

# Influence of temperature regime on the germination of seeds of ornamental grasses

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## Abstract

The results obtained show that of the ten varieties of turf grass tested: *Agrostis stolonifera*, *Cynodon dactylon* 'Ornamental', *C. dactylon* 'Premium', *Festuca arundinacea* 'HounDog', *F. arundinacea* 'Nebula', *F. arundinacea* 'Starlett', *F. rubra*, *F. rubra* var. *commutata*, *Lolium perenne* 'Fabian' and *Poa pratensis* 'Yvette', seven varieties show satisfactory germination under the temperature regime typical for the end of October and November. The closest germination to the control, that was held at 22 C<sup>0</sup>, was recorded in *L. perenne* 'Fabian', where germination decreased by only 4.06 %, a significant decrease in germination was found in *F. rubra* – by 10.95 %, *P. pratensis* "Yvette" – by 12.5 % and *F. rubra* var. *Commutata* – by 19.46 %. For varieties of *F. arundinacea*, the decline in germination is significantly higher: 21.76 % in 'Starlett', 30.53 % for 'HounDog' and 37.9 % for 'Nebula'. The temperature regime also affected the rate of germination as the first germinated seeds were reported 2-4 days later in the seeds kept cool. Three varieties, namely *A. stolonifera*, *C. dactylon* 'Ornamental' and *C. dactylon* 'Premium', showed very low germination or no germination was observed in the temperature regime typical for the second half of autumn.

The results obtained by us bring further clarity to the problem of late sowing of turf grass to produce sod or directly create a vegetative coating. The results obtained show that species with lower requirements for germination and sprouting temperatures such as *L. perenne*, *F. rubra* and *P. pratensis*, or sowing norms, may be selected to compensate for the lower germination.

**Key words:** turf grasses; germination; temperature regime

## INTRODUCTION

The seeding of areas to produce sod in the second half of the autumn is required in individual cases for producers in Bulgaria. The reasons for this are varied, but among the main ones are the late release of the areas and the preference for winter seeding. Seeding before winter has many advantages that are well appreciated by the sod producers. In the Sofia Plain, the grass growth season usually ends in mid-December, although in individual years, temperatures remain positive until the end of December. The fall of temperatures persistently below 0°C at the end of November or the beginning of December is rare. In addition, at least 10 days in February, as

well as most of March, have temperatures allowing the growth and development of grass species. On the other hand, changing and unpredictable conditions in early spring make seeding difficult and more uncertain than autumn. Even if the density of the late autumn crops is lower than optimal, this is usually compensated by higher levels of vegetative propagation of the plants sprouted in autumn and, if necessary, can be compensated by additional seeding for achieving optimal density.

In the scientific literature, there is a large amount of research on the germination of seeds of various fodder and turf grass species. Reported results are not always consistent, and the tested temperature regimes are usually above + 10°C. The opti-

mal temperatures for germination of grass seeds vary between different species. For example, for *L. perenne*, maximum germination was obtained at 25°C constant temperature (Javaid et al., 2022). *L. perenne* seeds have maximum germination and optimal germination time at different temperature regimes (night/day): 15/25, 20/25, 20/30 and 25/30 °C (Shen et al., 2008). In *F. arundinacea* seeds, maximum germination was observed at different temperatures: 20-25°C (Boyce, et al., 1976); 16-19°C (Danielson & Toole, 1976); 15-30°C (Sharifiamina et al., 2016). The minimum temperature for germination of *F. arundinacea* seeds is 3.5°C (Sharifiamina, et al., 2016). When examining the temperature requirements of 11 varieties (which belong to several species of the genus *Festuca*) large differences were found, with maximum germination being obtained in different varieties in the range of 10 to 30°C (Palazzo, 1997). Results obtained for *C. dactylon* seeds show various optimal temperature regimes for germination and sprouting 10/40°C. High germination is also obtained at a constant temperature of 25 or 30°C (Young et al., 1977); 25-35°C for whole seeds and 20-30°C for husked (Evers & Parsons, 2009).

The germination rate also differs between grass species. For example, at a constant temperature of 25°C *L. perenne* shows the fastest germination rate of all tested species — 92% in 7 days. *F. arundinacea* had a germination rate of 99% on day 9, and *A. stolonifera*, respectively, 89% also on the 9th day, the lowest germination rate was recorded for *P. pratensis* — 61% on day 30 (Charif et al., 2019). The rate of germination is dependent on temperature. Germination of *L. multiflorum*, *Bromus catharticus* and *F. arundinacea* seeds shows significant delay at 15 and 10°C. In addition, for *F. arundinacea* the germination rate at 10°C is lower compared to higher temperatures (Hill et al., 1985).

Although the optimal temperature regimes for germination and sprouting of most grass species have long been established in several cases seeding under unfavourable conditions is necessary for various reasons. For example, *C. dactylongrown* in areas with cooler climate should be sown as early as possible. This is done to early cover the soil to suppress the appearance of weed plants and strengthen the plants until winter to minimise the damage from frost. In experiments with early seeding of *C. dactylon*, the seeds were found to remain latent until

suitable soil temperatures were reached, without their remaining in the soil adversely affecting germination. After all, early seeded areas are of equal density or superior to those seeded later (Shaver et al., 2006). The timely repair of the grass cover in the golf courses before the start of the high season is of great importance. However, for this purpose, the seeds of *A. stolonifera* must be seeded at temperatures significantly lower than the optimum for the species. The germination of *A. stolonifera* seeds was found to be unreliable, and significant differences were also found in the ability of seeds of different varieties to germinate and sprout at low temperatures (Heineck et al., 2019). Differences in germination capacity at lower temperatures have been found not only between varieties but also between different samples of the same variety. This is explained by the different environmental conditions of production as well as differences in the methods of processing and storage of the seeds (Carroll et al., 2020).

Many factors affect the germination of grass seeds, their requirements for temperature regime and germination speed (Baghdadi et al., 2013; Sveinsson & Björnsson, 1994). In *L. perenne*, *F. rubra* and *P. pratensis*, a link was found between seed size, germination rate and germination speed. The largest germination rate at the lowest temperature is obtained for the seeds of the largest fraction (Larsen & Andreasen, 2004). The requirements for the temperature regime are also influenced by the temperatures during the ripening of the seeds and during their storage (Boyce et al., 1976). The storage of seeds of *Festuca spp.* between 120 and 150 days after harvesting increases germination and vitality (Dragičević et al., 2010).

Seed germination can also be influenced by several other factors, for example, pre-seeding preparation; clothing; temperature amplitudes; presence, spectrum, intensity and periods of light, etc. (Frett & Pill, 1996; Tool & Borthwick, 1971; Leggatt, 1946; Danielson & Toole, 1976).

The main aim of this study is: to establish the impact of temperature regime in late autumn on the germination of seeds of grass varieties used in practice; to identify differences in germination between species and varieties; to determine which species and varieties are more reliable for late seeding; to quantify the decline in germination that can be used in the future as a guideline to compensate for seed-

ing norms; to identify delays in germination, which may provide further guidance on acceptable time limits for late seeding.

## MATERIALS AND METHODS

### Plant material

The plant material used is of the following origin: *Festuca Rubra* and *F. rubra* var. *Commutata* are received by “Sortovi Semena Ltd”, produced in Bulgaria in 2020; *Cynodon dactylon* “Ornamental” and *C. dactylon* “Premium” were provided by Totex EOOD, produced in the USA in 2020; *Agrostis stolonifera* was obtained by the same company, the seeds were produced in 2020 in Denmark. Seeds of *F. arundinacea* “HounDog”, *F. arundinacea* “Nebula”, *F. arundinacea* “Starlett”, *Lolium perenne* “Fabian”, and *Poa pratensis* “Yvette” were provided for this study by “Totex EOOD”, produced in Denmark in 2021.

### Setting up the experiment

The seeds were placed in glass petri dishes with a diameter of 100 mm and a height of 20 mm, 100 seeds in each. Under the seeds were placed two sheets of filter paper cut in the shape of the petri dish, and a sheet of filter paper was placed over them. Each petri dish was moistened with 5 ml of distilled water, which moisturized the filter paper, keeping a small amount of water at the bottom of the petri dish. From each variety 10 petri dishes were laid, 5 of which were placed under a constant temperature regime of  $22 \pm 1$  °C in a thermostat in the dark. The other five were placed in a dark cabinet in an open room with a north exposure, in which the air temperature naturally copied that outdoors.

Two maximum-minimum thermometers were placed, the data from which were recorded twice a day at 7 and 19 hours. One thermometer was placed in the seed cabinet, while the second was outdoors on a northern wall (not lit by the sun). Maximum, minimum and instant temperature were recorded.

The average daytime and average night temperatures were recorded as an average between the lowest and the highest values of the day, respectively nights. The average daytime and average night temperatures were used to establish the average day-to-day temperature. By averaging the mean daily

or mean night temperatures over the 30-day period of the experiment, the average day temperature and the average night temperature for the period were obtained.

During the experiment there was a loss of water from the petri dishes, which is why they were further moistened with distilled water, through a pipette of 1 ml.

The experiment was set on 25 October 2021 and lasted until 25 November 2021.

### Reporting of results and statistical processing of data

The results were obtained by multiple counting of germinated seeds. The first counting was made on the 6th day after the start of the experiment, after which every day counting were carried out until the 30th day. The germinated seeds were removed from petri dishes to eliminate the risk of recounting and to facilitate the counting.

The time, in days, to reach 50% germination ( $DG^{50}$ ) was calculated as the average value between the day from the start of the test on which germination equal to or greater than 50% of the final germination was recorded and the day on which a germination equal to or greater than 50% of the planted seeds was reported. When final germination is 100%, the two values match and  $DG^{50}$  is the number of days to report germination equal to or greater than 50% of seed set. When the final germination rate is lower than 50%, the  $DG^{50}$  value is equal to the average value of the day on which germination equal to or greater than 50% of the final and the last day on which germination was recorded for the sample was recorded.

Germination rate index (GRI) is calculated according to Maguire (1962). It indicates the degree of germination of the seeds. For the calculation of the index, the number of germinated seeds at each reading is divided by the days from the beginning of the test, after which all the resulting values are aggregated.

The temperature sum is the sum of temperatures above + 5 °C by hours for the whole previous period. Hours with a temperature below + 5 °C are not aggregated.

The data were subjected to a one-way variance analysis (ANOVA) and the Duncan test with multiple ranges at probability level  $p < 0.05$  (SPSS, IBM) was used.

## RESULTS AND DISCUSSION

The results obtained from the measurement of the temperature regime at the end of October and November show that the average temperatures are close to the minimum ones indicated in the literature for most grass species (Table 1). The average temperature for the entire period of 30 days is +6.8°C, but germination is mostly realized during the first and partly during the second ten days, which is why, in our opinion, the temperatures in this period are of greater importance. Despite the low average temperatures, the maximum temperatures almost every day exceeded +10°C, not infrequently reaching values of 15 - 17°C. The minimum night temperatures on most nights were in the range between +2°C and +4°C. Several times during the experiment, a temperature of +1°C was recorded, and once during the 3rd ten-day period a temperature of 0°C was recorded. Compared to outdoor temperatures, the temperatures at which the seeds were planted show weaker dynamics, and in a day-to-day plan this mainly affects the absolute maximum and absolute minimum temperatures, which were usually 1-2°C higher, respectively lower, outdoors. We assume that the experimental design implemented in this way satisfactorily reproduces the temperature regime in the upper soil layer where the seeds would be located during real seeding. A disadvantage of the experimental design is the inability to capture the effect of sunlight and possible heating of the soil surface. On most days at the time of the experiment, there were at least 4 hours of sunshine. Bearing this in mind, we can assume with a high degree of certainty that to some extent the temperature regime reported by

us for the period is more unfavorable than the real temperature regime in the soil. However, we should note that in the same period there were at least 10 cases of frost and temperatures falling to and slightly below 0°C outside, which would have worsened the conditions for the seeds located immediately on and below the soil surface.

Germination of the studied grass species varied significantly, both at the constant temperature regime and in the samples kept at a temperature regime specific to late autumn (table 2). First of all, the low germination rate was clearly observed in both varieties of *C. dactylon*. By direct seeding in the spring from the same batch of seeds we obtained 45 – 61% germination (unpublished data). The reason for the low germination in this particular case is probably related to the unfavorable temperature regime, which is significantly below the optimum for the species of 25-30°C, but it may also be partly related to the cultivation of the seeds in the dark. Germination of the seeds of *C. dactylon* at the temperature regime specific to late autumn was not observed. This can be explained by the species' high requirements for temperatures during germination. Another species with low germination at temperatures characteristic of late autumn is *A. Stolonifera*. Unlike *C. dactylon*, *A. stolonifera* is a cool climate grass. Although the average temperature for the period was below the minimum temperature suitable for seeding this species (+7°C), germination was recorded, but it was 6 times lower than that observed at a constant temperature of 22°C. The remaining 7 varieties showed significant germination under the temperature regime specific to late autumn. *L. perenne* 'Fabian' showed the best germination at a

**Table 1.** General characteristics of the temperature regime in late autumn, in which the germination of seeds of 10 varieties of grass was tested

Temperature °C	10-day			Total for the period
	1-st	2-nd	3-rd	
Average daytime t°C	9,375	8,35	8,1	8,61
Average night t°C	5,38	5,2	4,48	6,16
Average 24-hour t°C	7,38	6,8	6,3	6,8
Average maximum t°C	12,7	12,55	12,45	12,57
Absolute maximum t°C	16	17	16	17
Average minimum t°C	3,95	3,65	1,7	3,1
Absolute minimum t°C	1	1	0	0

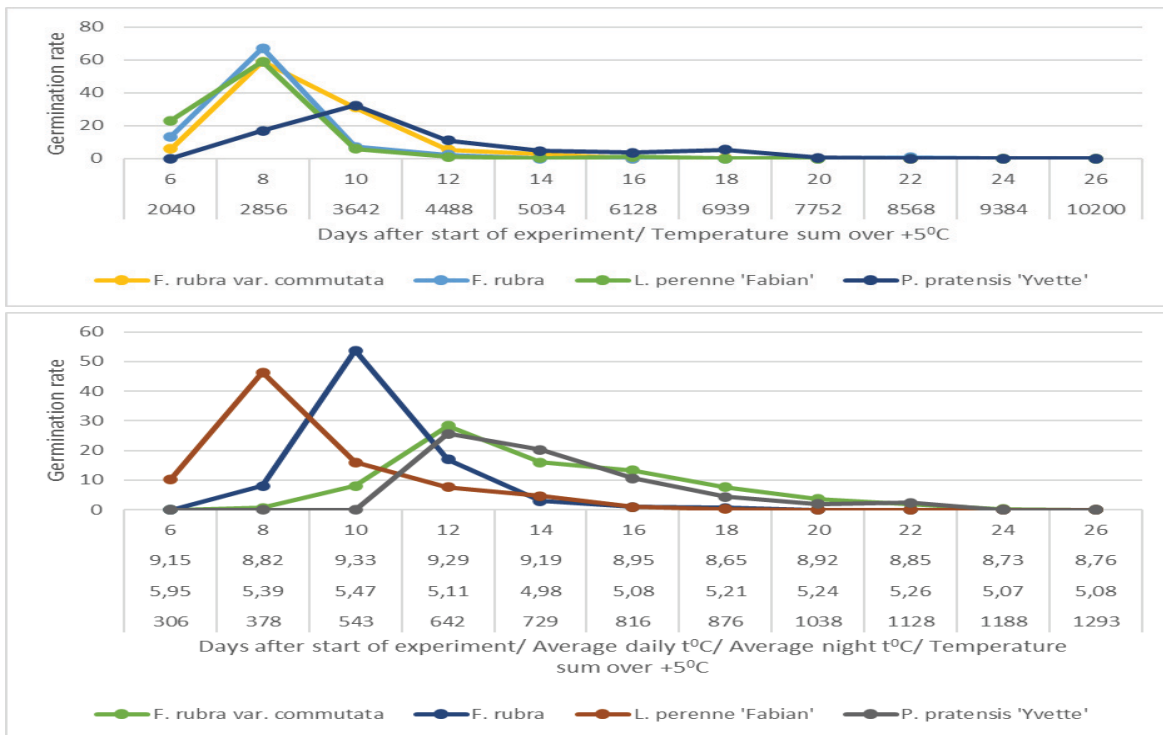
temperature regime specific to late autumn, which was the highest both in absolute terms and as a decrease compared to the germination recorded at a constant temperature of 22°C. This species is the only one in which no statistically significant difference in germination can be found at the two temperature regimes, and there is practically no difference in DG<sup>50</sup>, which for all the tested samples is on the 8th day. However, there is a decrease in GRI under a temperature regime specific to late autumn, which is associated with a certain delay in germination. The second highest germination rate was reported for *F. Rubra* 84% and *F. Rubra var. Commutata* 80%, the difference with the germination of the samples kept at 22°C in both species is significant for all indicators. In a temperature regime specific to late autumn, *F. Rubra* showed a decrease in germination by 10.3%, DG<sup>50</sup> was reached 2 days later, and due to a significantly extended germination time, GRI was 3.48 lower. In *F. Rubra var. Commutata* the decrease in germination was even greater (19.99%), and the number of days to reach DG<sup>50</sup> increased by 5.66. Although *F. Rubra* and *F. Rubra var. Commutata* lose some of their germination in

the temperature regime specific to late autumn, it remains very high compared to *F. Arundinacea*. At a temperature regime specific to the late autumn, *P. pratensis* 'Yvette' showed a decrease in germination by 9.33% compared to seeds kept at 22°C. Although the achieved germination rate of 65.33 is relatively low, we consider that the actual reduction related to temperature conditions is acceptable, therefore we consider this strain undemanding enough to be successfully used for late autumn seeding.

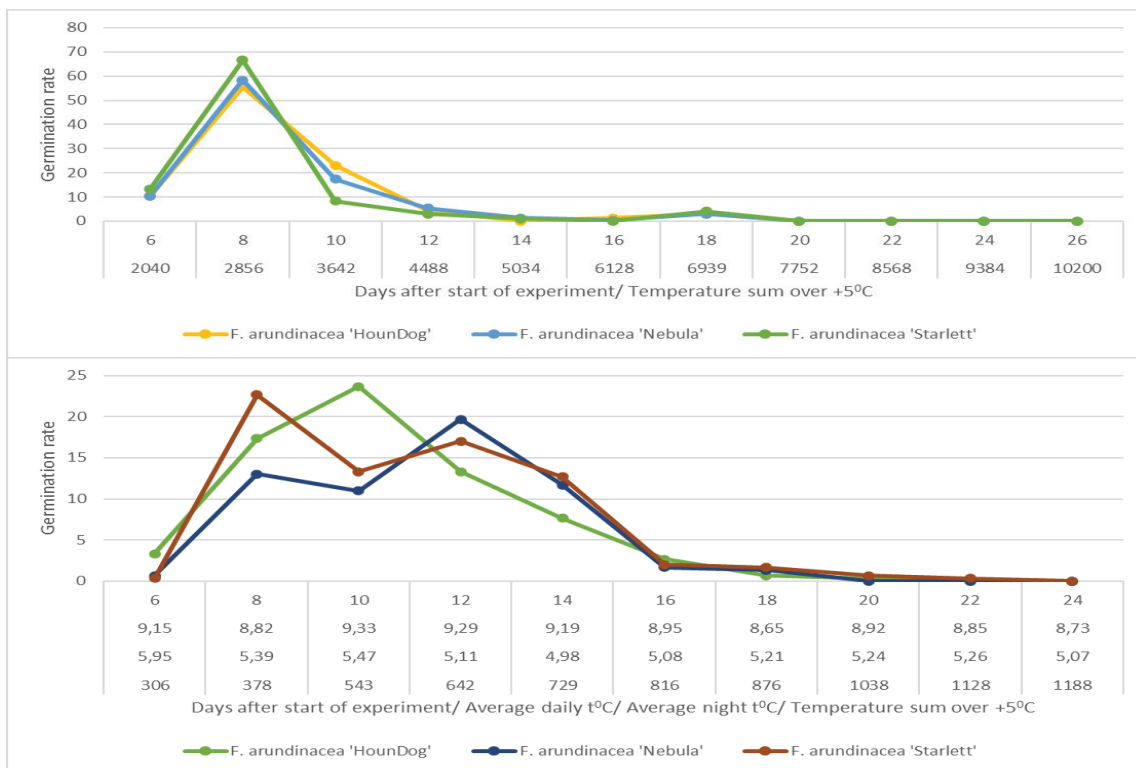
In the varieties of *F. arundinacea*, the decrease in germination, both in absolute terms and as a percentage compared to the germination reported for the seeds kept at 22°C, is significant. Compensating the reduced germination rate (25-35%) by increasing the seeding rates seems economically ineffective. On the other hand, it is an important component of grass mixtures due to its relatively high drought resistance and resistance to high temperature stress. Our results show significant varietal differences in *F. Arundinacea*. There is a significant difference in germination reduction between 'HounDog' and 'Starlett' on the one hand and 'Nebula', on the other (Table 2). We consider it appropriate to study a larg-

**Table 2.** Average final germination, %; average number of days to reach DG<sup>50</sup>; average values of GRI; calculated respectively for the seeds kept at 22°C and at a temperature regime specific to late autumn

Species/ Variety	Germination		DG <sup>50</sup>		GRI	
	Et 22 °C	TR l. autumn	Et 22 °C	TR l. autumn	Et 22 °C	TR l. autumn
<i>A. stolonifera</i>	63 ± 13,74 <sup>b</sup>	11,67 ± 3,21 <sup>a</sup>	13,66 ± 2,31 <sup>b</sup>	22 ± 1,15 <sup>d</sup>	5,82 ± 1,87 <sup>b</sup>	0,56 ± 0,15 <sup>a</sup>
<i>C. Dact.</i> 'Ornamental'	8,33 ± 5,56 <sup>b</sup>	0,33 ± 0,58 <sup>a</sup>	10,66 ± 0,58 <sup>b</sup>	0 ± 0 <sup>a</sup>	0,81 ± 0,39 <sup>b</sup>	0 ± 0 <sup>a</sup>
<i>C. Dact.</i> 'Premium'	3,33 ± 1,15 <sup>b</sup>	0 ± 0 <sup>a</sup>	12,33 ± 0,58 <sup>b</sup>	0 ± 0 <sup>a</sup>	0,27 ± 0,09 <sup>b</sup>	0 ± 0 <sup>a</sup>
<i>F. arund.</i> 'HounDog'	99,33 ± 1,15 <sup>b</sup>	69 ± 6,56 <sup>a</sup>	8 ± 0 <sup>a</sup>	12,66 ± 0,58 <sup>c</sup>	11,34 ± 0,6 <sup>b</sup>	5,75 ± 0,58 <sup>a</sup>
<i>F. arund.</i> 'Nebula'	95 ± 3,46 <sup>b</sup>	59,67 ± 7,55 <sup>a</sup>	8 ± 0 <sup>a</sup>	15 ± 2,65 <sup>c</sup>	11,47 ± 0,32 <sup>b</sup>	4,58 ± 0,71 <sup>a</sup>
<i>F. arund.</i> 'Starlett'	96,33 ± 1,53 <sup>b</sup>	70,67 ± 3,05 <sup>a</sup>	8 ± 0 <sup>a</sup>	13,33 ± 0,58 <sup>c</sup>	11,93 ± 0,44 <sup>b</sup>	5,67 ± 0,23 <sup>a</sup>
<i>F. rubra</i>	94,33 ± 2,52 <sup>b</sup>	84 ± 2,65 <sup>a</sup>	8 ± 0 <sup>a</sup>	10 ± 0 <sup>b</sup>	11,57 ± 1,02 <sup>b</sup>	8,09 ± 0,36 <sup>a</sup>
<i>F. rub. var. Comm.</i>	99,33 ± 1,15 <sup>b</sup>	80 ± 17,69 <sup>a</sup>	8 ± 0 <sup>a</sup>	13,66 ± 0,58 <sup>c</sup>	12,13 ± 0,77 <sup>b</sup>	5,93 ± 1,11 <sup>a</sup>
<i>L. perenne</i> 'Fabian'	90,33 ± 2,08 <sup>a</sup>	86,67 ± 7,64 <sup>a</sup>	8 ± 0 <sup>a</sup>	8 ± 0 <sup>a</sup>	12,09 ± 0,18 <sup>b</sup>	10,17 ± 0,59 <sup>a</sup>
<i>P. pratensis</i> 'Yvette'	74,66 ± 7,37 <sup>a</sup>	65,33 ± 8,62 <sup>a</sup>	10,66 ± 0,58 <sup>a</sup>	15 ± 1 <sup>b</sup>	7,17 ± 0,68 <sup>b</sup>	4,71 ± 0,65 <sup>c</sup>



**Figure 1.** Dynamics of seed germination of *F. rubra* var. *commutata*, *F. rubra*, *Lolium perenne* 'Fabian' and *Poa pratensis* 'Yvette' recorded respectively for the seeds kept at 22°C (above) and at a temperature regime specific to late autumn (below)



**Figure 2.** Dynamics of seed germination of *F. arundinacea* 'HounDog', *F. arundinacea* 'Nebula', *F. arundinacea* 'Starlett' recorded respectively for the seeds kept at 22°C (above) and at a temperature regime specific to late autumn (below). In the second figure, parallel to the dynamics of germination, some elements characterizing the temperature regime are shown

er number of cultivars of this species for their ability to germinate at sub-optimal temperatures.

The dynamics of germination are shown in Figure 1 and 2. The delay in germination at the temperature regime specific to the late autumn is clearly visible on the graphs, which was statistically processed using the DG<sup>50</sup> and GRI indicators. Attempts to detect a statistically significant correlation between germination and various elements in the temperature regime show a low level of credibility in terms of correlation with the temperature sum above +5°C, and the average day, night and 24-hour temperature. In our opinion, temperature maxima, which were not considered in the present experiment, are of primary importance for germination. In the second figure, parallel to the dynamics of germination, some elements characterizing the temperature regime are shown.

## CONCLUSIONS

The obtained results unequivocally show that the late autumn seeding of grass to produce sod is possible, but with the correct selection of the grass, species and varieties involved in the grass mixtures. The perennial ryegrass *L. perenne* 'Fabian' can be mentioned as the most suitable of the tested species and varieties for late seeding. Apart from it, *F. rubra*, *F. rubra* var. *commutata*, and *Poa pratensis* 'Yvette' can also be used. In case of late seeding with grass mixtures in which the described varieties and species participate, it is desirable to increase the seeding rate by an average of 10%. The last ten days of October can be specified as a permissible deadline for carrying out late seeding in the area of the Sofia Plain.

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