

HORTON, SHERRY JEAN. The Effects of 4000 Hz, 100 db Sound on the Kinetics and Thermodynamics of Isothermal Seed Germination. (1976) Directed by: Dr. Gaylord T. Hageseth.

It was the purpose of this study to determine the effects of 4000 Hz, 100 db sound on the germination rate of Grand Rapids Lettuce seeds. The data were tested for agreement with the mathematical model for germination developed by Dr. Gaylord T. Hageseth. Using this model the activation energy and other parameters were determined. Comparison was made with the results obtained from experiments under similar conditions except for a quiet environment, performed by Mr. Leon Jaynes.

Random samples of Grand Rapid Lettuce seeds were germinated under identical conditions of light, humidity and exposure to sound. Each sample was observed during germination at a different constant temperature.

Computer methods were used to analyze the data and to fit theoretical curves to the experimental data by the method of least squares. The chi-square test was used as the criteria for closeness of fit.

It was found that the Hageseth model fit the data and that while the kinetics for the noise groups and the quiet groups were different the thermodynamics were the same.

THE EFFECTS OF 400 HZ, 100 DB SOUND ON THE KINETICS AND

THERMODYNAMICS OF ISOTHERMAL

SEED GERMINATION

by

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A Thesis Submitted to the Faculty of the Graduate School at The University of North Carolina at Greensboro in Partial Fulfillment of the Requirements for the Degree Master of Science

> Greensboro 1976

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ACKNOWLEDGEMENTS

The author wishes to express appreciation for the guidance provided by Dr. Gaylord T. Hageseth and to thank the physics department of the University of North Carolina at Greensboro for providing the equipment necessary for this research. Also the author wishes to thank Mr. Leon Jaynes for the use of his data collected without the presence of 4000 Hz, 100 db sound.

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CHAPTER I

INTRODUCTION

Much work has been done studying the effects of different parameters on seed germination. Some of the most recent concerns the effects of sound on the germination rate of turnip seeds (Hageseth and Joyner, 1975). Dr. Hageseth has developed a mathematical model for the germination rate of seeds and the total number of seeds that germinate as a function of time based on kinetics and thermodynamic principles (Hageseth, 1974).

The purpose of this thesis was to see if the Hageseth model is applicable to Grand Rapid Lettuce seeds and to determine the effects of 4000 Hz 100 db sound on the germination of the seeds. Each sample was observed during germination for a different constant temperature. The number of seeds germinated each hour were counted. Then the kinetic and thermodynamic values determined from the data were compared with the kinetic and thermodynamic values Mr. Leon Jaynes determined for the germination of Grand Rapid Lettuce seeds in a quiet environment.

The Hageseth model is discussed in Chapter II. Chapter III deals with the method of experimentation. The analysis of data is presented in Chapter IV, and conclusions are found in Chapter V.

CHAPTER II

THEORY

The Hageseth model for seed germination assumes that the same mathematics which are applied to autocatalytic enzyme reactions can be applied to seed germination rates. An autocatalytic reaction is one in which the products of the reaction catalyse the reaction (Stevens, 1970). In the first reaction two enzyme substrate A and B combine to form complex AB,

$$A + B \ddagger AB$$
, (1)

in the second reaction AB reacts to from the final enzyme F by a first-order enzyme reaction,

$$AB \ddagger F + B,$$
 (2)

or by a second-order autocatalytic enzyme reaction,

$$AB + F + 2F + B.$$
 (3)

The result is the transformation of enzyme A into enzyme F.

The initial rate for the first-order reaction is

$$d[F]/dt = k_1[A].$$
(4)

[F] is the concentration of F, [A] is the concentration of A and k_1 is the rate constant. For the second-order reaction the rate is

$$d[F]/dt k_2([A]_-[F])([F]_+[F]).$$
 (5)

 $[A]_{0}$ and $[F]_{0}$ are the initial concentrations of A and F respectively and k_{2} is the rate constant. When equation (5) is integrated it gives the S-shaped curve which is typical for an autocatalytic reaction. Since $[A]_{o}$, $[F]_{o}$, [A] and [F] cannot be measured directly, external parameters which can be correlated to these concentrations need to be found. The concentration of F varies from the initial concentration, $[F]_{o}$ to a critical concentration, $[F]_{c}$, when germination, the breaking of the seed coat by a radical, occurs. An estimation of the average concentration of F can be found by counting the number of seeds which have reached the critical concentration, i.e. the seeds that have germinated. The assumption is made that each seed that has germinated contributes one unit of $[F]_{c}$ and that seeds which have not germinated contribute zero. Thus for a population of 400 seeds the average concentration of F divided by 400. By setting the value of $[F]_{c}$ divided by 400 to be the unit of enzyme concentration and equal to one, the average concentration of F is the number of F is the number of germinated seeds N, yielding

$$d[F]/dt = dN/dt.$$
 (6)

Substituting equation (6) into equation (5) gives

$$dN/dt = k_2([A] - [F])([F] + [F]).$$
 (7)

Equation (7) describes the rate of seed germination. Thus in equation (7) [F] is the number of seeds germinated and $[A]_0$ is the number of seeds that will germinate by the autocatalytic reaction.

The activation energies for $[A]_0$ and $[F]_0$ can be found by plotting ln $[A]_0$ versus 1000/T and $[F]_0$ versus 1000/T. For both plots a straight line results and the slopes are equal to $-E_a/k$, where E_a is the activation energy and k is Boltzmann's constant.

The Hageseth model uses the thermodynamics of the reversible

denaturation of proteins to calculate the change in enthalpy, the Gibb's free energy, and the entropy for the germination process after the equilibrium constant for each temperature has been determined.

The equilibrium constant for reactions (1), (2) and (3) are

$$K_1 = [AB]_{o} / [A]_{o} [B]_{o}, \qquad (8)$$

$$K_2 = [B]_0 [F]_0 / [AB]_0, \qquad (9)$$

and

$$K_{3} = [F]_{0}^{2} [B]_{0}^{2} [AB]_{0}^{0} [F]_{0}^{2} = [F]_{0}^{2} [B]_{0}^{2} [AB]_{0}^{2},$$
 (10)

where $[AB]_0$ is the initial concentration of complex AB. Equations (9) and (10) show that $K_2 = K_3$, thus if either reactions (1) and (2) or (1) and (3) occur consecutively, a new equilibrium constant may be defined which describes both reactions:

$$K_{12} = K_1 K_2 = [F]_0 / [A]_0.$$
 (11)

The equation

$$\Delta G = \Delta H - T \Delta S \tag{12}$$

relates the change in the free energy, enthalpy and entropy. ΔG is the change in free energy when one mole of enzyme A is changed into one mole of enzyme F, ΔH is the change in enthalpy per mole, ΔS is the change in entropy per mole, and T is the absolute termpeature. Taking the partial derivative of equation (12) with respect to T yields

$$d(\Delta G)/dt = -\Delta S.$$
(13)

The change in free energy is given by

$$\Delta G = -RT \ln K_{12} \tag{14}$$

or

$$K_{12} = \exp(-\Delta G/RT)$$
(15)

where R, the universal gas constant is equal to 1.98 calories.

Substituting equation (14) into equation (12) gives

$$-RT \ln K_{12} = \Delta H - T \Delta S$$
$$-\ln K_{12} = \Delta H/RT - \Delta S/R.$$
(16)

Differentiating equation (16) with respect to (1/T) yields

$$-d(\ln K_{12})/d(1/T) = \Delta H/R$$
(17)

this is van't Hoff's equation. Thus a plot of $\ln K_{12}$ as a function of (1/T) has a slope of - $\Delta H/R$. For a small temperature range the change in enthalpy is approximately constant. So we can write

$$\ln K_{12}(T_2)/K_{12}(T_1) = -\Delta H(1/RT_2 - 1/RT_1).$$
(18)

When the slope is negative ΔH is positive and the reaction is endothermic; when the slope is positive ΔH is negative and the reaction is exothermic.

Plotting ΔG as a function of T we can find the change in entropy. From equation (12) the slope is $-\Delta S$ and the intercept is ΔH . The change in enthalpy from equations (12) and (18) should be equal.

CHAPTER III EXPERIMENTAL CONSIDERATIONS

In each experiment 400 lettuce seeds were placed in a rectangular array on paper towels moistened with 200 ml of water in a special box; the bottom of which was sheet aluminum in contact with a constant temperature water bath and which was tightly covered to prevent evaporation. Joyner has a detailed description of the apparatus used.

The seeds were placed into the box simulanteously. Thirty minutes after wetting the seeds were exposed to 4000 Hz 100 db sound for one hour. The room was dark except for five minute hourly intervals after observations were begun. It is expected that this exposure to light had no effect since the exposure was the same for all.

A seed was considered germinated when the radical broke through the seed coat. During the observations the number of seeds that had germinated during the hour was recorded and the total number of seeds germinated was also recorded.

CHAPTER IV DATA ANALYSIS AND RESULTS

With the aid of computer programs the data for each temperature (see Appendix I) was analyzed. The best theoretical curve was found using a least-square fit and minimum chi-square fit to the germination rate and to the integrated germination rate. The best fit was found by varying k_2 , $[A]_0$ and $[F]_0$ until the minimum chi-square was found. Table 1 contains the values of k_2 , $[A]_0$ and $[F]_0$ for each temperature. Table 2 contains the sum of the chi-square and the confidence levels for the germination rate and the integrated germination rate. The data and the theoretical curve for the integrated rate were plotted for each experiment (see Appendix II).

The integrated rate curves are found in Appendix II. The total number of seeds germinated at a given hour were plotted versus that hour. The solid line represents the theoretical curve and the closed circles represent the experimental data points. This same method was used for all the graphs in this thesis.

After the minimum chi-square curves were found, it was observed that for the temperatures 11.5° C, 17.3° C and 26.0° C the values were not meaningful thermodynamically. The values were adjusted to yield better results. The fit to the data was not seriously altered (see Table 2). The rest of the analysis was done with the corrected values.

The confidence levels for the germination rates and the closeness

Table 1: Tabulated Experimental Values

Temp. (°C)		[F]	k_2^a	<u>t_(hrs)</u> **
28.5	58.0	0.50	5.96	16
26.0	227.0	4.40	2.21	18
26.0*	227.0	.44	2.42	18
22.5	291.0	1.05	1.88	15
19.2	360.0	6.20	1.00	18
17.3	279.0	.87	1.62	20
17.3*	279.0	2.03	1.56	20
14.5	400.0	1.00	.87	25
11.5	220.0	2.50	1.18	25
11.5*	220.0	1.06	1.24	39
10.2	189.0	2.20	3.05	48

a x 10⁻³

* revised value

** to is the dead time (time from wetting to germination of first seed)

Table 2: Goodness of Fit

Temp. (°C)	Sum of X^2 a	Confidence Level a(%)	Sum of X ² b	Confidence Level b
28.5	9.34	75.0	3.95	99.9+
26.0	11.94	50.0	3.93	08.0
26.0*	14.19	25.0	10.94	50.0
22.5	9.83	60.0	2.33	99 o ⁺
19.2	14.36	20.0	6.25	00.0
17.3	6.06	75.0	1.54	90.0
17.3*	6.56	70.0	2.72	99
14.5	5.83	98.0	1.19	00.0 ⁺
11.5	5.25	95.0	6.96	99.9
11.5*	7.31	88.0	13.62	91.0
10.2	12.84	80.0	6.99	45.0 99.0 %

a for germination rate

b for integrated germination rate

* revised values

0

of the theoretical integrated rate curves to the experimental data indicate that the Hageseth model is applicable to Grand Rapid Lettuce seeds.

Table 3 contains the experimental calculations used. The values were to plot the ln t_o versus 1000/T, the ln $[A]_o$ versus 1000/T. The ln $[F]_o$ versus 1000/T, the ln $[F]_o/[A]_o$ versus 1000/T, the ln k_2 versus 1000/T, and ΔG versus T.

Figure 1 is a plot of ln t_o (the dead time) versus 1000/T, for the range 10.2° C to 22.5° C. For this range there is a linear relationship. The equation of this relationship is

$$\ln t = m/T + b.,$$

where m is the slope and b is the y-intercept (Hageseth, 1976). The thermodynamics of the slope and intercept have not yet been determined. The slope is equal to 7.90 ± 0.86 . The product-moment correlation coefficient is 0.977 with an F-statistic of 85.38 on 1 and 4 degrees of freedom. The confidence level is 99.9%. The value of the slope for the quiet data obtained by Mr. Jaynes was 7.12 ± 0.84 with a confidence level of 99%. Thus sound had no effect on this parameter.

Plotting $\ln [A]_{0} (A]_{0}$ is interpreted as the number of seeds which will germinate by the autocatalytic reaction for a set of environmental conditions) versus 1000/T gives a straight line with a slope equal to $-E_{a}/k$ where E_{a} is the activation energy and k is Boltzmann's constant (Hageseth 1976). The ln A was plotted for two ranges $10.2^{\circ}C$ to $19.2^{\circ}C$ and $19.2^{\circ}C$ to $28.5^{\circ}C$, (see Figure 2). For the range $10.2^{\circ}C$ to $19.2^{\circ}C$ the slope is -5.25 ± 2.64 with a confidence level of 85%. The activation energy is 10.40 ± 5.23 . The slope for the second range has no meaning according to the model. Combining the noise data with the

Temp. (K)	$1000/T (1/k^{0})$	ln t _o	In [A]	ln [F]	$\ln [F] / [A]_{O}$	<u>ln k₂</u>	AG (kcal/mole)
301.5	3.316	2.77	4.06	-0.690	-4.75	-5.12	2 836
299.0	3.344	2.89	5.42	-0.820	-6.24	-6.02	3 694
295.5	3.384	2.71	5.67	0.049	-5.62	-6.27	3.288
292.2	3.422	2.89	5.89	1.820	-4.06	-6.90	2.349
290.3	3.445	3.00	5.63	0.708	-4.92	-6.46	2,828
287.5	3.478	3.22	5.99	0.000	-5.99	-7.05	3.410
284.5	3.515	3.66	2.39	0.058	-5.33	-6.69	3.002
283.2	3.531	3.87	5.24	0.788	-4.45	-5.79	2 495

Table 3: Tabulated Experimental Calculations





quiet data collected by Mr. Jaynes gives a slope of -7.07 ± 2.37 with a confidence level of 98%. For the second range both slopes are within one standard deviation. Again it appears that sound has no effect.

The activation energy for $[F]_{0}$ can also be determined by the same method used for $[A]_{0}$. Three ranges were used in plotting ln $[F]_{0}$ versus 1000/T (see Figure 3); 10.2°C to 14.5°C, 14.5°C to 19.2°C, and 19.2°C to 26.0°C. The only meaningful slope is for the range 14.5°C to 19.2°C. For this range the slope is -31.73 ± 7.44 with a confidence level of 85% and an activation energy of -66.82 ± 14.20. My data was combined with the data collected by Mr. Jaynes and analyzed over the same three ranges. For the range 14.5°C to 19.2°C the slope was -39.49 ± 5.41 with a confidence level of 99.8%. The values of the slopes for the three ranges are all within one standard deviation of the values for the noise data. Therefore it can be concluded that sound had no effect.

Figure 4 is a plot of the natural logerithms of the reaction constant, k_2 , versus 1000/T. The activation energy is equal to -R times the slope of the curve. For the noise data the slope is -4.12 ± 2.88 with a confidence level of 80%. The activation energy is 8.16 ± 5.70. For the combined noise and quiet data the slope is -4.69 ± 2.05 with a confidence level of 96%. Thus we can again conclude that noise has no effect on the activation energy.

The ratio $[F]_0/[A]_0$ was assumed to be the equilibrium constant in the Hageseth model. The natural logarithm of this ratio versus 1000/T was plotted for three ranges in Figure 5. -R times the slope of the curve yields the change in enthalapy. For the range 10.2° C to 14.5° C the slope is 27.09 ± 8.59 with a confidence level of 82%



Figure 3: Plot of ln [F] versus 1000/T



Figure 4: Plot of ln k_2 versus 1000/T





the change in enthalpy is -53.64 \pm 17.01; for the range 14.5°C to 19.2°C the slope is -34.32 \pm 1.37 with a confidence level of 99.8%, the change in enthalpy is 67.95 \pm 2.71; and for the range 19.2°C to 26.0°C the slope is 27.97 \pm 7.30 with a confidence level of 82%, the change in enthalpy is -55.38 \pm 14.45. When the noise data is combined with the quiet data obtained by Mr. Jaynes, the change in enthalpy for the ranges are -60.45 \pm 10.20 with a confidence level of 99.3%, 70.13 \pm 2.26 with a confidence level of 99.94%.

This plot indicates that there are separate reactions for the germination process. The reactions in the ranges 10.2° C to 14.5° C and 19.2° C to 26.0° C are exothermic while the reaction in the range 14.5° C to 19.2° C is endothermic. Comparing the standard deviations it can be concluded that noise has no effect on the change of enthalpy.

The Hageseth model predicts that if the free energy is plotted as a function of time the slope will be equal to the change in entropy and the y-intercept will be equal to the change in enthalpy. Figure 6 is a plot of ΔG versus T for the ranges, 283.2° K to 287.5° K, 287.5° K to 292.2° K, and 292.2° K to 299.0° K. For these ranges respectively for the noise data the change in entropy is $.199 \pm .059$ kcal/mole $-^{\circ}$ K, $-.224 \pm .012$ kcal/mole $-^{\circ}$ K and $.197 \pm .049$ kcal/mole $-^{\circ}$ K; and the change in enthalpy is -53.86 ± 16.76 kcal/mole, 67.94 ± 3.52 kcal/mole and -55.11 ± 14.36 kcal/mole. The confidence levels are 81%, 96% and 87%, respectively. For the combined data (noise and quiet) the change in entropy is $.224 \pm .036$ kcal/mole $-^{\circ}$ K, $-.233 \pm .010$ kcal/mole $-^{\circ}$ K and $.209 \pm .022$ kcal/mole $-^{\circ}$ K; the change in enthalpy is -60.86 ± 10.12 kcal/mole, 70.43 ± 2.75 kcal/mole and -58.76 ± 6.65 kcal/mole; and



Figure 6: Plot of AG versus T

confidence levels of 99.6%, 99.94% and 99.94% for the three ranges. It can thus be seen that noise has no effect on the change in enthalpy or the change of entropy.

Table 4 contains the calculated thermodynamic variables. From Table 4 it can be seen that the values of ΔH calculated from the plot $\ln [F]_{0}/[A]_{0}$ versus 1000/T and the plot ΔG versus T are in close agreement. It can also be seen that noise has no effect on the thermodynamic variables. Table 5 contains the linear regression statistics for the plots.

Anso and Mirky (1934) reported that the change in enthalpy of trypsin is 67.6 kcal/mole with a change in entropy of .213 kcal/mole $-^{\circ}$ K. These values are of the same magnitude as those calculated and are in close agreement for the values obtained in the range 14.5°C to 19.2°C.

For turnip seeds it was found that sound did affect the thermodynamic variables. The values of ΔH and ΔS for the quiet group below 33° C are very close to the values we obtained in the range 14.5° C to 19.2° C. It is interesting to note that the thermodynamic values for the noise data below 33° C are much closer to the values for the quiet data than those above 33° C for the turnip seeds.

Table 4: Calculated Thermodynamic Variables

Source	ΔH	<u>ΔS</u>	ΔE
	Range 10.2°C to 14	.5°C - Noise Data	
[F] _O /[A] _O AG	-53.64 ± 17.01 -53.86 ± 16.76	.199 ± .059	
	Range 10.2°C to 14	.5°C - Combined Data	
[F] ₀ /[A] ₀ ΔG	-60.45 ± 10.20 -60.86 ± 10.13	.224 ± .036	
	Range 14.5°C to 19	.2 ⁰ C - Noise Data	
[F] ₀ /[A] ₀ ΔG	67.95 ± 2.71 67.94 ± 3.52	224 ± .012	
	Range 14.5°C to 19	.2°C - Combined Data	
[F] _o /[A] _o AG	70.13 ± 2.26 70.43 ± 2.75	233 ± .010	
	Range 19.2°C to 26	.0°C - Noise Data	
[F] ₀ /[A] ₀ ΔG	-55.38 ± 14.45 -55.11 ± 14.36	.197 ± .049	
	Range 19.2°C to 26	.0°C - Combined Data	
[F]/[A] AG	-58.59 ± 6.47 -58.76 ± 6.65	.209 ± .022	
	Range 10.2°C to 28	.5°C - Noise Data	
k ₂			8.16 ± 5.70
	Range 10.2°C to 28	.5°C - Combined Data	
k.			9.29 ± 4.06

Table	5	5: 1	Linear	Regression	Statistics
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Graph	Range	Slope $\pm \sigma_{s}$	Y-Intercept ± o	PMCC	F-Statistic	Confidence Level %
ln t _o	10.2°C-22.5°C	7.90 ± 0.86	-24.14 ± 2.96	0.977	85.38 on 1 & 4 deg. of freedom	99.9 +
ln[A] _o	10.2 ^o C-19.2 ^o C	-5.25 ± 2.64	23.88 ± 9.17	-0.754	3.96 on 1 & 3 deg. of freedom	85
	19.2°C-28.5°C	15.39 ± 6.26	-46.54 ± 21.08	0.867	6.04 on 1 & 2 deg. of freedom	87
ln[F] _o	10.2°C-14.5°C	12.54 ± 10.18	-43.70 ± 35.72	0.776	1.52 on 1 & 1 deg. of freedom	60
	14.5 ^o C-19.2 ^o C	-31.73 ± 7.44	110.27 ± 23.64	-0.974	18.21 on 1 & 1 deg. of freedom	85
	19.2 ^o C-26.0 ^o C	+33.75 ± 7.17	-113.88 ± 24.26	0.978	22.15 on 1 & 1 deg. of freedom	85
ln k2	10.2°C-28.5°C	-4.12 ± 2.88	7.84 ± 9.86	-0.505	2.05 on 1 & 6 deg. of freedom	80
11 b/[A]	0 ^{10.2°} C-14.5 [°] C	27.09 ± 8.59	-100.29 ± 30.15	0.953	9.94 on 1 & 1 deg. of freedom	82
	14.5°C-19.2°C	-34.32 ± 1.37	113.37 ± 4.74	-0.999	624.42 on 1 & 1 deg. of freedom	99.8
	19.2°C-26.0°C	27.97 ± 7.30	-99.94 ± 24.69	0.968	14.69 on 1 & 1 deg. of freedom	82

Table	5.	Linear	Regression	Statistics	(Camp :
Table	5.	Linear	Regression	Statistics	(Continued)

Graph	Range	Slope $\pm \sigma_s$	<u>Y-Intercept $\pm \sigma_i$</u>	PMCC	F-Statistics	Confidence Level %
۵G	10.2°C-14.5°C	0.199 ± 0.059	-53.86 ± 16.76	0.959	11.50 on 1 & 1 deg. of freedom	81
	14.5°C-19.2°C	-0.224 ± 0.012	67.94 ± 3.52	-0.998	340.75 on 1 & 1 deg. of freedom	96
	19.2 ^o c-26.0 ^o c	0.197 ± 0.049	-55.11 ± 14.36	0.971	16.43 on 1 & 1 deg. of freedom	87

CHAPTER V

CONCLUSIONS

The Hageseth model of seed germination was found to fit the data obtained for Grand Rapid Lettuce seeds. Using this model the changes in enthalpy, entropy and free energy were obtained.

It was found that both endothermic and exothermic reactions take place depending on the ambient temperature. The effect of temperature is seen in the number of seed germinated, the germination rate and the dead time of the seeds.

Although the application of 4000 Hz 100 db sound affects the kinetics, it does not affect the thermodynamic variables of the reactions. All thermodynamic variables of the noise group were within one standard deviation of the variables of the combined noise and quiet groups.

Much work still remains to be done in this field. Other experiments should be carried out to see if the Hageseth model fits for other type of seed and to see if sound affects the thermodynamic variables.

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APPENDIX I

EXPERIMENTAL DATA

Table 6	
Experiment I	
10.2 °C	

Time (Hours)	Number Germinated	Total Germinated
48	1	1
49	1	2
50	1	3
51	2	5
52	2	7
53	3	10
54	2	12
55	2	14
56	2	16
57	2	18
58	2	20
59	2	22
60	6	28
61	8	36
62	12	48
63	14	62
64	7	69
65	7	76
66	8	84

Table 7 Experiment II 11.5 ^oC

Time (Hours)	Number Germinated	Total Germinated
38	0	0
39	3	3
40	1	4
41	5	9
42	4	13
43	6	19
44	6	25
45	6	31
46	8	39
47	9	48
48	11	59
49	12	71
50	12	83
51	17	100
52	16	116

Table 8 Experiment III

14.5 °C

Time (Hours)	Number Germinated	Total Germinated
24	0	0
25	1	1
26	1	2
27	1	3
28	2	5
29	3	8
30	5	13
31	7	20
32	11	31
33	10	41
34	15	56
35	21	77
36	23	100
37	37	137
38	26	163
39	40	203
40	34	237
41	28	265

Table 9

Experiment IV 17.3 ^oC

Time (Hours)	Number Germinated	Total Germinated
18	0	0
19	0	0
20	1	1
21	3	4
22	2	6
23	4	10
24	8	18
25	15	33
26	14	47
27	20	67
28	40	107
29	33	140
30	27	167

Table 10 Experiment V 19.2 ^OC

Time (Hours)	Number Germinated	Total Germinated
18	3	3
19	11	14
20	11	25
21	12	37
22	9	46
23	12	58
24	30	88
25	41	129
26	38	167
27	32	199
28	26	225
29	30	255
30	23	278

Table 11 Experiment VI

22.5 °C

Number Germinated	Total Germinated
0	0
3	3
1	4
3	7
10	17
26	43
26	69
40	109
40	149
37	186
32	218
14	232
19	251
15	266
	Number Germinated 0 3 1 3 10 26 26 26 40 40 37 32 14 19 15

Table 12

Experiment VII 26.0 ^OC

Time (Hours)	Number Germinated	Total Germinated
16	0	0
17	0	0
18	3	3
19	6	9
20	11	20
21	30	50
22	28	78
23	27	105
24	20	125
25	25	150
26	22	172
27	18	190
28	7	197
29	12	209
30	5	214

Table 13 Experiment VIII 28.5 ^OC

Time (Hours)	Number Germinated	Total Germinated
16	1	1
17	2	3
18	0	3
19	0	3
20	0	3
21	2	5
22	3	8
23	5	13
24	4	17
25	3	20
26	8	28
27	7	35
28	3	38
29	3	41
30	4	45

APPENDIX I

TIME - INTEGRATED GERMINATION RATE CURVES







Figure 8: 11.5 °C integrated rate curve



Figure 9: 11.5 °C integrated rate curve (revised)



Figure 10: 14.5 °C integrated rate curve



Figure 11: 17.3 °C integrated rate curve



Figure 12: 17.3 °C integrated rate curve (revised)



Figure 13: 19.2 °C integrated rate curve



Figure 14: 22.5 °C integrated rate curve







Figure 16: 26.0 °C integrated rate curve (revised)



Figure 17: 28.5 °C integrated rate curve