Original Paper

The Effect of Biostimulants on the Selected Parameters of Seed Germination of Genus *Festuca*

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In the laboratory experiments with selected grass species of the genus *Festuca* (F.) was evaluated the effect of 3 supporting preparations (plant growth stimulant, preparation with humic substances and extract from brown seaweed) on the germination process. There were evaluated total germination at the end of the experiment, the rate of germination and the mean germination time (MGT) in this experiment. Plant growth stimulant (*F. pallens* +7.21% and *F. rubra* +6.76% compared to control) and extract from brown seaweed (*F. filiformis* +28.09%, *F. pallens* +30.93% and *F. rubra* +17.07% and *F. arundinacea* +1.45% compared to control) showed stimulating effect on total germination from all used preparations. The effect was statistically significant (p = 0.0246). Those preparations can be evaluated positively in terms of their effect on the rate of germination, which at *F. filiformis* increased by approximately 0.34 seeds.day^{-1} to 0.36 seeds.day^{-1}, at *F. rubra* 0.05 seeds.day^{-1} to 0.14 seeds.day^{-1} and at *F. arundinacea* 0.04 seeds.day^{-1} to 0.11 seeds.day^{-1} compared to control. For acceleration of the germination of *F. pallens* (+0.39 seeds.day^{-1}) worked only plant growth stimulant. The values of mean germination time showed a shortened germination period after use of extract from brown seaweed at *F. filiformis* (-0.77 days), *F. pallens* (-0.26 days) and *F. rubra* (-0.41 days), preparation with humic substances at *F. filiformis* (-0.13 days) and plant growth stimulant at *F. pallens* (-0.02 days) and *F. rubra* (-0.14 days).

Keywords: brown seaweed, fescue, germinability, biostimulant, humines

1 Introduction

Grasses are the most widespread group of plants on the planet. Due to its modesty and diversity they are widely used. Some species are used as a component of permanent grasslands, forest communities, agrocenoses, some are grown as fodder on arable land (fodder grasses), others are components of lawns, and tall species grasses of the genus *Arrhenatherum*, *Molinia*, *Phalaris* and others can be used for energy purposes. Ornamental grasses find their application also in roof gardens because they can survive in extremely dry habitats, or vice versa, even in humid places (Nováková, 2004). Recently, we have met more and more often with recurrent periods of drought and heat, which can have very significant, mostly negative, effects on seed germination of grass. According to some authors (Copeland and McDonald, 1995; Pazderů, 2009) seed vigour and germinability, i.e. quality of seed, has an impact on competitiveness of the most species especially in the early stages of development. Knowledge of seeds vitality is desirable when establishing grasslands. It is also a prerequisite for the speed of germination and further development and formation of grassland. Ultimately, it also affects the performance of the required functions, as was confirmed in some experiments (Martinek et al., 2011; Hric et al., 2012). It is especially important in using of mixtures containing species with different speed of germination, emergence and initial growth. Fierce competition occurs from the species with faster development, while the slower species is usually disadvantaged to the extent that completely subsides from the grassland (Svobodová a Šantrůček, 2003). Therefore, some authors (Štranc et al., 2008; Przybysz et al., 2014) recommended using different preparations to support the initial development which stimulate seed germination, whereby the above drawbacks can be eliminated. According to Ali et al. (2021) and Franzoni

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et al. (2022), there are organic or inorganic compounds and/or microorganisms, that when applied to plants, stimulate several processes, leading to improved growth and productivity, and tolerance to stresses and display low or null toxicity and do not accumulate in long term (Sangiorgio et al., 2020). They can be classified based on the source of raw material into six major groups, such as seaweed, plant extracts, protein hydrolysates, humic substances, inorganic compounds, and microorganisms (Franzoni et al., 2021). They are available in liquid, granular form, or soluble powder and can be applied as foliar sprays or/and in soil near the root zone, respectively for the treatment of seeds (Lau et al., 2022). Rai et al. (2021) state that the application of biostimulants represents a significant scientific breakthrough toward sustainably safeguarding future food production.

This paper aims to evaluate the effect of supporting preparations on seed germination ability and germination speed of grains of selected grass species from the genus *Festuca*.

2 Material and methods

2.1 Biological material and treatments

The experiment was realised at the Institute of Agronomic Sciences (workplace Department of Plant Production and Grassland Ecosystems), Faculty of Agrobiology and Food Resources, Slovak University of Agriculture in Nitra.

As biological material were used seeds of the following grasses from the genus *Festuca*:

- *Festuca filiformis*,
- *Festuca pallens*,
- *Festuca rubra*,
- *Festuca arundinacea*.

For this experiment, four treatments were created in which the following preparations were used at concentrations recommended by the manufacturer:

- Treatment 1: Distilled water – in text "control",
- Treatment 2: Preparation 1 (concentration of a solution: 0.003%) – in text "plant growth stimulant",
- Treatment 3: Preparation 2 (concentration of a solution: 0.50%) – in text "preparation with humic substances",
- Treatment 4: Preparation 3 (concentration of a solution: 0.04%) – in text "extract from brown seaweed".

2.2 Characteristics of preparations

Preparation 1 – it is a plant growth stimulant with aromatic nitro compounds as active substances. It supports the rapid synthesis of all vital substances (proteins, fats, sugars, enzymes) for the healthy growth and development of plants. It stimulates the root system.

Preparation 2 – it contains humic substances and their salts (13%), a mixture of oligopeptides and amino acids (10%), an extract of seaweed – adaptogens, a wetting agent having an adhesive effect and other substances that increase the penetration of enhancers and nutrients via membranes. It increases resistance to cold, drought and salinisation, and increases the threshold of tolerance to diseases.

Preparation 3 – the organic extract from brown seaweed containing a high concentration of organic nutrients, minerals, vitamins and bioactive substances. Ingredients: organics (55%), alginic acid (15%), amino acids (6%), a total of N (1%), C (17%), S (1%), Ca (0.5%), Fe (0.15%), Mg (0.04%), copper (25 ppm), natural growth hormones (PGR) (600 ppm) such as auxins, gibberellins, cytokinins and abscisic acid.

2.3 Realization of the experiment and evaluated parameters

The seeds of the evaluated grass species (100 seeds in 4 replicates) were uniformly distributed on the filter paper in a Petri dish of the size 120 × 120 mm. The filter paper was moistened with 10 ml of distilled water or a solution prepared according to the above-mentioned treatments.

Petri dishes with the seeds were placed into a growth chamber (Climacell 404) and incubated at 23 °C (day)/15 °C (night) with a photoperiod 12 hours light/12 hours dark, and relative air humidity $rh = 70\%$.

During the experiment, the filter paper was continuously moistened as needed by prepared solutions according to individual variants. The number of sprouted seeds was detected on the 7th, 14th, 21st and 28th day of the experiment.

Germinability of the seeds at the end of the experiment was expressed as % of control.

The speed of seed germination according to the formula:

$$S = (N_n - N_{n-1})/(t_n - t_{n-1})$$

where: S – the speed of germination (number of seeds. day^{-1}); N – the number of germinated seeds at n^{th} and n - 1st assessment; $t_n - t_{n-1}$ – the number of days between n^{th} and n - 1st assessment

The mean germination time (MGT) according to the formula:

$$\text{MGT} = \sum(D \times n)/\sum n$$

where: *D* – the number of days from the start of the experiment; *n* – the number of seeds germinated at day *D* (Ellis a Roberts, 1980)

2.4 Statistical analysis

The values of total germinability, speed of seeds germination and the mean germination time of grass seeds were evaluated in the program Statistica (Statsoft Inc. (2011). Statistica Cz, version 10) by one-way analysis of variance (ANOVA) followed by testing the evidential differences with Fischer LSD test at 95% level of the probability (α = 0.05). For the graphical processing of results MS Excel program was used.

3 Results and discussion

The total germination of grass seeds, expressed as % of control, is presented in Figure 1. In the case of *Festuca* (*F.*) *filiformis* the germination was increased by 28.09% (treatment 4 – extract from brown seaweed) and 17.20% (treatment 3 – preparation with humic substances), while the plant stimulant (treatment 2) was effected slightly inhibitory (by 1.89%) compared to the control (treatment 1). Humic substances evidenced an inhibitory effect (by 21.69%) in *F. pallens*, while the plant stimulant and extract from brown seaweed increased the germination by 7.21% and 30.93% as compared to the control, respectively. In the case of *F. rubra* grain germination was higher by 6.76% (plant stimulator) and 14.07% (extract from brown seaweed) than the control treatment, whereas the preparation with humic substances was slightly inhibitory (by 3.13%). The germination of *F. arundinacea* seeds was moderately inhibited (treatment 2 – by 2.04%, treatment 3 – by 2.31%). Only the extract from the brown seaweed (treatment 4) slightly stimulated seed germination (by 1.45%) in this grass species. In general, we can conclude a statistically significant influence of the supporting preparations on the germinability (p = 0.0246). The number of germinated seeds increased on average by 2.53% by using the plant stimulant (treatment 2) and the extract from brown seaweed (treatment 4) by an average of 18.64%. Preparation with humic substances (treatment 3) showed a negative effect and by its influence germinability of seeds decreased on average by 2.48% as compared to the control (treatment 1) regardless of grass species. Similarly, Macháč (2011), in its experiment after the application of the growth regulator observed significant differences in the germination values in the case of *Festuca* sp., but also in the case of *Arrhenatherum elatius*. Likewise, Yadav et al. (2024) found enhancing in the germination rate and germination percentage of wheat seed after treatment with herbal extracts.

In the case of the second evaluated characteristic – the speed of seed germination (Figure 2) – differences between individual species are visible. It is a species

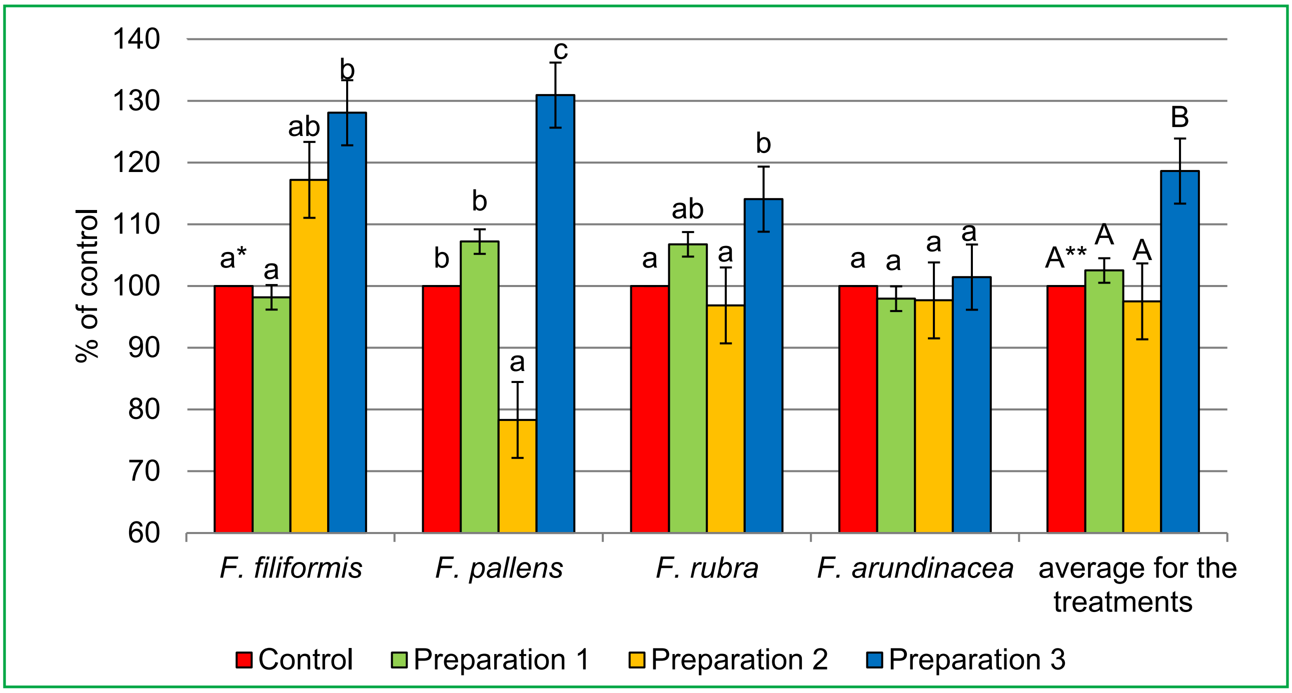

Figure 1 Germinability of seeds of selected grass species (% of control)
* different lowercase letters (a–c) show statistical significance between individual treatments within each grass species (Fischer LSD test, α = 0.05); ** different capital letters (A–B) show statistically significant differences between individual treatments (Fischer LSD test, α = 0.05); error bars represent standard deviation (SD); Control – distilled water; Preparation 1 – plant growth stimulant; Preparation 2 – preparation with humic substances; Preparation 3 – extract from brown seaweed

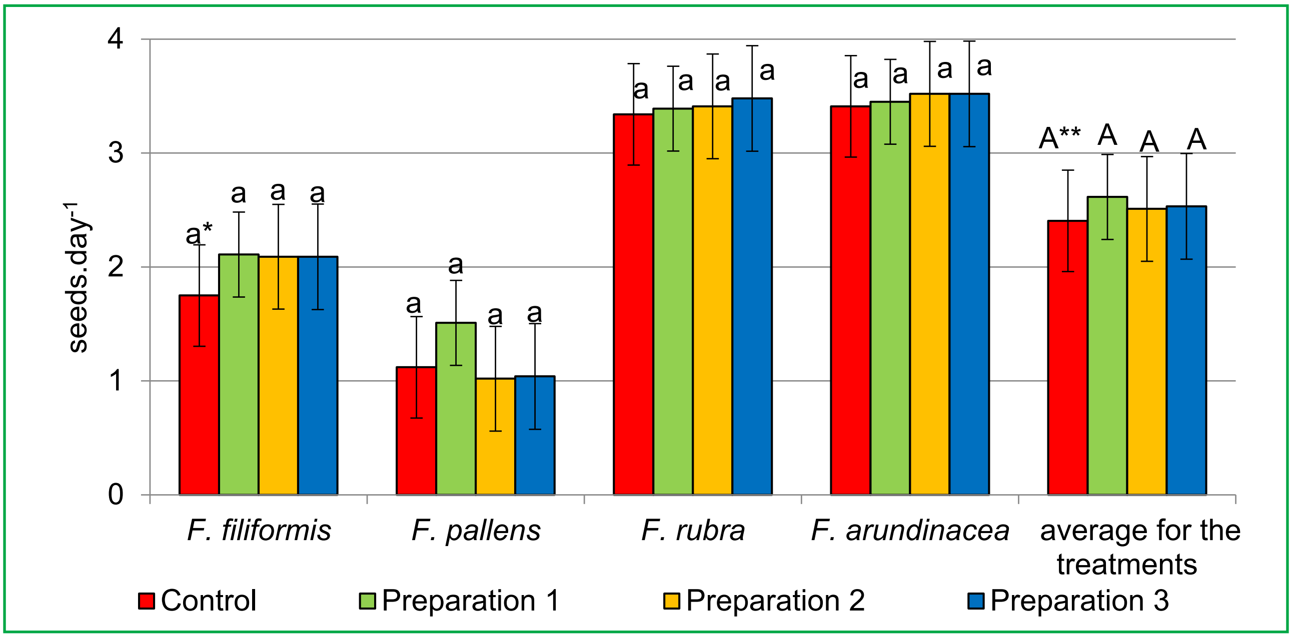

Figure 2 Average seeds germination rate of selected grass species (seeds.day^{-1})
* the same lowercase letter (a) shows statistical insignificance between individual treatments within each grass species (Fischer LSD test, α = 0.05); ** the same capital letter (A) shows statistically insignificant differences between individual treatments (Fischer LSD test, α = 0.05); error bars represent standard deviation (SD); Control – distilled water; Preparation 1 – plant growth stimulant; Preparation 2 – preparation with humic substances; Preparation 3 – extract from brown seaweed

issue, as the speed of germination is one of the natural biological properties of grasses. It is influenced also by the quality of seeds (e.g. age, condition before and after the harvest etc.). However, the comparison of seed germination speed between treatments within individual *Festuca* species showed insignificant (p = 0.9154–0.9999) differences. As in our previous experiments (Kovár et al., 2015; Kovár et al., 2017), there was a different reaction of grass species to the used preparations. The increase in the speed of seed germination due to used preparations was found in *F. filiformis*, where compared to the control, the values of this indicator were 0.34–0.36 seeds.day^{-1}. *F. pallens* responded positively only to the plant stimulant (treatment 2), where the germination rate was 0.39 seeds.day^{-1} higher than in the control treatment. As with *F. filiformis*, the mild stimulating effect of the used preparations was also found for *F. rubra* and *F. arundinacea*, where the germination rate was increased by 0.05–0.09 seeds.day^{-1} and 0.04–0.11 seeds.day^{-1} compared to control (treatment 1), respectively. It can be concluded that between the treatments there were minimal and insignificant (p = 0.9964) differences in the speed of seed germination. The most pronounced acceleration of seed germination as compared to the control (treatment 1 – 2.41 seeds.day^{-1}) was achieved by using a plant stimulant (treatment 2) – on an average of 2.62 seeds.day^{-1}. A less pronounced increase in the germination rate was in treatment 4 (extract from brown seaweed) and treatment 3 (preparation with humic substances) with values of 2.53 seeds.day^{-1} and 2.51 seeds.day^{-1}, respectively. Although in our experiment this increase in the germination rate was less pronounced (by 4.15–8.71% compared to the control), several authors (Rayorath et al., 2008; Ali et al., 2020; Ali et al., 2021) report that these extracts have been shown to positively affect seed germination and other plant growth. In addition, seaweed products can promote an increase in seedling vigor by enhancing root size and density.

Mean germination time (MGT), i.e. the day from the beginning of the experiment, when the level of 50% of the germinated seeds is reached, is documented in Figure 3. From the values of this indicator, we can see certain differences not only between species, but also between treatments. The acceleration of seed germination (i.e. decrease in MGT value) was caused, in particular, by the extract from brown seaweed (treatment 4) at *F. filiformis* (by 0.77 days), *F. pallens* (by 0.26 days) and *F. rubra* (by 0.41 days), a plant stimulant (treatment 2) at *F. pallens* (by 0.02 days), and *F. rubra* (by 0.14 days) and a preparation with humic substances (treatment 3) at *F. filiformis* (0.13 days). In other cases, the supporting preparations had rather the opposite effect, i.e., slowed the germination compared to the control treatment. The general effect of the supporting preparations was characterized by only minimal and insignificant differences (p = 0.8442) between the treatments. The earliest seed germination was achieved after the application of extract from brown seaweed (treatment 4) – on average after 19.25 days.

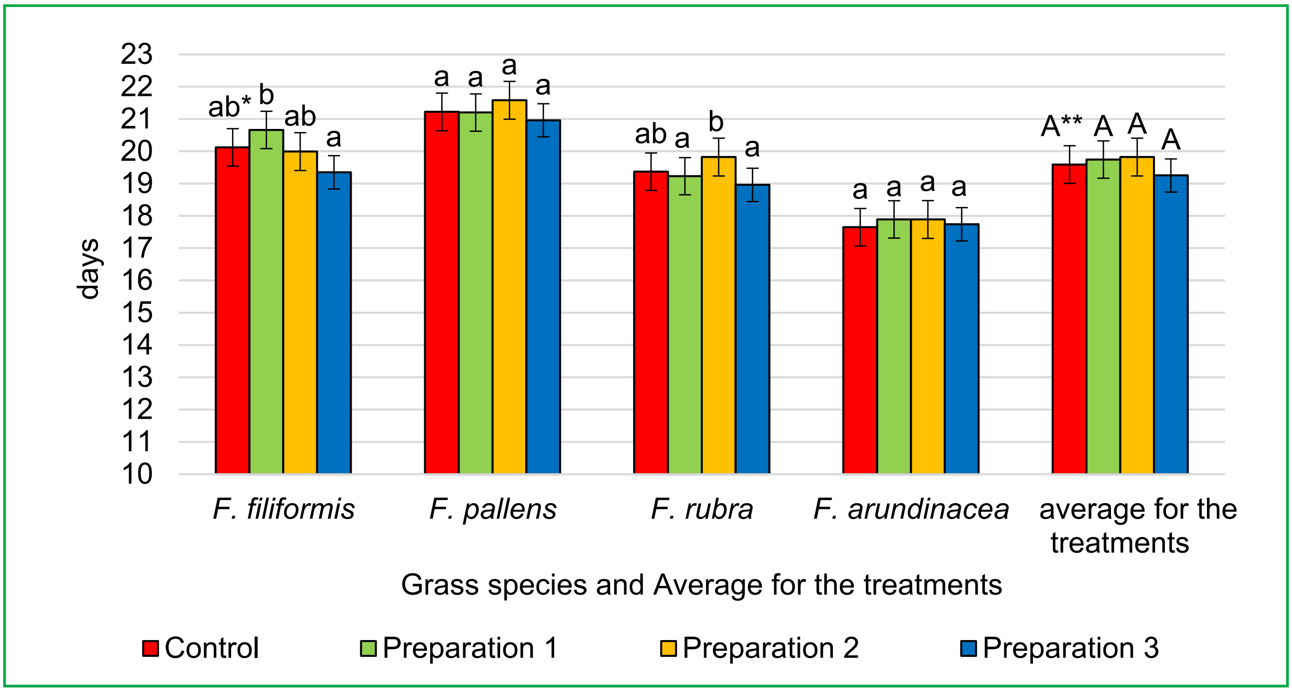

Figure 3 Mean germination time of selected grass species (days)
* different lowercase letters (a–b) show statistical significance between individual treatments within each grass species (Fischer LSD test, α = 0.05); ** the same capital letter (A) shows statistically insignificant differences between individual treatments (Fischer LSD test, α = 0.05); error bars represent standard deviation (SD); Control – distilled water; Preparation 1 – plant growth stimulant; Preparation 2 – preparation with humic substances; Preparation 3 – extract from brown seaweed

The stated finding is thus in agreement with some works (Kovár et al., 2015; Kovár et al., 2017; Badawi et al., 2020; Makhaye et al., 2021; Mzibra et al., 2021), where the stimulating effect of seaweed extracts was shown. It followed treatment 1 (control) with 19.59 days, followed by treatment 2 (plant stimulator) with 19.75 days, and finally 19.82 days for treatment 3 (preparation with humic substances).

4 Conclusions

The results of the experiment aimed at the effect of selected supporting preparations showed that the stimulation effect on the germination process of *Festuca* grass species was achieved using a plant growth stimulant and extract from brown seaweed. Total seed germinability was increased by extract from brown seaweed, especially in *F. filiformis*, *F. pallens* and *F. rubra*. The most obvious effect on germination rate had plant growth stimulant at *F. pallens*. The effect of extract from brown seaweed was manifested by decreasing of the mean germination time at almost all evaluated grass species except for *F. arundinacea*.

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References

Ali, O. et al. (2020). Phytoelicitor activity of *Sargassum vulgare* and *Acanthophora spicifera* extracts and their prospects for use in vegetable crops for sustainable crop production. *J Appl Phyco*, 33, 639–651. https://doi.org/10.1007/s10811-020-02309-8

Ali, O. et al. (2021). Biostimulant properties of seaweed extracts in plants: implications towards sustainable crop production. *Plants*, 10(3), 531. https://doi.org/10.3390/plants10030531

Badawi, M. A. et al. (2020). Improvement of wheat germination by using some biostimulants substances. *J. of Plant Production*, 11(2), 139–144. https://doi.org/10.21608/JPP.2020.79107

Copeland, L. O., & McDonald, M. B. (1995). Principles of Seed Science and Technology. (3rd ed.), Chapmann & Hall.

Ellis, R. H., & Roberts, E. H. (1980). Towards a rational basis for testing seed quality. *Seed Production*, 605–635.

Franzoni, G. et al. (2021). Maceration time affects the efficacy of borage extracts as a potential biostimulant on rocket salad. *Agronomy*, 11(11), 2182. https://doi.org/10.3390/agronomy11112182

Franzoni, G. et al. (2022). Biostimulants on crops: Their impact under abiotic stress conditions. *Horticulturae*, 8(3), 189. https://doi.org/10.3390/horticulturae8030189

Hric, P. et al. (2012). Alelopatické vzťahy trávnikových druhov tráv počas klíčenia [Allelopathic relationships of lawn grass species during germination]. In *VII. vedecká konferencia doktorandov s medzinárodnou účasťou, konaná pri príležitosti Európskeho týždňa vedy*. SUA (pp. 20–23).

Kovár, P. et al. (2017). Klíčenie osiva vybraných druhov rodu *Festuca* po aplikácii podorných prídavkov [Seed germination of selected species of the genus *Festuca* after the application of supporting additives]. In *Osivo a sadba*. Česká zemědělská univerzita (pp. 203–208).

Kovár, P. et al. (2015). Využitie podporných prípravkov pre zlepšenie klíčenia zŕn vybraných druhov kŕmnych tráv [The use of supporting preparations to improve the germination of selected types of fodder grasses]. In *Osivo a sadba*. Česká zemědělská univerzita (pp. 175–180).

Lau, S. E. et al. (2022). Microbiome engineering and plant biostimulants for sustainable crop improvement and mitigation of biotic and abiotic stresses. Discov. *Food,* 2, 9. https://doi.org/10.1007/s44187-022-00009-5

Macháč, R. (2011). Vliv trinexapac-ethylu na kvalitu osiva vybraných druhů trav [Effect of trinexapac-ethyl on seed quality of selected grass species]. In *Osivo a sadba*. Česká zemědělská univerzita (pp. 88–92).

Makhaye, G. et al. (2021). Biopriming with seaweed extract and microbial-based commercial biostimulants influences seed germination of five *Abelmoschus esculentus* genotypes. *Plants*, 10(7),1327. https://doi.org/10.3390/plants10071327

Martinek, J. et al. (2011). Evaluation of *Deschampsia caespitosa* /L./ Beauv. competition ability in mixtures with main turfgrass species. *Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis*, 59(5), 159–168. https://doi.org/10.11118/actaun201159050159

Mzibra, A.et al. (2021). Biostimulants derived from moroccan seaweeds: seed germination metabolomics and growth promotion of tomato plant. *J Plant Growth Regul,* 40, 353–370. https://doi.org/10.1007/s00344-020-10104-5

Nováková, A. (2004). *Okrasné trávy*. Grada Publishing.

Pazderů, K. (2009). Význam energie klíčení pro hodnocení kvality osiva [Significance of germination energy for seed quality assessment]. In *Osivo a sadba*. Česká zemědělská univerzita (pp. 56–60).

Przybysz, A. et al. (2014). Biological mode of action of a nitrophenolates-based biostimulant: a case study. Front. *Plant Sci.,* 713(5). https://doi.org/10.3389/fpls.2014.00713

Rai, N. et al. (2021). Prospects for abiotic stress tolerance in crops utilizing phyto- and bio-stimulants. Front. Sustain. *Food Syst*., 5, 54853. https://doi.org/10.3389/fsufs.2021.754853

Rayorath, P. et al. (2008). Extracts of the brown seaweed *Ascophyllum nodosum* induce gibberellic acid (GA3)-independent amylase activity in barley. *J Plant Growth Regul,* 27, 370–379. https://doi.org/10.1007/s00344-008-9063-6

Sangiorgio, D. et al. (2020). Facing climate change: Application of microbial biostimulants to mitigate stress in horticultural crops. *Agronomy*, 10(6), 794. https://doi.org/10.3390/agronomy10060794

Statistica program/documentation: StatSoft, Inc. (2011). STATISTICA (data analysis software system), version 10. www.statsoft.com

Štranc, P. et al. (2008). Výsledky pokusů s vybranými stimulátory v chmelařství. Moderní trendy v zemědělství [Results of experiments with selected stimulators in hop farming. Modern trends in agriculture]. *Agromanuál*, 4(6), 50 –53.

Svobodová, M., & Šantrůček, J. (2003). Vztah jílku vytrvalého a lipnice lúční při zakládaní trávníků [The relationship between perennial ryegrass and Kentucky bluegrass in the establishment of lawns]. In *Trávníky 2003*. Agentura Bonus (p. 34).

Yadav, A. et al. (2024). Screening herbal extracts as biostimulants to increase germination, plant growth and secondary metabolite production in wheatgrass. *Sci Rep,* 14, 607. https://doi.org/10.1038/s41598-023-50513-6