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## Germination : - A N Formula

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### Introduction

Many seed analysis and authors have tried to derive a formula that evaluates germination tests. The principal aim of to obtain such formula is to present a standard procedure for the viability of a seed lot, the effectiveness of various presowing treatments, and expected seedling survival from field or nursery sowings.

CZABATOR (1962) Proposed a new formula for the objective evaluation of germination tests and also demonstrated the problems which were encountered using previous methods.

He mentions that results of germination tests are too often subjective, with tables of experience, graphs of the course of germination, or statistical transformation of the data being used to qualify the expressed percentage of germination. Furthermore, CZABATOR (1962), after a review of the literature, established that with conventional methods of evaluating the results of germination tests could be different and misleading. Since CZABATOR reviewed the literature most adequately, no further review is necessary. Therefore this discussion will be limited to the validity of CZABATOR's formula, plus the presentation of a new formula for germination evaluation.

### Czabator's Formula

CZABATOR mentioned that a practical formula for evaluating germination tests should meet the following specific requirements:

1. Both speed and totality of germination should be considered, but with more emphasis on germinative energy than on totality.
2. Values should be comparable regardless of the duration of the tests.
3. Differences in frequency of counting germinated seed should not render the results erratic.
4. Each seed lot should be evaluated as a single figure, to allow for direct comparisons and to simplify statistical analysis of the results.
5. A more vigorous lot of seed should be indicated by a higher value, and this value should vary directly with

increased speed of germination, or higher total germination, or both.

6. The formula should be sensitive to relatively minor differences in speed or totality of germination.
7. The calculations should be simple.
8. The application of the formula should not involve drastic changes in commonly accepted methods of conducting and recording germination tests.

CZABATOR's formula is as follows:

$$GV = MDG \times PV$$

where GV is germination value, MDG is mean daily germination calculated as the percentage of full-seed germination at the end of the test divided by the number of days to the end of the test; PV is peak value, or the maximum quotient derived from all of the cumulative full-seed germination percentages on any day divided by the number of days to reach this percentages. The peak value is the mean daily germination of the most vigorous component of the seed lot, and is a mathematical expression of the break, or shoulder, of a sigmoid curve representing a typical course of germination. To demonstrate the formula's use the following table cited by CZABATOR is given in Table 1.

The tables germination value will be:

$$GV = MDG \times PV = 4.81 \times 5.08 = 24.43$$

Table 1. — Cumulative germination percent, T-value

| Days since beginning<br>of test | Cumulative<br>germination percent (germination speed) | T-value |
|---------------------------------|-------------------------------------------------------|---------|
| 8                               | 20                                                    | 2.50    |
| 9                               | 34                                                    | 3.78    |
| 10                              | 42                                                    | 4.20    |
| 11                              | 49                                                    | 4.45    |
| 12                              | 60                                                    | 5.00    |
| 13                              | 66                                                    | 5.08    |
| 14                              | 70                                                    | 5.00    |
| 15                              | 74                                                    | 4.93    |
| 16                              | 77                                                    | 4.81    |

Critical points of CZABATOR's formula

CZABATOR's formula seems to be adequate for the seed germination testing of pines. It has been used by many authors (See all literature cited except BATES and CZABATOR).

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However, a number of critical points might be considered.

1 — The use of such formula means that after reaching a peak value, the more germinating seeds we have the lower the germination values we obtain. Consequently, seed lots which continue to germinate after the peak value will have a lower apparent germination value than those that stopped at the peak. Table 2 (germination of *Pinus ponderosa* at 25° C) demonstrates how erroneous results could be produced by use of this formula. These results will be compared with results of a new formula which is discussed later. The germination value for each day is calculated by supposing that germination ceases after that day. The peak value is 9.50 and if germination ceased after the peak value day, the GV would be (peak value)<sup>2</sup> or 9.50<sup>2</sup> = 90.25, but germination continued thus continuously decreasing the GV, therefore, seed lots in which germination stopped would have higher values for the GV than others that continued. (See table 2.)

### Developing a New Formula

Three factors are very important in evaluating seed germination:

1 — Daily germination speeds: These are computed daily by dividing cumulative germination percentages by the number of days (this is equal to the T value of CZABATOR). Taking the maximum speed germination (which CZABATOR calls peak value) and using only this value in a formula will give erroneous results. Generally peak value cannot represent total speed germination during the length of the test. Table 4 will confirm this suggestion. In table 4 the peak values for both sets are 7 and the DGS for both sets are 5.76. So the GV by CZABATOR's formula is 40.32. However the second seed lot has higher germination speeds than the first seed lot due to higher germination percentages during the days 3rd to 11th.

2 — The germination percent: germination percent could

Table 2. — Germination value

| N<br>(Frequency) | Days Since<br>beginning<br>of test | Cumulative<br>germination | Daily<br>Germination<br>speed<br>(DGS) | $\frac{\sum DGS}{N}$ | Germination<br>value |                   |
|------------------|------------------------------------|---------------------------|----------------------------------------|----------------------|----------------------|-------------------|
|                  |                                    |                           |                                        |                      | by<br>Czabator's     | by new<br>formula |
| 1                | 3                                  | 17                        | 5.66                                   | 5.66                 | 32.03                | 9.62              |
| 2                | 4                                  | 37                        | 9.25                                   | 7.45                 | 85.50                | 27.58             |
| 3                | 5                                  | 47.5                      | 9.50                                   | 8.13                 | 90.25                | 38.64             |
| 4                | 6                                  | 52.5                      | 8.75                                   | 8.29                 | 83.12                | 43.52             |
| 5                | 7                                  | 54.5                      | 7.78                                   | 8.18                 | 73.91                | 44.62             |
| 6                | 8                                  | 57                        | 7.12                                   | 8.01                 | 67.64                | 45.65             |
| 7                | 9                                  | 58.5                      | 6.50                                   | 7.79                 | 61.75                | 45.57             |
| 8                | 10                                 | 61                        | 6.10                                   | 7.58                 | 57.95                | 46.23             |
| 9                | 11                                 | 65                        | 5.90                                   | 7.39                 | 56.06                | 48.03             |
| 10               | 12                                 | 68.5                      | 5.70                                   | 7.22                 | 54.15                | 49.45             |
| 11               | 13                                 | 72.00                     | 5.53                                   | 7.07                 | 52.53                | 50.90             |
| 12               | 14                                 | 75                        | 5.35                                   | 6.92                 | 50.82                | 51.90             |
| 13               | 15                                 | 78                        | 5.20                                   | 6.79                 | 49.40                | 52.65             |
| 14               | 16                                 | 80                        | 5.00                                   | 6.66                 | 47.50                | 53.28             |
| 15               | 17                                 | 80                        | 4.70                                   | 6.53                 | 44.65                | 52.24             |
| 16               | 18                                 | 81                        | 4.50                                   | 6.40                 | 42.75                | 51.84             |
| 17               | 19                                 | 81                        | 4.26                                   | 6.28                 | 40.47                | 50.85             |
| 18               | 20                                 | 82                        | 4.10                                   | 6.16                 | 38.95                | 50.51             |

2 — If we look at the germination value (column of Table 2), we see that all the values calculated for the 3rd through 9th days are higher than the germination percent values. Therefore, using CZABATOR's formula gives germination values which are higher than actual germination percentages. Then CZABATOR's formula would have us believe 90 seedlings could be expected on the fifth day of the test, when only 47 germinated under the controlled condition of the lab.

3 — The germination value calculated by this formula, for pine and other tree seeds gives results which are far less than would be expected for seedlings. For example, *Pinus ponderosa* cultivated at zero bar on standard blue blotter paper shows 78 percent of germination after 26 days (Table 3). peak value occurred after 5 days of culture which was 5.70; MDG is 3. Therefore GV = MDG × PV = 3 × 5.70 = 17.1, which seems to be much less than the expected survival rate of nursery seedlings. If germination ceased after peak day, we might expect 32 seedlings from the same seed lot (5.70<sup>2</sup> = 32.49), and if germination ceased after 10 days, we could expect 25 seedlings (4.45 × 5.70 = 25).

4 — This formula does not define the length of the test, which could totally change the test results and interpretation of the data.

be calculated in the formula as independent of time, because the time factor or length of germination is already considered in daily germination speed.

3 — Length of test: the formula must be suitable to enumerate the length of the germination test to its termination.

Considering these three factors and observation on many germination tests of various species (*Pinus eldarica*, *Pinus ponderosa*, *Pinus taeda*, *Eucalyptus* spp., *Juniperus vir-*

Table 3. — Germination percent of *Pinus ponderosa* at 25° C, at zero moisture stress, based on amount of 400 seeds

| Days | Germ.<br>percent | GS   | Days | Germ.<br>percent | GS   |
|------|------------------|------|------|------------------|------|
| 3    | 4.0              | 1.33 | 15   | 55               | 3.66 |
| 4    | 14.5             | 3.62 | 16   | 58               | 3.62 |
| 5    | 28.5             | 5.70 | 17   | 59               | 3.47 |
| 6    | 29.5             | 4.91 | 19   | 65               | 3.42 |
| 7    | 33.5             | 4.78 | 21   | 67.5             | 3.21 |
| 8    | 37.0             | 4.62 | 23   | 74.5             | 3.23 |
| 9    | 41.0             | 4.55 | 24   | 76.0             | 3.16 |
| 10   | 44.5             | 4.45 | 25   | 76.0             | 3.04 |
| 12   | 47.5             | 3.95 | 26   | 78.0             | 3.00 |
| 14   | 54.5             | 3.89 |      |                  |      |

Table 4. — Germination speed

| Days | First seed lot |             | Second seed lot |             |
|------|----------------|-------------|-----------------|-------------|
|      | germ. percent  | germ. speed | germ. percent   | germ. speed |
| 3    | 3              | 1.00        | 15              | 5           |
| 4    | 9              | 2.25        | 22              | 5.50        |
| 5    | 15             | 3.00        | 30              | 6.00        |
| 6    | 20             | 3.33        | 37              | 6.16        |
| 7    | 25             | 3.57        | 44              | 6.28        |
| 8    | 30             | 3.75        | 52              | 6.50        |
| 9    | 41             | 4.55        | 60              | 6.66        |
| 10   | 53             | 5.30        | 68              | 6.80        |
| 11   | 60             | 5.45        | 76              | 6.90        |
| 12   | 84             | 7.00        | 84              | 7.00        |
| 13   | 87             | 6.69        | 87              | 6.69        |
| 14   | 90             | 6.42        | 90              | 6.42        |
| 15   | 93             | 6.20        | 93              | 6.20        |
| 16   | 95             | 5.93        | 95              | 5.93        |
| 17   | 98             | 5.76        | 98              | 5.76        |

*giniana*, *Juniperus polycarpus*, *J. scopulorum*, *Quercus* spp. etc.) and including data available from seed laboratories and nurseries, we propose the following formula.

$$GV = \frac{\sum DGS}{N} \times (GP \times 10)$$

where GV is germination value, DGS is daily germination speed which is computed by dividing cumulative germination percent by the number of days since beginning the test. DGS should be calculated every day but if no data are available each day, then could be calculated at two day intervals. N is frequency or number of DGS that are calculated during the test. GP is germination percent at the tests conclusion and is used in the formula as the number of germinated seeds over 100 (for example if the germination percent at the end of the test is 89. in the formula will be  $\frac{89}{100}$ ). So the formula could be explained as  $GV = \frac{\sum DGS}{N} \times \frac{\text{number of germinated seed}}{100} \times 10$ . Then number 10 is a constant through experience and by many germination tests, it has been determined that results of the formula using the constant can be close to the number of surviving seedlings, hence increasing the formula's objectivity.

#### Formula's Indication of Test Termination

It is very important to determine the length of the test. A few weak or sluggish seeds which germinate the last days of the test could change the interpretation of the results. There are no standard methods of determining the end of a test. Periods suggested in the literature for length of germination, for example 42 days for slash pine and longleaf pine, or 25 to 30 days for a dry sown of a Southern pine and 10—20 days for a stratified lot etc. are only general ideas for the length of the germination completion. These figures are not utilized in the formula because the length of germination tests are related to many factors such as physical, chemical condition of the culture medium, seed lots and pretreatments. So it is very variant within a same species. Seed germination generally begins to wane in the last days of the test, and this slow germination could be continued for several days or weeks. So when can the test be considered terminated?

CZABATOR admits that his formula fails to give a constant difference in germination values between any lots that vary in test period length. BATES (1913) stated that the germinative energy period ends when germination on any day drops below 2 seedlings, and on the following days the number of seedlings does not exceed 2. This kind of inter-

pretation could not be applied in all cases and with all species, because:

- (1) It depends on the rate of germination speed in early days of the test, i.e., if there were very high germination speeds in the first stage of germination the number 2 could conceivably be too low, or if the rate of germination speeds were low in the early stage the number 2 could be too high.
- (2) Some seeds have a rate of germination per day that is not more than 2 or 3. For example *Juniperus scopulorum*, when not chilled for a sufficient period of time to break dormancy has a slow rate of germination. This slow rate can also be expected for immature seeds, or when moisture stress of the medium is inadequate (Such as a culture of seeds on high moisture stress, produced by polyethylglycol, or d-manithol, etc.). The formula presented herein is suitable of determining the end of the experiment very satisfactorily. If we calculated the GV from the beginning of the test it establishes a curve based up GV and days. Following are two examples of how the formula can assess the end of the test.

A. In *fig. 1* curve A tends to increase its GV each day of the experiment. This indicates ideal germination and no sluggish or weak seeds. The maximum GV for this curve is at the end of the test where the curve is at its highest point.

B. Curve B of *fig. 1* increases and reaches to maximum GV, for example, at the 25th day, then begins to decrease due to low germination rates as compared to the previous days. So the top of the curve B will be the maximum GV (GV = 55) and also the end of the test. This maximum GV will be the true germination value for the lot. The maximum GV was exemplified in table 2 where the GV was at its highest on the 16th day, at 53.28, and not on the 20th day where the GV had begun to decrease at 50.51.

#### The Formula's Advantages

1. An exact definition of the germination value is the expected seedlings from field or nursery sowings. The value obtained by this formula is very close to the number of seedlings survived in the field. To confirm this suggestion the following laboratory and field experiment analyses of *Pinus eldarica* and *P. ponderosa* seed germination is given here. The soil in this experiment was equal volume of peat and Gross vermiculite which were kept well wet (*Table 6*).

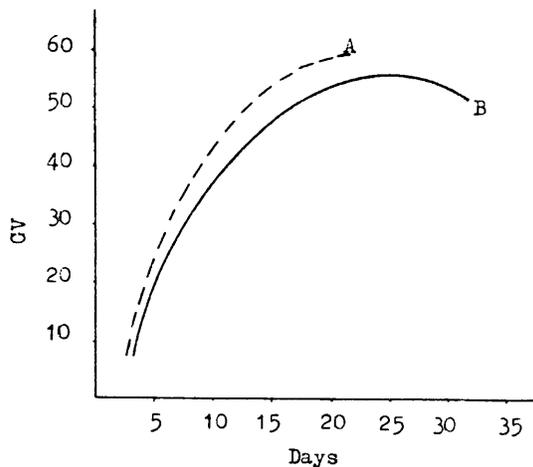


Figure 1. — Germination value.

Table 5. — A Table Model for Facilitating the Mechanism of Calculations

| D  | GP | $\Sigma$ DGS | DGS $\div$ N = $\frac{\Sigma$ DGS}{N} | GV    |
|----|----|--------------|---------------------------------------|-------|
| 3  | 6  | 2.00         | 2.00 $\div$ 1 = 2.00                  | 1.20  |
| 4  | 15 | 3.75         | 5.75 $\div$ 2 = 2.87                  | 4.30  |
| 5  | 20 | 4.00         | 9.75 $\div$ 3 = 3.25                  | 6.50  |
| 6  | 30 | 5.00         | 14.75 $\div$ 4 = 3.68                 | 11.04 |
| 7  | 38 | 5.42         | 20.17 $\div$ 5 = 4.03                 | 15.31 |
| 8  | 48 | 6.00         | 26.17 $\div$ 6 = 4.36                 | 20.92 |
| 9  | 65 | 7.22         | 33.39 $\div$ 7 = 4.77                 | 31.00 |
| 10 | 72 | 7.20         | 40.59 $\div$ 8 = 5.05                 | 36.50 |

D — days since beginning of test; GP — germination percent; DGS — daily germination speed; N — frequency, is written beside DGS to avoid any error in calculation. GV will be very easy to obtain by multiplying the figures of  $\frac{\Sigma$ DGS}{N} by GP, then dividing by 10, for example for the 3rd day will be  $6 \times 2 = 12$  but will be written 1.2 because the index of formula is  $\frac{10}{100}$  (10 for constant, 100 for percent). DGS and  $\frac{\Sigma$ DGS}{N} and GV are rounded off to two decimal places.

- The formula does not give erroneous results due to different germination speeds which occur during the test. In table 4 CZABATOR'S formula gives the same GV of 40.32 for two sets of seed lots which are indeed different, but the proposed formula gives 45.2 ( $\frac{\Sigma$ DGS}{N} \times GP \times 10) for the first seed lot and 61.27 for the second lot.
- The formula determines the end of the test by a mathematical operation, hence a standard model for the test

length common to all tree seeds. By this formula the quality of the seed lot will not decrease by sluggish seeds which germinate on the last days. This formula is very sensitive in its determination of test termination for any kind of germination (slow or fast). In table 2 the number of germinated seeds per day from the 3rd through the 8th days was respectively 17, 20, 10, 5, 2, 3 but on the 9th day this number dropped to 1.5 per day, so the GV decreased from 45.65 to 45.57 showing that a high drop of germination could define the end of the experiment, but there was more germination on the 10th and following days, then the GV increased again and arrived at its maximum on the 16th day which is the end of the test. In the case when the number of germinated seeds per day is very low for example 2 or 3 seeds per day, the formula will show the end of the experiment when the number of seeds per day drops to less than  $\frac{1}{2}$  (one seed germinated per two days). It also depends on the number of seeds germinated in the beginning of the test and the day on which the seed lot started to germinate. Generally in common germination of pines, the formula shows the end of the test when the number of germinated seeds drops to 1.5 per day (in a seed lot of 100 seeds).

- This formula gives better results than CZABATOR'S formula for the cases where daily germination records are not possible and data is available from 2 day intervals.
- The formula could be applied not only to pines, but for all tree species with an average daily speed of germination ( $\frac{\Sigma$ DGS}{N}) less than 10. There are very few tree species that have an average germination speed more than 10. If the average germination speed is 10 or more than 10, for example, 100 percent germination in 10 days the GV will be 100. therefore we have to modify the constant 10 of the formula to 7 or 8 only for such species if they exist.

Such species, however, do not need GV calculations because they are so vigorous that the expected seedlings from the lots are very close, or are the same as the germination percent.

#### Mechanism of Calculation for Germination Value

The germination value could be early calculated by the formula  $GV = \frac{\Sigma$ DGS}{N} \times GP \times 10 as explained previously.

This germination value need not be calculated for each day as it is done in Table 2, but can be calculated for the last days and from there we can work back from the end of the test to get the maximum GV.

If it is desired to calculate the GV for each day establishment of the table 5 facilitate the calculations.

Table 6. — Germination value

| Cumulative <sup>1)</sup> germination (%) in petri plates |                     | Germination value by CZABATOR'S |                 | Germination value by new formula |                 | Number of seedlings <sup>2)</sup> survived in the field during 8 weeks (%) |                 |
|----------------------------------------------------------|---------------------|---------------------------------|-----------------|----------------------------------|-----------------|----------------------------------------------------------------------------|-----------------|
| <i>P. eldarica</i>                                       | <i>P. ponderosa</i> | <i>P. eld.</i>                  | <i>P. pond.</i> | <i>P. eld.</i>                   | <i>P. pond.</i> | <i>P. eld.</i>                                                             | <i>P. pond.</i> |
| 94 $\pm$ 0.20 <sup>a)</sup>                              | 81 $\pm$ 0.50       | 31.68                           | 33.44           | 61.24                            | 53.28           | 75 $\pm$ 3 <sup>b)</sup>                                                   | 66 $\pm$ 2      |

- Seven hundred seeds were used in seven replications
- 2100 seeds were used in field experiment in seven replications
- Mean  $\pm$  S. E. of all seeds in seven replications
- Mean  $\pm$  S. E. of all seedlings in seven replications

### Summary

The formula proposed here for evaluation of germination test has taken into account both daily germination speed for each day, and also total germination percent which gives as composite value called germination value

$$GV = \frac{\sum DGS}{N} \times GP \times 10$$

GV is germination value. DGS is daily germination speed which is obtained by dividing the cumulative germination percent by the number of days and can be calculated for each day or at 2 day intervals depending on collection of data. GP is the percent of germination at the end of the test. N is frequency of DGS. This formula indicates when test could be considered terminated, by determining the maximum GV. The proposed formula is applicable for most tree seeds, and is practically very objective and it could be used to standardize germination value in seed germination tests.

*Key words:* New formula, seed germination value.

### Zusammenfassung

Die in dieser Arbeit vorgestellte Formel für die Beurteilung von Keimtests berücksichtigt sowohl die tägliche Keimgeschwindigkeit, als auch das Gesamtkeimprozent. Aus beiden zusammen ergibt sich dann der sog. Keimwert nach der Formel:

$$GV = \frac{\sum DGS}{N} \times GP \times 10$$

GV ist der Keimwert, DGS die tägliche Keimgeschwindigkeit, welche ermittelt wird, indem man das kumulative Keimprozent durch die jeweilige Anzahl der Tage dividiert. Es kann sowohl für jeden Tag einzeln, als auch für Intervalle von 2 Tagen errechnet werden, je nach Zweckmäßigkeit der Datenerhebung. GP ist das Keimprozent am Ende des Tests und N die Häufigkeit der DGS.

Durch die Bestimmung des maximalen GV, zeigt diese Formel auch an, zu welchem Zeitpunkt der Test als be-

endet angesehen werden kann.

Aufgrund der nach unseren Feststellungen objektiven Ergebnisse dieser Formel, könnte sie zur Standardisierung von Samenkeimtests dienen.

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### Short Note

## Investigations on the Dependence of Flowering in Spruce (*Picea abies* (L.) Karst.) upon Age and Hormone Treatment

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### Introduction

Since 1950 seed orchards have been established for all important forest tree species. Unfortunately the beginning of fructification, especially in Norway spruce, has been delayed for a long time. Therefore many scientists all over the world (MELCHIOR, 1961 a, 1961 b; HASHIZUME, 1968; and many others) have worked to solve the problems of fructification in forest tree species. Again and again it appeared in these studies that fructification in Pinaceae depends very strongly on the age of the plants. In 1973, investigations were started at our institute to elucidate

whether Norway spruce plants react differently to treatments with growth regulators at different ages.

### Materials and Methods

In the spring of 1973, 20 3-year-old seedlings and grafts of spruce were sprayed in a greenhouse 3 times at weekly intervals with 100 ppm gibberellic acid (GA<sub>3</sub>), 1000 ppm indoleacetic acid (IAA) or 10 ppm kinetin (6-furfurylamino purine = KI) as soon as the buds began to open. Subsequently the plants were sprayed 3 times with 2000 ppm CCC ((2-chloroethyl)-trimethyl-ammonium chloride) or 1000 ppm