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The Effect of Audible Sound on the Germination and Root Elongation of Oats and Wheat

Ferne George Ellis
Western Michigan University

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THE EFFECT OF AUDIBLE SOUND ON THE GERMINATION
AND ROOT ELONGATION OF OATS AND WHEAT

by
Ferne George Ellis

A Thesis
Submitted to the
Faculty of The Graduate College
in partial fulfillment
of the
Degree of Master of Arts

Western Michigan University
Kalamazoo, Michigan
August 1973
ACKNOWLEDGEMENTS

I deeply appreciate the advice and guidance in all aspects of this work of Dr. Leo C. Vander Beek and of the other members of my thesis committee, Dr. Richard W. Pippen and Dr. Eugene Bernstein.

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Finally, an affectionate word of appreciation to my wife, Theresa, for countless hours as my secretary extraordinaire throughout this work as well as all my graduate days.

Ferne George Ellis
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Physiology
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The Effect of Audible Sound on the Germination and Root Elongation of Oats and Wheat

INTRODUCTION

The sound that surrounds all living organisms, except in unusual or experimental circumstances, is divided into two general areas, audible and ultrasonic. This classification is based upon the average man's ability to hear sound: sounds ranging from 20 to about 15,000 Hertz (Hz) are considered audible and those above 15,000 Hz are considered ultrasonic.

Sound is measured in Hertz or cycles per second and intensity of sound is expressed in decibels (db) or force per area. The reference value taken in computing decibels is the minimum intensity the ear can detect in air or $2 \times 10^{-4}$ dynes/cm² (sound pressure).

Most experiments reported in the literature involve the ultrasonic region where energy levels are higher. Ultrasound owes its destructive effectiveness in large part to the cavitation of water (Liu and Yen, 1934). Microflows created at the liquid-cell boundary and within cells can also have destructive effects (El'Piner, 1970).

Experiments with audible sound reported in the literature generally involved animals having hearing systems; plant experimentation generally concerns germination and growth.
This work was initiated to examine the effects of audible sound on the germination and growth of oats and wheat.
LITERATURE REVIEW

Most of the work on the effects of audible sound on animals involves animals with hearing systems. Koitchev (1969) found that sodium distribution changed in the organ of Corti under conditions of relative rest and after exposure to sound. The increase in sodium was attributed to possible changes in the permeability of the excited membranes. Aleksandrovskaia and Chezhenkova (1970) found that the rabbit's cerebral cortex under sound exposure had enhanced slow waves and spindles, particularly in the motor area. They also found that there was an increase in astrocytes in the deep layers of the motor area but a decrease in the deep layers of the projection area (auditory cortex).

Anichen (1968) found that in the hair cell nuclei of the organ of Corti, sound treatment caused the nuclei number to increase and the RNA bundles to be more compact. No effects were noted in the nuclear volume or content of Nissl substances in the nerve cells of the cochlear nuclei of the guinea pig using four different frequencies (Wustenfeld et al., 1970). Sviderskaya (1968) studied the effects of two frequencies on the motor activity of chicks. Different frequencies gave different responses and the degree of response increased after the auditory systems of the chicks had developed.

The effects of audible sound on plants and plant cells has been examined in a number of different ways. Studies by Northen
and Mac Vicar (1939) showed that sound lowered the elasticity of the cytoplasm of *Spirogyra* by 20 per cent to 64 per cent. Higher rates of photosynthesis for plants exposed to sound have been reported by Gnanam (1959) in *Spirogyra*, and by Singh (1959) for the water plant *Hydrilla verticillata*.

Popular magazines and daily newspapers frequently feature reports which concern the effect of music or the spoken word on plants. Some reference to such effects have been recorded in scientific literature. Ponniah (1958) played a single note to *Mimosa pudica*, *Impatiens balsamina*, *Tagetes erecta*, and *Hydrilla verticillata* and found an increase in growth. Singh and Ponniah (1954, 1955) showed that plants exposed to musical tunes had an increase in growth in all parameters measured.

Lisenkov (1966) found that seeds exposed to "sound treated water" had an increase in "ground germination" (quotation marks are mine), in frost resistance and better growth. Amylase activity increased in those seeds exposed to the sound for periods between 0.5 and 1.0 hours. Longer treatment decreased enzyme activity.

The effects of a single frequency at any given time on the growth of wheat reveal differences between varieties of wheat, frequency used, and time of exposure. Weinberger and Measures (1968) using spring and winter wheats (*Triticum aestivum*) found an increase in germination and better growth in winter wheat (var. *Rideau*) and in some cases temperature dependency or frequency dependency was observed. With spring wheat (var. *Marquis*)
germination was increased at 2°C and 10°C but not at 25°C. The growth response of the spring wheat was dependent upon treatment.

The same workers in a modification of the above experiment using spring wheat noted a significant increase in some of the parameters measured when 300 Hz and 5,000 Hz were employed (Measures and Weinberger, 1970).

While most of the literature reports positive (or stimulative) growth effects, some negative effects have been reported. Weinberger and Das (1972) using synchronized cultures of Scenedesmus obtusiusculus found that 4,000 Hz produced a decrease in the rate of cell division and that after treatment normal cell division did not return until two life cycles had elapsed. Other negative effects were found by Woodlief et al., (1969) in tobacco plants subjected to random noise. These plants showed a decline in the rate of growth by over 40 per cent.
METHODS AND MATERIALS

Plant Material

The two types of certified grains used in these experiments, oats (Avena sativa var. AuSable) and wheat (Triticum aestivum var. Ionia) were purchased from Farm Bureau Services Inc., Kalamazoo, Michigan. All seeds were stored during the course of the experiments in a refrigerator at about 8°C.

Sound Source

Sound at a frequency of 300 Hz was generated by an audio-oscillator\(^1\) connected to an amplifier\(^2\). The sound intensity was measured by means of a sound level meter\(^3\) using the 'C' weight. The audio-oscillator was then adjusted to give 100 ± 3 db (1 \times 10^{-2} dynes/cm\(^2\)) in each of two growth chambers. Background from all frequencies was 82 ± 3 db in each of the growth chambers. A nine inch speaker was used in all experiments suspended by heavy string near the center of the chamber. Using a system of hooks, the speaker could be removed and returned to the same position in either chamber.

\(^{1}\)Audio-oscillator made by the Hewlett Packard Co., Pala Alto, California (Model 200D).

\(^{2}\)Amplifier made by the David Bogen Co., New York, New York (Model E14).

\(^{3}\)Sound level meter made by the General Radio Co., West Concord, Massachusetts.
Growth Conditions

Both control series and experimental series consisted of 5 plastic dishes\(^1\) (155 mm X 63 mm) each lined with Armstrong \# 6 filter paper\(^2\) and each containing 15 ml of distilled water. All containers were placed in clear, polyethylene bags\(^3\) which were closed by means of wire twists. The dishes were placed in Sherer Controlled Environment Chambers\(^4\). Experiments in which oats were tested and those in which wheat were tested were run simultaneously.

Dishes were placed in the center of the chamber with the experimental dishes under the speaker. They were placed in a pattern as shown in Figure 1. The containers were kept in constant darkness at 20°C ± 1°C. The experimental series was exposed to sound continuously. A control group was run simultaneously in the second chamber under the same conditions but with no sound, except normal background. All seeds were counted after 5 days. A total of 8 separate runs, 4 replications of the experiment, were made and the growth chambers were interchanged for each succeeding run so that an 'experimental' chamber became a 'control' chamber on

---

\(^1\) Plastic containers were obtained from Bradley Industries, Inc., Franklin Park, Illinois.

\(^2\) Paper manufactured by the Armstrong Cork Co., Lancaster, Pennsylvania.

\(^3\) Bag made by Union Carbide Corp., Consumer Products Division, 270 Park Avenue, New York, New York, and sold under the trade name of 'Glad'.

Figure 1. Photograph showing inside of experimental growth chamber with dishes (alternating, oats when wheat) under speaker. Control chamber same but with no speaker.
on the next run systematically.

Determination of Germination and Growth

A seed was considered to have germinated when its root obtained a length of 1 mm or more. Growth was assessed using the method of Thompson et al., (1945) where the longest root of each seed was measured in mm. A mean root length for both controls and experimentals was then calculated using the root lengths of all seeds that did germinate.

Statistical Analysis

Root elongation data were analyzed by means of a two-way analysis of variance (unbalanced case) where factor 1 is the difference between control and experimental treatments and factor 2 is the difference between runs. Analysis was carried out using Western Michigan University's PDP-10 computer and library program # 1.9.2. The analysis was carried out using only those seeds that did germinate. If interaction was at a significant level (.05) for the root data, it was further analyzed, using mean values only, by library program # 1.9.1 (version 2). This two-way analysis of variance program showed the significance of control/experimental, replication, and growth chamber differences while not looking at the within cell variability. Per cent germination data was also analyzed by means of this program as there was only one observation per cell with germination.

In both programs, from the 'F' value generated, a probability level was calculated.
RESULTS

Root Elongation

Tables 1 and 2, pages 11 and 12, show the average length of the longest root of germinated oat and wheat seeds under both control and experimental conditions. In both cases the grand mean (GM) of the control group is significantly greater than the grand mean of the experimental group. Tables 3, 4, and 5 on pages 13, 15, and 16 show the results of the statistical analysis.

Oat seeds

With oats (see Table 1, page 11), the grand mean of the control group is 118.04 mm while the grand mean of the experimental group is 108.64 mm. The difference observed is significant at below the 0.1 per cent level\(^1\) (see Table 3, page 13). A significant difference (below the 0.1 per cent level) is found between the 8 runs of the experiment. Interaction is significant at below the 0.1 per cent level.

Table 5, page 16, shows the analysis of root elongation data using mean values only and library program \# 1.9.1 (version 2). The data here are grouped so one can now see that the difference between growth chambers A and B is not significant (93.27 per cent level).

\(^1\)As recommended by Remington and Schork (1970), the level of significance will be reported along with the statement if the level is at a significant level for this experiment (5.0 per cent level).
Table 1. Mean length in mm of longest root of germinated oats for control and sound treated seeds. GM = Grand Mean.
Table 2. Mean length in mm of longest root of germinated wheat for control and sound treated seeds. GM = Grand Mean.
**Grand mean for control** 118.04 mm

**Grand mean for experimental** 108.64 mm

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Table 3. Two-way analysis of variance (unbalanced case) for root elongation data of oats

(library program # 1.9.2).
Wheat seeds

The data for wheat (see Table 2, page 12) follow a similar pattern as in oats showing an inhibition of growth in the sound treated group. The control group has a grand mean of 111.66 mm and the grand mean of the experimental group is 108.90 mm. The difference between the two is a significant difference at the 4.1 per cent level (Table 4, page 15). There is a significant difference between the 8 runs (below the 0.1 per cent level); and the interaction is significant also being below the 0.1 per cent level.

Table 5, page 16, shows the analysis of root elongation data using mean values only. The difference between the four replications is significant below the 1.0 per cent level but the difference between growth chambers A and B is not significant (94.56 per cent level).

Germination

Tables 6 and 7, pages 17 and 18, show the per cent germination for both oats and wheat. Table 8, page 19, shows the level of significance for the differences. In both oats and wheat, a significant difference was not found between per cent germination for either growth chambers, replications, or control/experimental differences.

Oat seeds

A significant difference was not found in per cent germination between control and experimental plants (10.03 per cent level),
Grand mean for control 111.66 mm  
Grand mean for experimental 108.90 mm  

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<td>Interaction</td>
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Table 4. Two-way analysis of variance (unbalanced case) for root elongation data of wheat (library program # 1.9.2).
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<tr>
<td>Oats</td>
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<td>Difference between chamber A and B</td>
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Table 5. Two-way analysis of variance of oats and wheat root elongation data using mean values only (library program # 1.9.1 (version 2)).
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<td>Control</td>
<td>88.88</td>
<td>90</td>
<td>92</td>
<td>89</td>
<td>90</td>
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<tr>
<td>Sound</td>
<td>89.63</td>
<td>92</td>
<td>86</td>
<td>91</td>
<td>90</td>
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Table 6. Per cent germination of oats for control and sound treated seeds. GM = Grand Mean.
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<td>97</td>
<td>95</td>
<td>97</td>
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Table 7. Per cent germination of wheat for control and sound treated seeds. GM = Grand Mean.
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<td>0.1003</td>
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<td>Difference between replications</td>
<td>1.183</td>
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<td>Difference between chamber A and B</td>
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<td>0.6611</td>
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<td><strong>Wheat</strong></td>
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<tr>
<td>Difference between control/experimental</td>
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<td>0.5306</td>
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<td>Difference between replications</td>
<td>0.798</td>
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<td>Difference between chamber A and B</td>
<td>0.000</td>
<td>1.0000</td>
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</table>

Table 8. Two-way analysis of variance of oats and wheat germination data (library program 1.9.1 (version 2)).
between replications (36.48 per cent level), or between growth chambers A and B (66.11 per cent level).

**Wheat seeds**

As in oats, a significant difference was not found between per cent germination of control and experimental plants (53.06 per cent level), between replications (52.27 per cent level), or between growth chambers A and B (100.00 per cent level).
Plants treated with 300 Hz sound showed a significant inhibition of root elongation in both oats and wheat although there was a greater inhibition in oats. However, no significant change was observed in their germination.

In all runs, considerable variation is observed in both control and experimental groups. In oats, control plants show a low mean of 105.54 mm and a high mean of 131.53 mm; experimental plants on the other hand show a low mean of 92.37 mm and a high mean of 124.46 mm. Wheat control series shows a low mean of 105.45 mm and a high mean of 119.28 mm. The experimental series in wheat shows a low mean of 103.61 mm and a high mean of 117.32 mm.

The reasons for this variability cannot be attributed to growth chamber differences (see Table 5, page 16). This means that each run of the experiment can be considered a replication of the experiment. A statistical check showed that the differences in mean root length in growth chamber A and in growth chamber B are not significant (i.e. about the 94.00 per cent level). However, visual inspection of the seeds showed differences in morphological types and therefore the observed variability may be due to genetic factors.

Moreover, it is possible that if there occurred any variation of noise in the chambers, whatever the source, root elongation could be affected (Woodlief et al., 1969). Background
noise can reasonably be assumed to be nearly of the same intensity in both growth chambers since the two chambers employed are only a foot or so apart and thus any laboratory noise would impinge upon each chamber with equal intensity. Furthermore, the circulating fans in both chambers are identical. Finally, the compressors are far removed from the chambers and thus their noise can safely be eliminated from consideration. It appears, therefore, to completely eliminate the above possibility, i.e. variation of noise, would require growth chambers with no background sound, which are both difficult and expensive to construct and beyond the scope of this study.

In general, mean values in all series, control or experimental, increase or decrease simultaneously; i.e. if a control mean value increases, when compared with the previous run, the experimental mean value also increases and vice versa. This data suggests that the variability between runs might be due to a 'biological rhythm'. It would be of value to pursue this interesting possibility in future experimentation.

It is possible that the reversal observed in run # 2 for wheat (Table 2, page 12) is not significant. However, the observation that run # 7 did not show root inhibition with sound (Tables 6 and 7, pages 17 and 18) while the other runs did for both oats and wheat is disturbing.

Lowest germination values also are observed in run # 7 for both oats and wheat (Tables 6 and 7, pages 17 and 18). These values cannot be attributed to differences in growth chambers. It
is possible, therefore, that germination and elongation are related in these experiments.

Furthermore, since there is significant interaction in this experiment (Tables 3 and 4, pages 13 and 15), we cannot be sure that the observed effects, i.e. inhibition of root elongation, is due to sound. It is interesting to note, however, that if run #7 is not included in the analysis for both oats and wheat, the interaction is no longer significant (above the 50.00 per cent level) and the data point to a most dramatic inhibition.

Germination in both oats and wheat treated by 300 Hz sound fails to show significant differences (Table 8, page 19). It might be desirable to carry on additional experimentation with oats because the data are not significant at the 5.0 per cent level although the data do show significance at the 10.03 per cent level.

In these experiments only 300 Hz was employed. Lisenkov (1966), Measures and Weinberger (1970) and Weinberger and Das (1972) report differences in the same species when the frequency is varied. Preliminary experiments not ready for inclusion in this report indicate that 600 Hz (104 db) sound treated plants show a significant (2.00 per cent level) inhibition of root elongation in oats and in cucumber. In both cases, germination was not affected.

In conclusion, I have rejected the null hypothesis — $U_C = U_e$ (root elongation). I am not able to state conclusively, on the basis of a statistical analysis which includes all runs, that the reduction in root elongation is due to the sound
treatment. However, if run #7 is not included in the analysis, the data point conclusively to inhibition of root elongation by 300 Hz sound in oats and wheat. In future experiments, more attention should be given to reducing the variability between runs, or to account for such variability by means of statistical treatment. Also, experiments should be designed which consider genetic variability, biological rhythms, or the effect of low germination and their interaction with sound treatment. The null hypothesis for the effect of 300 Hz sound on germination — \( U_c = U_e \) — is not rejected.
LITERATURE CITED


