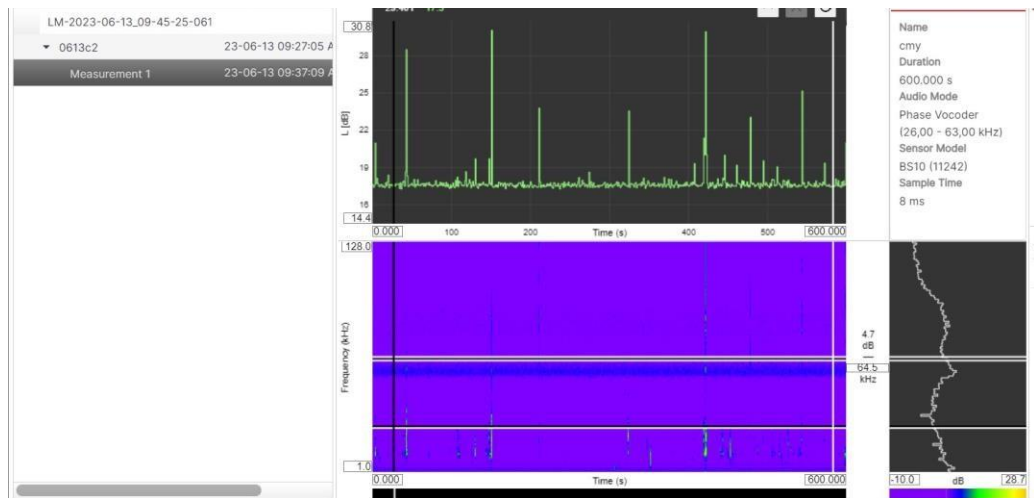


Fig. 6 10-minute recording of ultrasonic spike signal in SONAPHONE DataSuite

1. Water Plant
2. Collection of a 10-Minute Ultrasonic Signal Using the Testing Device 1 hour after watering and 9 hours after watering
3. Interpretation and Comparative Analysis of Test Signals

Note: Conduct tests during periods of minimal environmental and artificial noise to significantly reduce the likelihood of interference, thereby enhancing data quality and accuracy.

After initially watering the plants on the first morning, watering was halted, and ultrasonic tests were conducted at 1 hour after watering and 9 hours after watering. This was to verify

that the sounds produced by watered plants differ from those produced by droughted plants.

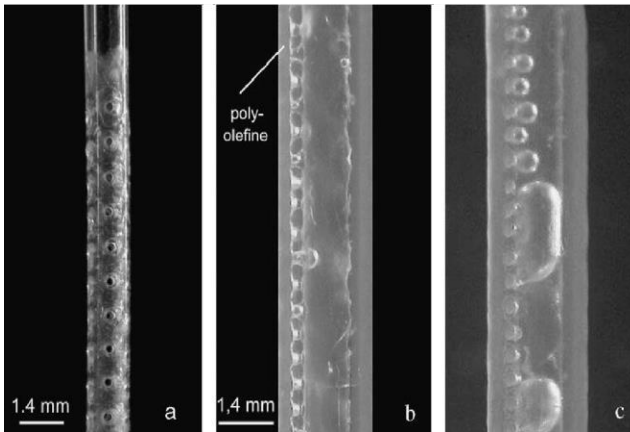


Fig 7 Xylem cavitation in woody parts

<https://plos.figshare.com/articles/figure/>

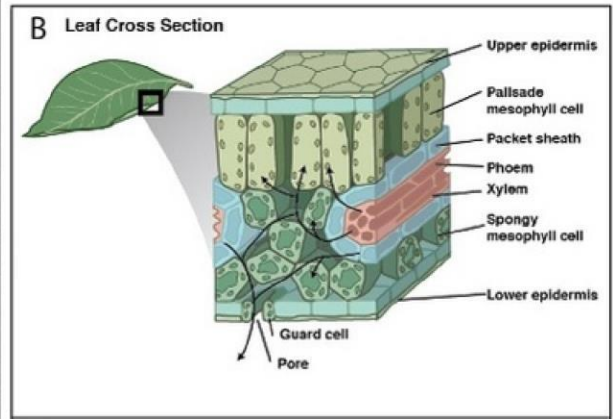


Fig 8 Leaf Cross Section

<https://www.nature.com/scitable/knowledge/>

Experiment Setup



Fig. 9 Experiment setup surrounded by acrylic cover to minimize interference

In the experimental setup, an acrylic cover was utilized to insulate the testing environment from external auditory disturbances, including wind, rain, avian vocalizations, and

other forms of noise. This measure ensures the integrity of the sound data by minimizing interference.

7. Research Outcomes

After conducting countless experimental tests, I will use one as an example.

Experimental Test 1: Jun 14th, Watered at 7:00 am and tested at 8:17 am

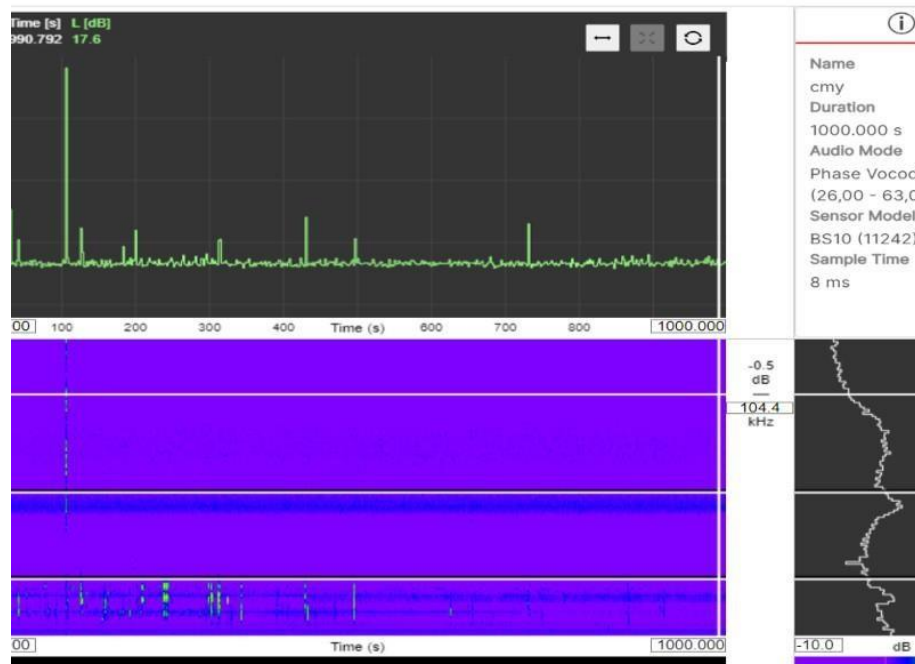


Fig. 10 Post-Watering: Simple, High-Frequency Wave

After watering, the sharp wave is simple but with a higher frequency , dBmax = 104.8 KHz (17.1dB) , frequency dBmax = 73.7KHz (17.1dB)

Experimental Test 2: After watering at 7:00 a.m. and testing at 16:17 p.m.

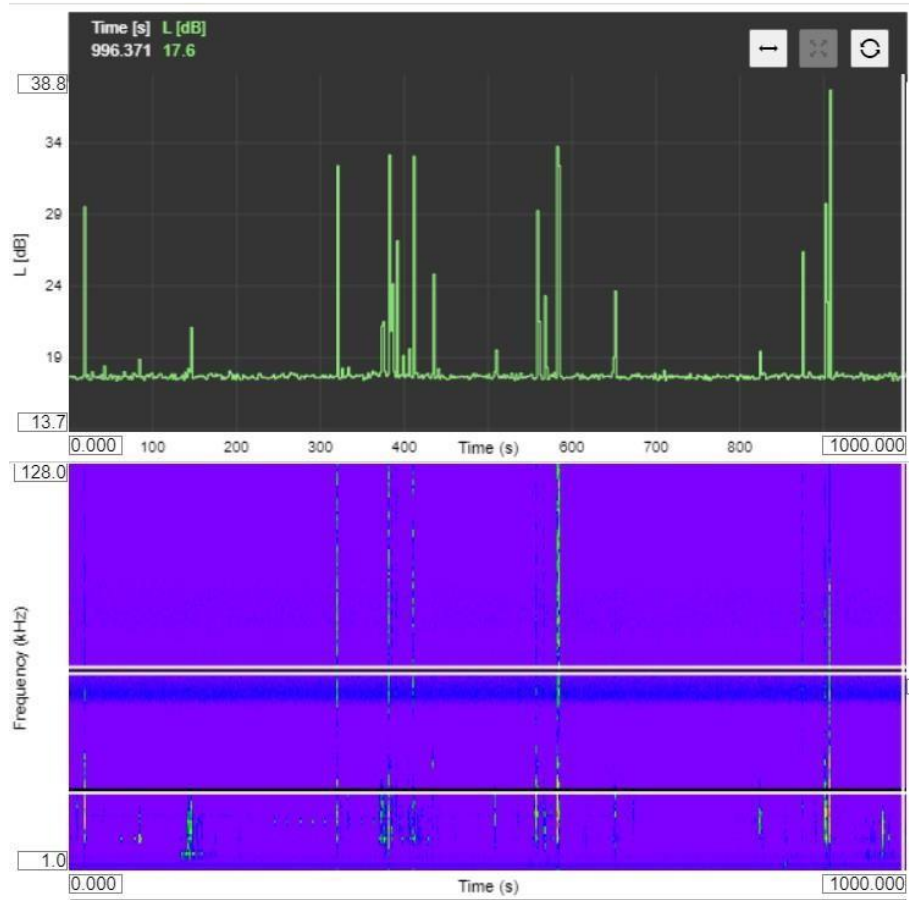


Fig. 11 Dry Conditions & Cavitation: Complex Sharp Slopes

Airborne sounds are more complicated due to the lack of water.

Experimental Test 3 : Test at 8:17 am after watering at 7:00am

Fig 12 Waveform Analysis

Appearance time	total wave dBmax	# of small spikes	Total wave Width ms	wave dBmax frequency KHZ		wave dBmax freq. KHZ	
				dB	KHZ	dB	KHZ
105.67	31	1	7	17.1	73.7	17.1	104.8

Small spike width, high spike dBmax frequency > 60KHZ. Experimental

Test 4 : Tests at 16:17 after watering at 7:00

Fig. 13 Overview of Experiment Data 1

wave number	length of appearance (s)	total wave dBmax	# of small spikes	length of small spike appearance (s)				total wave width (ms)	a wave width ms		a wave dBmax freq. KHZ		b wave width ms		b wave dBmax freq. KHZ		c wave width ms		c wave dBmax freq. KHZ		d wave width ms		d wave dBmax freq. KHZ	
				a	b	c	d		dB	ms	dB	KHZ	dB	ms	dB	KHZ	dB	ms	dB	KHZ	dB	ms	dB	KHZ
				1	18.92	29.6	3		18.9	19.4	19.53	19.6	676.1	23.6	54.5	12.9	29.7	26.9	17	24.1	18.4	24.3	14.1	23.6
2	320.4	32.4	3	320.4	320.5	320.53		138.9	32.4	7.6	20.9	28.3	21.6	7.6	14.5	21.7	26.7	7.5	19.9	28.3				
3	381.8	33.2	3	381.8	382.1	385.8		3998	26.1	8.2	18	11.3	33.2	8.2	21.1	18.8	24.1	7.5	14.4	14.1				
4	390.4	27.1	1	390.4				17.3	27.1	16.5	11.3	88.5												
5	410.9	33.1	1	410.9				8.5	33.1	8.5	18.1	58.8												
6	435.2	24.8	1	435.2				7.8	24.7	7.8	16.5	34.4												
7	558.6	29.3	1	558.6				15.1	29.3	15.1	27.4	19.8												
8	581.8	33.8	3	581.8	581.9	582.9		1142	29.5	70	28.6	19.3	33.8	33.1	35.4	15.1	32.4	17.7	18.8	79.1				
9	651.2	23.6	1	651.2				7.9	23.6	7.9	17.2	18.8												
10	873.1	26.4	1	873.1				8.6	26.4	8.6	12.7	21.7												
11	900.4	29.8	4	900.4	900.45	900.5	900.57	168.7	22.7	8.5	14.3	19.8	22.7	7.8	20.2	15.6	23.2	14.9	11.8	15.1	29.7	14.9	28.3	19.8
12	906.1	38.8	4	906.1	906.68	907.18	907.39	1327.2	22.2	8	22.6	11.3	37.7	32.1	29.6	21.7	25.6	7.7	22.3	11.3	23.5	16.6	22.8	19.8

Fig. 14 Overview of Experiment Data 2

Number	Length of Appearance(s)	Travel wave Width(ms)	dBmax	dBmax (KHZ)
1a	18.9	54.5	23.6	29.7
1b	19.4	17	26.9	18.4
1c	19.53	14.1	24.3	22.1
1d	19.6	13.7	29.6	15.6
2a	320.4	7.6	32.4	28.3
2b	320.5	7.6	21.6	21.7
2c	320.53	7.5	26.7	28.3
3a	381.8	8.2	26.1	11.3
3b	382.1	8.2	33.2	18.8
3c	385.8	7.5	24.1	14.1
4	390.4	16.5	27.1	88.5
5	410.9	8.5	33.1	58.8
6	435.2	7.8	24.8	34.4
7	558.6	15.1	29.3	19.8
8a	581.8	70	29.5	19.3

8b	581.9	33.1	33.8	15.1
8c	582.9	17.7	32.4	79.1
9	651.2	7.9	23.6	18.8
10	873.1	8.6	26.4	21.7
11a	900.4	8.5	22.7	19.8
11b	900.45	7.8	22.7	15.6
11c	900.5	14.9	23.2	15.1
11d	900.57	14.9	29.7	19.8
12a	906.1	8	22.2	11.3
12b	906.68	32.1	37.7	21.7
12c	907.18	7.7	25.6	11.3
12d	907.39	16.6	23.5	19.8
Average		16.36ms	27.25dB	25.86KHZ

This data is zoomed in in Figure 15, showing the wave width, maximum dB, and maximum dB frequency of each sharp wave. The twelve sharp waves observed had an average width of 16.36 milliseconds, a peak decibel level of 27.25 dB, and their highest frequency reached 25.86 kHz.

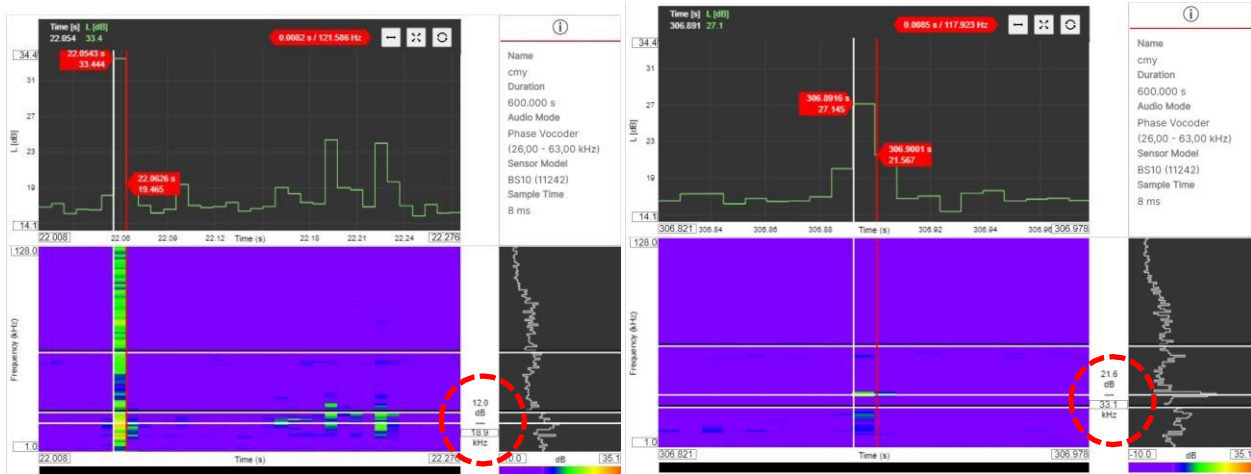


Fig. 15

18.9

Fig.16

33.1

Fig. 15 Spike Analysis 1

dB_{max} Frequency = 18.9kHz, wave width 8.3ms.

Fig. 16 Spike Analysis 2

dB_{max} Frequency = 33.1kHz, sharp wave width 8.5ms.

When the plant is well-watered, the number of sharp waves emitted is fewer, and the sound lasts shorter, with a width of around 7ms. Additionally, the dB_{max} frequency is higher than

(>50KHz). When the plant has less water, the number of sharp waves emitted will be more, and a single sharp wave may contain 2-3 smaller sharp waves, resulting in a sharp wave width of about 16ms, and the dBmax frequency is lower (<30KHz). This data is helpful for the future design of precision irrigation systems.

Conclusion

The comprehensive analysis across multiple experiments and conditions underscored significant findings regarding plant responses to water availability. When adequately watered, plants emitted fewer sharp waves with short widths (about 7ms) and high dBmax frequencies (over 50kHz). Conversely, water-stressed plants produced more sharp waves, including 2-3 smaller spikes within a single wave, leading to longer wave widths (about 16ms) and lower dBmax frequencies (below 30kHz). These results not only confirm our initial hypothesis but also highlight the potential of using plant's airborne sounds as a novel approach to monitor plant drought stress.

This research lays foundational work for future studies aimed at developing precision irrigation systems. By harnessing the nuanced responses of plants to water stress, as evidenced through ultrasonic emissions, we can move towards more sustainable and efficient agricultural practices. The ultimate goal is to tailor watering strategies that meet the specific needs of plants, reducing water waste and enhancing crop yield in environments prone to water scarcity

References

- Baldwin IT, Halitschke R, Paschold A, von Dahl CC, Preston CA. Volatile signaling in plant-plant interactions: "talking trees " in the genomics era. *science*. 2006;311:812-5. doi: 10.1126/science.1118446.
- Dutta, S., Chen, Z., Kaiser, E., Matamoros, P. M., Steeneken, P. G., & Verbiest, G. J. (2022). Ultrasound pulse emission spectroscopy method to characterize xylem conduits in plant stems. *research*.
- Gilbert, L., & Johnson, D. (2017). Plant-plant communication through common mycorrhizal networks. in *Advances in botanical research* (Vol. 82, pp. 83-97). Academic Press.
- Ghosh, R., Mishra, R. C., Choi, B., Kwon, Y. S., Bae, D. W., Park, S. C., ... & Bae, H. (2016). Exposure to sound vibrations lead to transcriptomic, proteomic and hormonal changes in *Arabidopsis*. *scientific reports*, 6(1), 33370.
- Haswell, E. S., Phillips, R., & Rees, D. C. (2011). Mechanosensitive channels: what can they do and how do they do it? *Structure*, 19(10), 1356-1369.
- Heffner, H. E., & Heffner, R. S. (2007). Hearing ranges of laboratory animals. *Journal of the American Association for Laboratory Animal Science*, 46(1), 20-22.
- Joshi, J., Stocker, B. D., Hofhansl, F., Zhou, S., Dieckmann, U., & Prentice, I. C. (2022). Towards a unified theory of plant photosynthesis and hydraulics. *Nature Plants*, 8(11), 1304-1316.
- Website: <https://www.nature.com/articles/s41477-022-01244-5>
- Khait, I., Lewin-Epstein, O., Sharon, R., Saban, K., Perelman, R., Boonman, A., ... & Hadany, L. (2018). Plants emit informative airborne sounds under stress. *bioRxiv*, 507590.
- Website: <https://www.sci.news/biology/plants-ultrasonic-clicks-07895.html>

Khait, I., Lewin-Epstein, O., Sharon, R., Saban, K., Goldstein, R., Anikster, Y., ... & Hadany, L.

(2023). Sounds emitted by plants under stress are airborne and informative. *cell*, 186(7), 1328-1336.

McElrone, A. J., Choat, B., Gambetta, G. A., & Brodersen, C. R. (2013). Water uptake and transport in vascular plants. *Nature Education Knowledge*, 4(5), 6.

Website: <https://www.nature.com/scitable/knowledge/library/water-uptake-and-transport-in-vascular-plants-103016037/>

M. Zhang and J. H. M. Willison, "Electrical impedance analysis in plant tissues: the effect of freeze-thaw injury on the electrical properties of potato tuber and carrot root tissues," *Canadian Journal of Plant Science*, vol. 72, pp. 545-553, 1992.

Oda, S., Sakaguchi, M., Yang, X., Liu, Q., Iwasaki, K., & Nishibayashi, K. (2021). Ultrasonic treatment suppresses ethylene signaling and prolongs the freshness of spinach. *Food Chemistry: Molecular Sciences*, 2, 100026.

Ponomarenko, A., Vincent, O., Pietriga, A., Cochard, H., Badel, É., & Marmottant, P. (2014). Ultrasonic emissions reveal individual cavitation bubbles in water-stressed wood. *Journal of the Royal Society Interface*, 11(99), 20140480.

Rafał Chudy, 2016, Trees call for water when they are thirsty.

Website: <https://www.forest-monitor.com/en/trees-call-for-water-when-they-are-thirsty/>

Y.Z. Lin, 1982, Research on the new technology of stomatal opening and closing and its application, National Elementary and Middle School Science Exhibition, Biology First Place, Taiwan Network Science Museum

Website: <https://www.ntsec.edu.tw/science/detail.aspx?a=21&cat=7128&sid=7719&print=1>

Chan, L. S.. (2013). Evapotranspiration in the growing environment of crops. Special issue of
Taichung Agricultural Improvement Center, 109-114.

Website: https://www.tcdares.gov.tw/upload/tdais/files/web_structure/8187/TC0211619.pdf

Yim Hongyang, 2013, Biological Signal Communication Pipeline - The Negative Effect of
Human Interference, Science and Technology Grand Park

Website: [https://scitechvista.nat.gov.tw/Article/C000009/detail?ID=ec4fb250-6b40-4c0d-9910-
dee8a9e8fec8](https://scitechvista.nat.gov.tw/Article/C000009/detail?ID=ec4fb250-6b40-4c0d-9910-dee8a9e8fec8)