

Mathematical Statistical Modelling of Germination Kinetics

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Abstract - In present work a mathematical-statistical description for the germination process has been found, in which the different parameters have direct physical-chemical meanings. The result of the model was controlled by experimental data given by seed germination trials of the common reed (*Phragmites australis*). The germination time being different seed-by-seed can be considered a random variable of normal distribution according to the model. The distribution function of this random variable gives the proportion of germinated seeds as the function of time, which can be characterized by two parameters, the mean value and the standard deviation of the germination time. Germination process can be described by a „two-type processes model”, where the rate of one type of processes is controlled by the material transport and the rate of the other type is determined by chemical reactions. Comparing the results of the model with the experimental data, the ratio of these is 30%-70% in the germination time, while the transport processes give the main part (>80%) of the standard deviation.

Keywords - reed, seed, germination, mathematical model, normal distribution

I. INTRODUCTION

Studying a seed population the germination time, t_G is a function of temperature, in such a way, that the thermal time, $TT = (T - T_B)t$ is accumulated, where T_B is base temperature, under which there is no germination [1]. A seed germinate when the TT reaches the value necessary for germination, that is

$$TT_G = (T - T_B)t_G. \quad (1)$$

This model is called TT -model and is generally used for description of germination time [2-3].

In the population the germination times of seeds, t_G are different [4], therefore it can be regarded as random variable. There is a lot of (logistic) models known for the probability distribution of germination time and for the proportion of seeds germinated within time t [5-6]. The disadvantage of these models is that their parameters have no physical-chemical meaning, so they can not refer to the processes during germination.

From the (1) the reciprocal of germination time, the germination rate is proportional to the temperature, T [7-8]. Because of it the germination time, t_G on this temperature T is proportional to TT_G , which has normal

distribution for lot of the seeds [9-10], while the base temperature, T_B is a parameter being characteristic of the lot of seeds and probably one for the species [11]. From this, according to the (1) the germination time has a normal distribution, while the proportion of lot of seeds germinated within time t will be ratio to the (normal) distribution function of germination time [12].

In present work this model of normal distribution has been developed, considering germination process as the resultant of physical-chemical processes and, as a consequence of this, the germination time being the sum of these various process times. Therefore, the values of the model parameters can be in direct relation to the characteristics of the physical-chemical processes during the germination mentioned above. The model having no connection to a specific material can be of universal validity. In this present work we use it to explain seed germination of Common Reed (*Phragmites australis*).

II. MATHEMATICAL STATISTICAL MODEL

A. Statistical characteristics of germination

Take a multitude of N elements (seeds of reed) and suppose $N(t)$ number of seeds germinated within time t . Look at the following volume

$$R(t) = \frac{N(t)}{N}, \quad (2)$$

which is called *germination ratio* (of mutually).

As germination of seeds belonging to a variety of germination parameters is not complete, only a part of them will do, $N(t \rightarrow \infty) = N_G$ [13], therefore the probability of germination of a seed picked spontaneously from the mutually is

$$p_G = \frac{N_G}{N} \quad (3)$$

equal to the ratio of germinated seeds ($t \rightarrow \infty$).

The volume p_G ($0 \leq p_G \leq 1$) is called *germination probability* [14] and it is regarded as one of characteristics of the germination process. Its value depends on the genetical characteristics of the lot of seeds and the parameters of germination, mainly on the amplitude of diurnal temperature cycles [13].

Take the multitude of germinated seeds, N_G . As within this the germination time of seeds is different, therefore germination time of a seed picked spontaneously from the (partial) multitude of germinated elements can be considered a random variable (τ_G). The probability of $\tau_G < t$ is

$$P\{\tau_G < t\} = \frac{N(t)}{N_G} = F(t) \quad (4)$$

equal to the *distribution function* of germination time, $F(t)$.

Comparing (2)-(4), the germination ratio (2.) will be the following

$$R(t) = p_G F(t), \quad (5)$$

which shows the germination time-course of the *germination kinetics*.

B. The linear process model

Germination process consists of a mixture of numerous sequent and/or parallel (sub)processes in which - from the beginning till the end - the composition of the seed changes continuously and finally the seed germinates. A linear approximation is used in the model, which means that the germination process is considered as a sum of sequent (n) processes, where the process $i+1$ starts when the process i has already finished (or has overcome a certain level).

According to this, germination time, τ_G as a random variable

$$\tau_G = \tau_1 + \tau_2 + \dots + \tau_n = \sum_{i=1}^n \tau_i \quad (6)$$

will be the sum of process times, τ_i , which are also random variables.

The mean value, t_G and standard deviation, σ_G of germination time according to the calculation law of the sum of mean values and standard deviations of the random variables are

$$t_G = \sum_{i=1}^n t_i, \quad (7.a)$$

$$\sigma_G^2 = \sum_{i=1}^n \sigma_i^2. \quad (7.b)$$

Since the germination process consists of thermal activated processes, therefore the the process times, τ_i follow an Arrhenius-type kinetics [14]. For this reason the mean value, t_i and standard deviation, σ_i will be Arrhenius-types (Appendix I.) as follows

$$t_i = A_i \exp\left\{\frac{E_i^*}{RT}\right\}, \quad (8.a)$$

$$\sigma_i = B_i \exp\left\{\frac{E_i^*}{RT}\right\}, \quad (8.b)$$

where E_i^* is the activation energy of process i , and A_i and B_i are the pre-exponential coefficients. The R is the universal gas constant and T is the germination temperature. In (8) both the mean value and the standard deviation depend on the temperature in the same way.

Two types of processes exist during germination: In case of one type the rate of the process (activation energy) is defined by the material transport (processes controlled by transport), while in the case of the other type the rate of process is determined by chemical reactions (processes controlled by chemical reactions).

C. The distribution function of the germination time

The distribution function of germination can be defined by the distribution functions of the processes. It is not necessary to know their exact form, if the next can be assumed: the processes are (almost) independent of each other, they are large in number ($n \gg 1$) and their distribution is approximately the same. In this case the distribution function, (4) of the germination time, (6), independently of the distribution functions of the different individual processes, can be approximated by the normal distribution function (central limit theorem), as

$$F(t) = \frac{1}{\sqrt{2\pi}\sigma_G} \int_{-\infty}^t \exp\left\{-\frac{(x-t_G)^2}{2\sigma_G^2}\right\} dx, \quad (9)$$

where the two parameters, t_G and σ_G are the *mean value* and *standard deviation* of the germination time and those can be got by comparing (7) and (8). It will be

$$t_G = \sum_{i=1}^n A_i \exp\left\{\frac{E_i^*}{RT}\right\}, \quad (10.a)$$

$$\sigma_G^2 = \sum_{i=1}^n B_i^2 \exp\left\{2\frac{E_i^*}{RT}\right\} \quad (10.b)$$

Validity of the normal distribution approximation has to be checked in every case by means of a statistical hypothesis test.

The kinetics of germination, (5) using distribution function (9.) will be the following:

$$R(t) = p_G N(t, t_G, \sigma_G), \quad (11)$$

which can be characterised by three parameters, the germination probability, p_G , the mean value, t_G and the standard deviation of the germination time, σ_G . These parameters are called *kinetical parameters* of germination.

The germination probability p_G has a weak dependence on the temperature [4], because the kinetics of germination (11) depends at first on the temperature through the mean value and the standard deviation of the germination time. This temperature dependence is quite

complicated using approximation is better to find suitable formulas.

D. "One-type processes model" approximation

Assume the processes taking place during germination are one-type processes with approximately the same activation energies. In this case $E_i^* = E^*$, further $A = \sum A_i$ and $B^2 = \sum B_i^2$. According to this

$$t_G = A \exp\left\{\frac{E^*}{RT}\right\}, \quad (12.a)$$

$$\sigma_G = B \exp\left\{\frac{E^*}{RT}\right\}, \quad (12.b)$$

the temperature dependency of the two parameters, the mean value and the standard deviation will be the same.

E. "Many-type processes" model, average activation energy approximation

Let us form mean value and standard deviation of germination time, they are

$$t_G = \bar{A} \exp\left\{\frac{\bar{E}_M^*}{RT}\right\} \quad (13.a)$$

$$\sigma_G = \bar{B} \exp\left\{\frac{\bar{E}_D^*}{RT}\right\}, \quad (13.b)$$

where now the temperature dependence of the mean value and the standard deviation is expressed in the same form as it is done in the case of one-type processes model. The only difference is, that average activation energy would be different at the two kinetical parameters (M mean value, D standard deviation).

The two average activation energies combining (10) and (13) can be written as follows

$$\bar{E}_M^* = RT \ln \left(\sum_{i=1}^n \frac{A_i}{A} \exp\left\{\frac{E_i^*}{RT}\right\} \right) \quad (14.a)$$

$$\bar{E}_D^* = \frac{RT}{2} \ln \left(\sum_{i=1}^n \frac{B_i^2}{B^2} \exp\left\{2\frac{E_i^*}{RT}\right\} \right). \quad (14.b)$$

The cause of the difference is that the preexponential parameters being in (14) are weight factors and they are different for the mean value and for the standard deviation, therefore the average activation energies, \bar{E}_M^* and \bar{E}_D^* can differ from each other.

III. EXPERIMENTS AND RESULTS

A. Materials and experiments

Germination trials of seeds *Phragmites australis* (harvested in Balatonfüzfő 2002. november) were carried

out on four average temperatures, $\bar{T} = (T_{MAX} + T_{MIN})/2$, which were 15°C, 20°C, 25°C and 30°C, the amplitude of diurnal temperature fluctuation, $\Delta T = T_{MAX} - T_{MIN}$ was of 20°C, 12+12 hours. Number of seeds were 500 in each cases.

At T_{MAX} seeds were exposed to white warm fluorescent light, while at T_{MIN} seeds were kept in darkness. Germinated seeds were counted once a day ($j=0, 1, \dots, J, J=14$) for 14 days. We considered a seed to be germinated, when radicle had broken through the seed coat. The ratio of germinated seeds (R_j) as a function of time (t_j) can be seen in Figure 1.

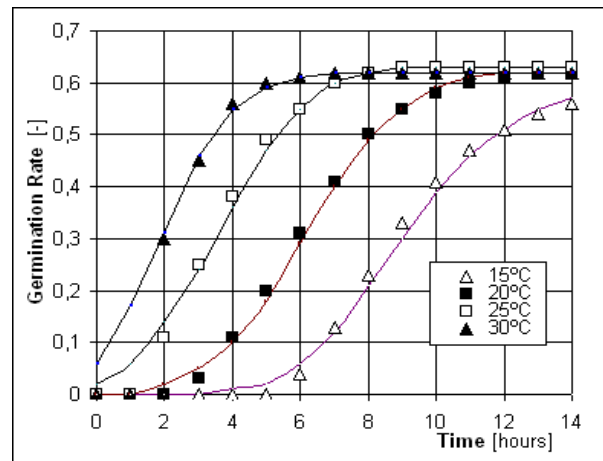


Figure 1. Ratio of seeds germinated in $j \leq 14$ days as a function of the germination time for the samples at temperatures. Symbols are measured values (R_j), the curves drawn are calculated from (11) using the estimated characteristics from (15). Number of seeds $n=500$

B. Results

The three parameters of the germination process were calculated by the method of parameters variation (Appendix II) seeking the minimum of (15)

$$SSR = \sum_{j=1}^J [R_j - p_G F(t_G, \sigma_G, t_j)]^2 = \min \cdot \quad (15)$$

Values of these parameters estimated in such way for the different (k) average germination temperatures ($T = \bar{T}_k$) are shown in Table 1.

Table 1. Estimated values of kinetic and fitting parameters for the average germination temperatures

\bar{T}_k [°C]	15	20	25	30
\hat{t}_G [Days]	9.0	6,2	3.6	2.0
$\hat{\sigma}_G$ [Days]	2,51	2,25	2,09	1.75
\hat{p}_G [-]	0.58	0.62	0.63	0.62
p	0.0001	0.001	0.005	0.01

The calculation of average activation energies we took the logarithm of (13), this way the regression functions will be lines (Figure 2.)

$$\ln t_G = \ln \bar{A} + \frac{\bar{E}_M^*}{R} \frac{1}{T} \quad (16.a)$$

$$\ln \sigma_G = \ln \bar{B} + \frac{\bar{E}_D^*}{R} \frac{1}{T} \quad (16.b)$$

from this the estimated values of average activation energies have been calculated using linear regression (EXCELL). Values are given $\bar{E}_M^* = 73\text{kJ/mol}$ and $\bar{E}_D^* = 21\text{kJ/mol}$.

Variance analysis (ANOVA) was used for the hypothesis test of the regression functions, where we computed F_0 and p level of significance belonging to the sample. If $p < 0.05$ than we accept with a probability of 95% that the measured data can be written with (11) and (16) relations. Values of significance belonging to the kinetics (11) can be seen in Table 1 as those given from defining activation energy are at the mean value $p=0.01$, at the standard deviation $p=0.01$.

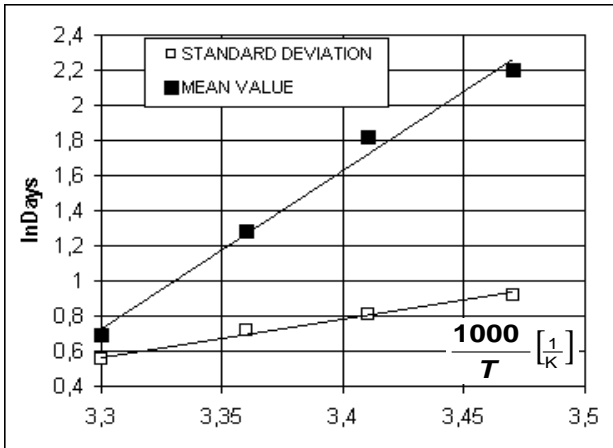


Figure 2. Regression lines (16) for calculation of the average activation energies from the mean values and standard deviations (Table 1.)

IV. DISCUSSION

The kinetics (11) according to Figure 1 and the level of significance ($p < 0.05$) fit to the measuring data showing the validity of the model for description of the germination of reed. The better the fitting is, the higher the mean value of germination in relation to the standard deviation of germination. A considerable difference can be found only in the case of $t_G \approx \sigma_G$ and $t \rightarrow 0$, because in this case $N(t=0, t_G, \sigma_G) > 0$, while, $R(t \rightarrow 0) = 0$ which means the limit of the approximation. The approximation within this range is also corrigible, having concrete assumptions for the distribution function, the mean value and the standard deviation of the different processes [14].

The mean value of the germination time is significantly, while standard deviation of the germination time is marginally influenced by the temperature. With

increasing temperature there is a decrease in both cases. Their logarithms fit to the regression lines at $p=0.05$ significance level, which means that both the mean value and the standard deviation can be defined by Arrhenius-type characteristics according to expression (16).

The different activation energies mean, that the germination process according to the „many-types processes” model can be defined as being a resultant of the processes ($\bar{E}_M^* = 73\text{kJ/mol}$) controlled by material transport and chemical reactions ($E^* = 0-30\text{kJ/mol}$ for transport process, $E^* = 40-60\text{kJ/mol}$ enzyme and $E^* = 100-250\text{kJ/mol}$ non-enzyme controlled processes [15], [16]).

The value of the activation energy belonging to the standard deviation ($\bar{E}_D^* = 21\text{kJ/mol}$) shows that the standard deviation is mostly originated from the transport processes (measured value of pea 18kJ/mol , [14], [17]).

APPENDIX

I. Temperature-dependence of mean value and standard deviation of the process time

Process time being a random variable is a function of two other random variables, namely ε_i^* activation energy and α_i pre-exponential coefficient

$$\tau_i = \alpha_i \exp \frac{\varepsilon_i^*}{RT} \quad (A.1)$$

Expand (A.1) into series according to α_i and ε_i^* around their mean value, $A_i = M\{\alpha_i\}$ and $E_i^* = M\{\varepsilon_i^*\}$ to the first term. It will be

$$\tau_i = A_i \exp \left\{ \frac{E_i^*}{RT} \right\} \left(1 + \frac{\alpha_i - A_i}{A_i} + \frac{\varepsilon_i^* - E_i^*}{RT} \right), \quad (A.2)$$

Form the mean value of (A2) using the formula for the mean value of the sum of random variables is

$$t_i = M\{\tau_i\} = A_i \exp \left\{ \frac{E_i^*}{RT} \right\}, \quad (A.3)$$

as $M\{\alpha_i - A_i\} = 0$ and $M\{\varepsilon_i^* - E_i^*\} = 0$.

Using the formula for the standard deviation of the sum of random variables

$$D\{\tau_i\} = B_i \exp \left\{ \frac{E_i^*}{RT} \right\}, \quad (A.4)$$

where

$$B_i^2 = A_i^2 \left(\frac{D^2\{\alpha_i\}}{A_i^2} + \frac{D^2\{\varepsilon_i^* - E_i^*\}}{(RT)^2} \right) \quad (A.5)$$

they turn on temperature the same way with different pre-exponential coefficients.

II. Method of parameter variation

The minimum of the (15) is given by the following system of equations

$$\frac{\partial SSR}{\partial p_G} = \frac{\partial SSR}{\partial t_G} = \frac{\partial SSR}{\partial \sigma_G} = 0 \quad (\text{A.6})$$

The (A.6) is a non linear equation system, its solving is possible only in a numerical way.

III. Thermal Time

Instead of TT using average activation energy approximation we can use the following parameter

$$Z = t \exp\left\{-\frac{\bar{E}_{Mn}^*}{RT}\right\} \quad (\text{A.8})$$

and

$$Z_G = t_G \exp\left\{-\frac{E_M^*}{RT}\right\} = \bar{A} \cdot \quad (\text{A.9})$$

Expand (A.10) into series around T_B till the first order term

$$Z = \left(\frac{\bar{E}_M^*}{RT_B^2} \exp\left\{-\frac{\bar{E}_M^*}{RT_B}\right\}\right)(T - T_B)t = \text{const} \cdot TT. \quad (\text{A.10})$$

In a specific little range of temperature it will be proportional to thermal time.

REFERENCES

- [1] J. Garcia-Huidobro, J. L. Monteith and G. R. Squire, "Time, temperature and germination of pear millet (*Pennisetum typhoides* S. & H) I. Constant temperature", *Journal of Experimental Botany*, 33: pp. 288-296, 1982
- [2] E. Kebreab and A. J. Murdoch, "Modelling the effects of wather stress and temperature on germination rate of *Orobanche aegyptiaca* seeds", *Journal of Experimental Botany* 50, pp. 655-664, 1999
- [3] F. Forcella, R. L. Arnold, R. Sanchez, and C. M. Chersa, "Modeling seedling emergence", *Field corps Research*, 7, pp. 123-139, 2000
- [4] B. Ekstam and A. Forseby, "Germination response of *Phragmites australis* and *Typha latifolia* to diurnal fluctuations in temperature", *Seed Sci. Res.* 9 (3), pp. 157-163, 1999
- [5] A. Bosch, R. A. Maronna and O. M. Yantorno, "A Simple Descriptive Model of Filamentous Fungi Spore Germination", *Process Biochemistry*, 30, No7, pp. 599-606, 1995
- [6] R. F. Brown and D. G. Mayer, (1988): "Representing cumulative germination. 2. The use of Weibull function and other empirically derived curves", *Ann. Bot.* 61, pp. 127-138, 1988
- [7] L. G. Labouriau, "On the physiology of seed gesmination in *Vicia grainea* Sm.", *Anais da Academia Brasileira de Ciencias*, 42 pp. 235-262, 1970
- [8] K. Bradford, "Water relations in seed germination", In: J. Kigel and G. Galili, eds: "Seed development and germination", *New York, Marcel Dekker*, pp. 351-396, 1995
- [9] S. Covell, R. H. Ellis, E. H. Roberts, and R. J. Summerfield, "The influence of temperature on seed germination rate in grain legumes. I. A comparison of chickpea, lentil, soyabean and cowpea at constant temperatures", *Journal of Experimental Botany*, 37 pp. 1441-1453, 1986
- [10] R. H. Ellis, S. Covell, E. H. Roberts and R. J. Summerfield, "The influence of temperature on seed germination rate in grain legumes. II. Intraspecific variation in chickpea (*Cicer arietinum* L.)", *Journal of Experimental Botany*, 37, pp. 1503-1515, 1986
- [11] R. H. Ellis and P. D. Butcher, "The effects of priming and „natural” differences in quality amongst onion seed lots ont he response of the rate of regmination to temperature and identification of the characteristics under genotyp control" *Journal of Experimental Botany*, 39, pp. 935-950, 1988
- [12] G. Lakner, J. Lakner, T. Naszradi and O. Oláh, "Mathematical modeling of reed germination", *BULLETIN of Szent István University*, Gödöllő, Hungary, 2002
- [13] B. Ekstam, R. Johannesson and P Milberg, "The effect of light and number of diurnal temperature fluctuations on germination of *Phragmites australis*", *Seed Sci. Res.* 9 (1), pp. 165-170, 1999
- [14] G. Lakner and J.Lakner, "Mathematical modelling for stages in germination of common reeds (*Phragmites Australis*)", *Acta Botanica Hungaria*, 52 (3-4) pp.341-361, 2010
- [15] T. P. Labuza, "Self-life dating of foods" *Food and Nutrition Press*, Westport, conn., pp.57-62, 1982
- [16] S. Yan, C. Huawei, Z. Limin, R. Fezheng, Z. Luda and Z. Hengtao, "Development and characterization of a new amylase tipe time-temperature indicator", *Food Control* 19, pp. 315-319, 2008
- [17] A. M.Mayer and A. Poljakoff-Mayber, "The germination of seeds", pp. 37-43, *Pergamon Press*, London. J. R. Pappenheimer, *Physical. Rev.* 33, pp. 387 1963