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# WHEN THE LAB GETS LOUD: TESTING NOISE EFFECT OF PLANTS

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## ABSTRACT

Noise pollution is increasing in both scope and intensity due to the growth of the human population and urban development, significantly impacting terrestrial and aquatic habitats. Anthropogenic noise is pervasive in nature and has been shown to affect a wide range of animal taxa. Recent studies, however, reveal that noise and vibrations can also influence plants, altering their morphological, physiological, and genetic traits. This suggests that noise pollution may exert effects on ecosystems at more complex levels than previously understood. In this study, we investigated the effects of broadband noise (pink noise) on two plant species (one herbaceous and one tree species) to explore its potential impact on vegetation. Laboratory experiments were conducted under controlled conditions at the Department of Earth and Environmental Sciences, University of Milano-Bicocca, Italy. Seeds were planted in soil-filled pots and placed within two phytotrons -one designated as the treatment chamber, where pink noise was continuously emitted through a speaker, and the other as the control chamber, with no noise exposure. We assessed whether noise influenced germination rates, growth and survival of plants. Preliminary results show an effect on germination and development of the herbaceous species.

**Keywords:** *anthropogenic noise, laboratory experiment, plant, soil, microbiome.*

## 1. INTRODUCTION

The rapid expansion of urban areas has led to a significant increase in noise pollution, which now poses a serious threat to both terrestrial and aquatic ecosystems [1]. Among the various sources of anthropogenic noise, traffic and industrial activities are particularly disruptive to biodiversity [2–3]. While noise pollution is widely recognized as an environmental stressor, its impact extends beyond animals to affecting a broad range of organisms [4]. Previous research has extensively documented how noise interferes with animal communication, reproduction, and spatial behaviors [1–5–6].

More recent findings indicate that plants, too, can perceive and respond to sound stimuli [7–8]. Unlike animals, plants are immobile and must endure environmental stressors, relying on complex signaling networks and adaptive mechanisms to cope with fluctuating conditions [9]. Studies suggest that exposure to certain sound frequencies can alter seed germination, plant growth, oxidative stress levels, and the expression of stress-related genes [8]. Prolonged noise exposure has also been linked to reductions in photosynthetic and transpiration rates, ultimately affecting plant development and survival, particularly in urban environments [10–11].

Beyond physiological changes, noise pollution may also interfere with ecological interactions, such as plant-pollinator relationships. Excessive noise can disrupt pollinators' ability to detect floral cues, which may negatively impact pollination success and, consequently, plant reproduction [10].

Despite growing concern about noise pollution, most studies have focused on specific sound frequencies (e.g., 0.4kHz, 1kHz, 5kHz), limiting our understanding of the effects of broader noise exposure [12] as road traffic noise. Research on noise-induced changes in plant physiology is still scarce, with only one study examining how traffic noise affects urban vegetation [13]. Given the increasing presence of anthropogenic noise across landscapes, further research is needed to assess its potential effects on plants [14].

This study investigates the impact of broadband noise (pink noise) on an herbaceous and a tree species to evaluate how noise pollution may influence plant-microbe interactions and overall ecosystem dynamics.

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## 2. MATERIAL AND METHODS

### 2.1 Species studied

This study focused on two species, one herbaceous, *Trifolium pratense L.* and one woody *Ulmus minor L.* The first one, commonly known as red clover, is a temperate perennial herb commonly growing wild in meadows throughout Europe and Asia. The second one is also widely planted in both countryside and urban areas, across almost all Europe. Both species are known for their tolerance to adverse environmental conditions and stress [15–16].

### 2.2 Experimental design

The study took place in 2023 at the University of Milan-Bicocca, Italy, in two phytotrons (growth chambers - Sanyo, 180Hx80Lx55W), maintaining a temperature of 25°C and a day/night cycle from 9 a.m. to 9 p.m. (light) and 9 p.m. to 9 a.m. (dark). The experiment was conducted using a speaker (JBL Control 1 Pro) placed on the top shelf X (Fig.1) emitting pink noise continuously for 24 hours (stimulus constructed using Audacity software). To mimic natural conditions, fluorescent lights alternated between 12-hour light/dark cycles. Seeds of *T. pratense* and *U. minor*, certified by research institutions, were sown in 24 soil-filled pots (12 per species, 50 seeds per pot). The pots were placed in shelfs Y and Z (Fig.1).

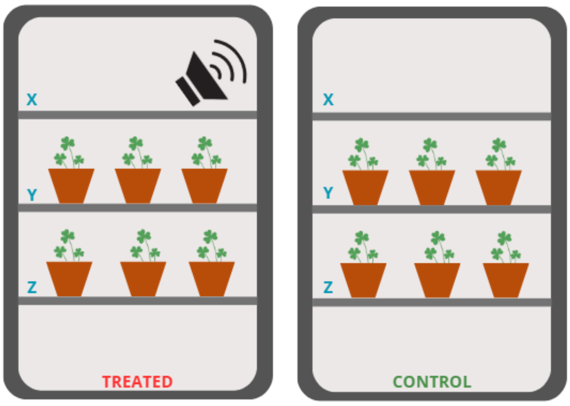


**Figure 1**. Treat phytotron (left) with a pink noise signal continuously emitted by a loudspeaker in shelf X, 24h a day and control phytotron (right) with no noise.

### 2.3 Acoustic data collection and analysis

Acoustic data collection was carried out using a Class 1 calibrated sound level meter (SLM -Larson Davis - 831C) equipped with a wired microphone, allowing precise placement inside the phytotron to record noise levels. To characterize the noise perceived by the plants both background and treatment noise levels were measured. The growth chamber was divided into four sections by three horizontal shelves: the speaker was placed on the first shelf (Figure 1), while the plants were positioned on the second and third shelves (three pots per species per shelf). Nine measurements were taken for each shelf (column A, B, C, row 1, 2, 3 - see Figure 2), following a regular grid in the treated and control conditions, with lights on/off, totaling 36 measurements per phytotron. Each recording lasted one minute, with a sampling rate of 48.000 Hz.
The acoustic data collected was extracted from the SLM and analyzed using its corresponding software, BZ5503 and Evaluator 7820. Data exported included LeqZ 1/3 octave bands . Sound pressure levels were measured at each points to characterize levels of noise arriving to the plants for both treatment and control phytotrones.

### 2.4 Acoustic data collection and analysis

The following plant morpho-functional traits were measured for both species: total and thorough time emergence (%), survival and growth (in terms of plant height, number and area of leaves).

### 2.5 Statistical analysis

We performed a linear mixed-effects model (LMM) using treatment, light condition and shelf as fixed factors and dB level as response variable. Data exploration followed the protocol described in [17]. We ran the models using packages "lme4" [18]. Visual analysis and figures were computed with R Studio (RStudio). Regarding biological parameters, univariate analyses (ANOVA and SNK tests) were conducted with GMAV software to analyze data regarding the morpho-functional traits considered for the two species and the bacterial abundances.

## 3. RESULTS

### 3.1 Acoustic characterization of the phytotrons

The Linear Mixed Model (LMM) analysis (Fig.2; Tab.1) revealed that background noise levels (dBZ) in treated phytotron were higher compared to control ($p < 0.001$).

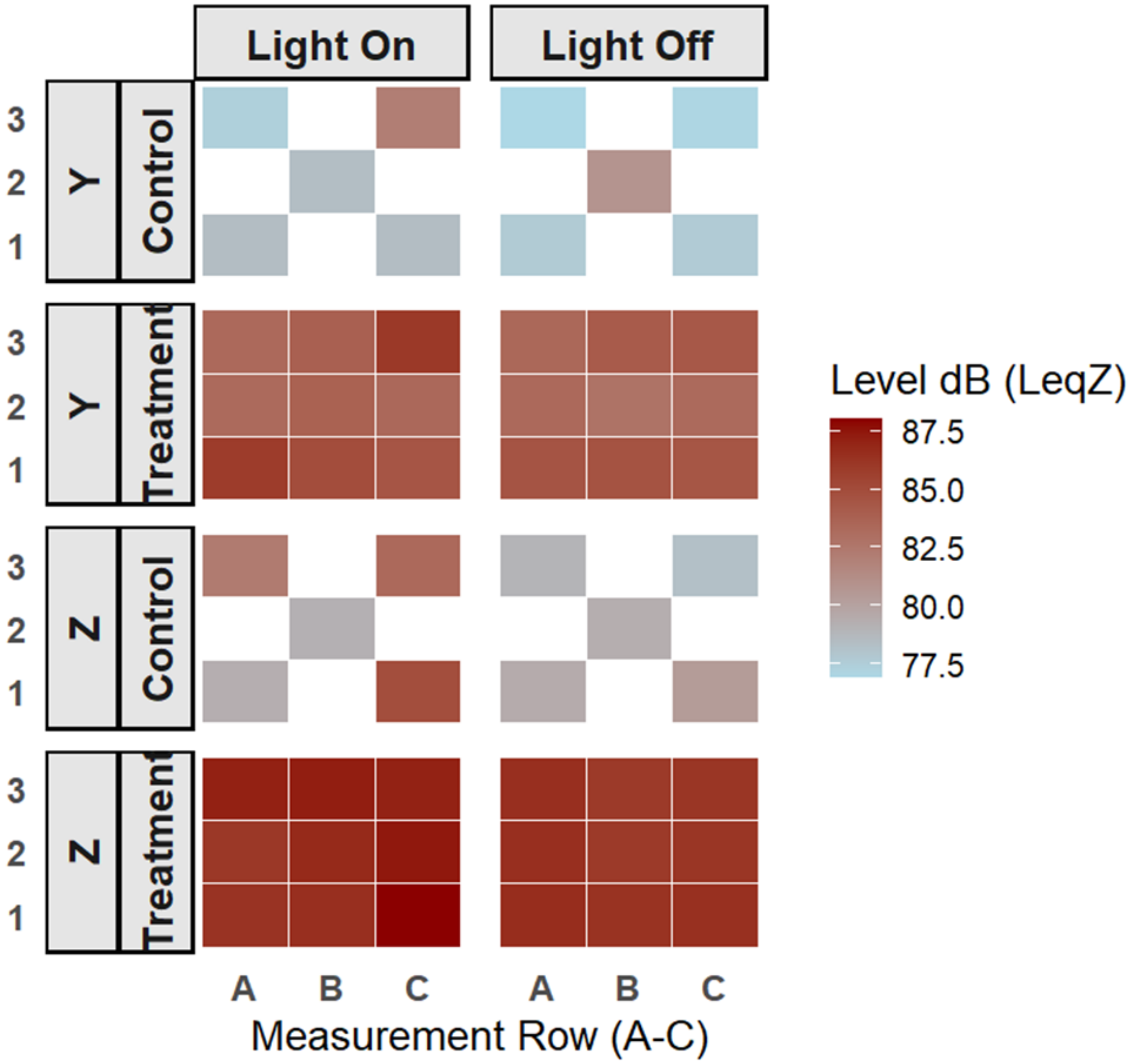


**Figure 2.** Heatmap of Noise Level dB (Z) by measurement site per shelf per treatment under light on/off condition.

**Table 1.** Summary of the Generalized Linear Mixed Model. Baseline reference: control, light off, shelf Y.

| Factor | Estimate | SE | df | t-value | p-value |
|---|---|---|---|---|---|
| Intercept | 78.9 | 0.55 | 28.45 | 143.90 | <.0001 |
| Treatment | 5.37 | 0.636 | 44.4 | 8.453 | <.0001 |
| Light On /Off | -0.95 | 0.720 | 43.9 | -1.313 | 0.196 |
| Shelf (Y/Z) | 2.83 | 0.720 | 43.9 | 3.934 | <.0001 |
| Treatment × Light | 0.50 | 0.89 | 43.9 | 0.559 | 0.579 |
| Treatment × Shelf | -0.23 | 0.89 | 43.9 | -0.257 | 0.798 |
| Light × Shelf | -1.59 | 1.018 | 43.9 | -1.529 | 0.133 |
| Treatment × Light × Shelf | 1.36 | 1.27 | 43.9 | 1.077 | 0.288 |

Furthermore, the shelf position also influenced the level's noise with the higher levels in Z respect to Y ($p < 0.001$). On the contrary, the light condition, although a bit higher in the light condition, was not significantly different ($p = 0.196$). Regarding interactions, no significant two-way or three-way interactions were found (p-values > 0.05). This suggests that the effects of Treatment, Light, and Shelf Position were largely independent rather than influencing each other. These differences in noise distribution between shelves and treatments could influence plant physiological responses. For this reason, the pots were rotated within the phytotron throughout the experiment to ensure equal exposure to noise.

### 3.2 Effect of noise treatment on plant species

The effect of the treatment exposure on the target species showed that noise negatively affected the germination of *U. minor*, causing a significant delay and a reduction in the number of germinated seeds compared to the control. In contrast, *T. pratense* showed no significant differences between the two conditions. At the end of the experiment, no significant differences in plant survival were observed between species and treatments, although *U. minor* showed slightly lower values under noise exposure. However, with regard to the growth, *U. minor* displayed significantly lower values for the height, the leaf number and area under noise exposure. In contrast, for *T. pratense* no significant variations between the noise exposure and the control treatment were observed.

## 4. DISCUSSION

The results from the acoustic characterization highlight key factors for improving experimental design when studying noise effects on plants. As expected, the treated phytotrons had significantly higher noise levels than the controls. However, the shelf position also influenced the noise distribution by having higher levels observed on the lower shelf. Meanwhile, light conditions showed no significant effect. The absence of significant interactions indicates that treatment and shelf position acted independently. These findings emphasize the need to account for spatial noise variability. Rotating pots, as done in this study, helps ensure equal exposure, reducing bias. Future experiments should integrate such controls to improve reproducibility and isolate noise effects more effectively.
Our results showed an effect of noise mainly on one of the considered plants, *Ulmus minor*, suggesting that species-specific responses to noise occur. This supports previous findings by [19], who observed varied plant reactions to different sound frequencies, with noise having both negative and, in some cases, positive effects on growth. Similar to other environmental factors like moisture, light, wind, and temperature, plant responses to noise appear to be primarily physiological and dependent on species-specific traits and functional groups [20].

## 5. CONCLUSION

Our results bring two important conclusions regarding both experimental design and noise impact on plants:

- Phytotrons represent a good environment for testing the effect of noise on plants, provided pot rotation is considered, as noise distribution is not homogeneous.
- Noise had an effect on plants, particularly on *Ulmus minor*, suggesting species-specific responses. These findings confirm previous studies and indicate that plant responses to noise, as well as to other environmental factors, depend on the species' physiological and functional traits.

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research structures and creation of R&D "innovation ecosystems", 657 set up of "territorial leaders in R&D.

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