

PhD DISSERTATION THESIS

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Gödöllő

2025



VEGETATION OF GRAZED DRY PANNONIAN PLAIN GRASSLANDS
AND DOMINANT *FESTUCA* SPECIES

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The background to the work, introduction

Grasslands make up 30% of the world's terrestrial habitats. They represent one of the most extensive and diverse ecosystems in the world. They provide a wide range of ecosystem services, such as water and carbon storage, erosion control, climate change mitigation, and play a crucial role in agriculture.

The present study is based on the examination of grasslands, specifically pastures, where vegetation types dominated by various *Festuca* taxa have emerged. The grazing animals also differed; we examined the vegetation of pastures grazed by Hungarian gray cattle and sheep. The dominant grass in the vegetation of the pastures, in this case the species and taxa of the *Festuca* genus, was examined in more detail, which became a key part of the research.

Festuca (*Loliinae*, *Poaceae*) is one of the most widespread and, in many cases, dominant cosmopolitan genera in the grasslands of the world. For a long time, their taxonomic classification was based solely on morphological characteristics, but due to their high degree of plasticity, these often did not provide completely reliable information about the actual number of species and their exact distribution.

Many species belonging to the *Festuca* genus dominate grassland communities that survive in nutrient-poor or extreme environmental conditions. In this way, fescues often determine the structure of these communities and provide habitat for many protected animal species (e.g., Great bustard (*Otis tarda*), European roller (*Coracias garrulus*), Balkan wall lizard (*Podarcis tauricus*), Sand lizard (*Lacerta agilis*), European ground squirrel (*Spermophilus citellus*)).

Objectives:

The objectives of the research can be divided into two main parts.

I. Objectives and questions concerning pasture vegetation:

I.1. On one hand, the study of typical Pannonian vegetation units dominated by *Festuca* species with filamentous leaves, which have been grazed for a long time, even hundreds of years.

I. 2. Do grazing animals change the species composition of dry Pannonian grassland types?

I.2. What *Festuca* species will be dominant in different vegetation types?

I.3. Does the ratio of vegetation and dominant *Festuca* species change, as well as the dynamics of the species within the year?

I.4. How do different grazing pressures affect the species composition of the grassland? It is particularly important to determine the grazing pressure that is most suitable in terms of vegetation composition and grassland management.

II: Examination of dominant *Festuca* species.

II.1. Comprehensive literature review and critical analysis of characteristic species or dominant *Festuca* taxa in the sample areas, and comparison of the taxa occurring.

II.2. Morphological, ploidy, and molecular genetic examination of the *Festuca* species occurring.

Materials and Methods

Coenology studies

I conducted my coenological studies in Hortobágy and on the Danube-Tisza floodplain. An important goal of the coenological surveys was to monitor changes caused by grazing pressure, so the surveys were conducted at several distances, in strips exposed to different grazing pressures.

The coenological surveys were conducted in spring and autumn based on the Braun-Blanquet method, but with the percentage coverage of species given using 2×2 m quadrats. Six surveys were conducted in each of the three area categories in the studied areas.

I assessed the degradation status of vegetation based on Borhidi's (1993) social behavior types (SBT) and Simon's (2000) nature conservation categories. Of the relative ecological indicator values, we used the following based on Borhidi (1995): relative groundwater and soil moisture (WB), relative nitrogen demand (NB). The life form analysis was performed based on Pignatti's (2005) life form types. When processing the values of the grassland management categories applied to the species, we also used the designations of TASI (2000, 2007, 2018, 2020). Grassland production was estimated according to the Balázs method (BALÁZS 1960) using the following formula.

We used a nonparametric statistical method to analyze the coverage values of species belonging to different groups, as these variables were not normally distributed according to the Shapiro-Wilk test ($p < 0.05$). Accordingly, we used the non-parametric Kruskal-Wallis test ($\alpha = 0.05$) and the non-parametric Dunn test with Bonferroni correction for multiple pairwise comparisons. All statistical procedures were performed using XL-STAT software (Addinsoft, N.Y., USA 2016; UShey et al. 2024). The data from these analyses are currently being

published. Shannon and Simpson diversity indices were used to measure vegetation diversity (Tóthmérész 1995).

The specimens used as the basis for our ploidy studies of *Festuca* species were collected from five locations along the Danube. The sampling areas and species were as follows: *Festuca vaginata* – Homoktövis nature reserve, hereinafter referred to as Homoktövis NR, *Festuca pseudovaginata* – Homoktövis NR, *Festuca tomani* – Homoktövis NR, *Festuca brevipila* – Barcs, *Festuca rupicola* – Györszentiván, *Festuca javorkae* – Csenke (Čenkov), *Festuca wagneri* – Kunbaracs.

Anatomical examinations are supplemented by cytogenetic data. The ploidy level (chromosome number) is determined by flow cytometry (Qiu et al., 2019). Different ploidy levels (diploid, tetraploid, hexaploid, etc.) often correlate with morphological characteristics, ecological niche, and reproductive isolation (Šmarda, 2008; Šmarda et al., 2008). To examine the ploidy level, we cut 100 mg of young, green, viable leaves per sample from each taxon intended for testing. We worked with a total of 101 individuals. Flow cytometry measurements were performed at the Szeged Cereal Research Institute using the CytoFLEX Flow Cytometer (Beckman Coulter Inc., Brea, California, USA) based on the method of Lantos et al. (2012) using a CytoFLEX Flow Cytometer (Beckman Coulter Inc., Brea, California, USA). As a reference, we used the ploidy test graph of proven diploid *Festuca vaginata* individuals collected on Homoktövis NR.

Results

The dry alkaline grassland of Hortobágy (*Artemisio santonici-Festucetum pseudovinaere*) are suitable for grazing and, from a nature conservation perspective, tolerate grazing well in terms of biomass conversion and species diversity. I have confirmed that it is reasonable to examine both aspects on saline pastures: autumn conditions that develop after spring conditions can also be evaluated. Wet alkaline grassland type with foxtail (*Alopecuretum pratensis*), on the other hand, are sensitive to grazing, resulting in significant deterioration and species impoverishment, but are suitable for mowing. The species composition of the wet alkaline grassland and the dry alkaline grassland also degrades along the spatial gradient of grazing pressure, away from the corral and livestock farm. In both cases, the vegetation closest to the enclosure is the most degraded. I have shown that vegetation exposed to lower grazing pressure develops in completely different ways in saline and sandy vegetation.

Different dominant *Festuca* species appear in the pasture area. *Festuca pseudovina* was the dominant species in the saline area. In addition to *Festuca vaginata*, *F. wagneri*, *F.*

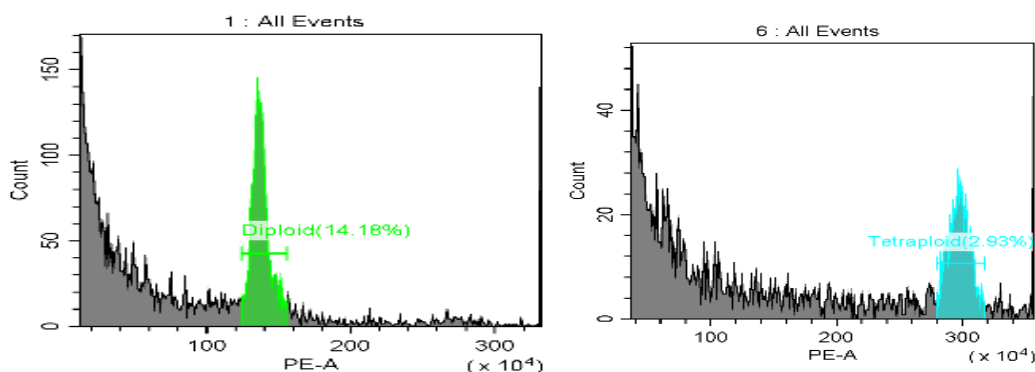
pseudovaginata, *F. tomanii*, and *F. javorkae* also occur in sandy pastures. The occurrence of these species was first reported in these sandy areas. I was the first to analyze the coenological records of the vegetation types formed by these species. *Duna-Tisza* (Kunpeszér

In the pastures examined in the area around Kunpeszér, the number of species increased steadily with distance from the barn. In the most heavily used strip, grazing pressure was highest at 0-50 m, exceeding 1 animal/ha. This also involves significant trampling. Nevertheless, weeds do not have good environmental conditions to develop, and usually only 1-1 weed species can dominate. The analyzed values for total species number and average species number showed a linear increase with decreasing grazing pressure.

Results of ploidy tests

Based on preliminary morphological and histological examination, we potentially identified the taxa and specified their ploidy level based on literature data. After the examination, we corrected the identifications based on the ploidy level and specified the names of the species. The device we used was able to measure the G₀/G₁ phase of the cell cycle. This is the growth phase of the cell cycle.

Based on the results of ploidy tests, alapján az ábrán the figure shows the ploidy level of *Festuca pseudovaginata* from the Homoktövis Nature Reserve, which is a tetraploid species. The tetraploid nature of the species can be seen from the fact that the peak measured on the x-axis during the G₁ growth phase (cell proliferation) is twice as far as the diploid *Festuca vaginata* reference.



Comparison of the ploidy of diploid Festuca vaginata (Homoktövis NR) and tetraploid Festuca pseudovagina (Homoktövis NR)

During the ploidy tests, the ploidy analysis of 101 samples collected from four local and one foreign location yielded several new and interesting results. In the present study, the domestic specimens potentially identified as *Festuca rupicola*, which were collected near Györszentiván, were hexaploids, confirming the aforementioned assumption. However, it refutes the possibility that the species belongs to the previously assumed *Festuca javorkae* species (Soó 1964). It follows that further studies are needed to prove the domestic occurrence of *Festuca javorkae*. The classification of the *Festuca* specimens with bristly outer glumes and strongly silvery, rough leaves collected from the Homoktövis Nature Reserve was also questionable. They could have belonged to the hexaploid *Festuca tracyhyphylla* taxon, but these are tetraploids, so the above assumption can be ruled out. During our investigations, we found the northerly distributed species *Festuca brevipilla* in planted grasslands. In natural vegetation, it occurs in the acidic sandy area near Barcs. The ploidy test seems to confirm our assumption, as previous literature indicates that the species is hexaploid.

Conclusions

Overall, the coenology study area was suitable for analyzing changes in the vegetation of dry alkaline grasslands and wet alkaline grasslands in parallel. This allowed us to confirm not only that animal husbandry is a human activity that significantly affects grassland ecosystems, plant biomass, and biodiversity, but also that significant differences arise even under similar grazing pressures. During the study, we identified parallels, similar trends, and differences. This study confirmed what has been reported in numerous studies on grazing, namely that grazing can alter resource competition between plants and soil properties through various mechanisms, thereby also altering plant biomass and the diversity of grassland ecosystems. The food choices of animals can directly alter the composition of plant communities by selecting certain species and removing dried biomass, which can reduce competition for light among plant species and thus alter plant biomass and area diversity over time.

In terms of biodiversity, grazing can both harm and help the grassland ecosystems. While excessive grazing pressure can lead to a significant decline in native plant species, balanced grazing has been shown to promote conditions favorable to certain grassland birds and other wildlife (Kotsonas et al., 2021). The interactions between grazing and other ecological processes are complex; for example, while moderate grazing can increase plant species

diversity, the absence of grazing can lead to the encroachment of woody plants, further degrading grassland habitats.

In sandy areas, however, there was a direct correlation between vegetation degradation and grazing pressure. Sandy grasslands do not tolerate grazing pressure to the same extent as alkaline grasslands. Comparing the richness of vegetation developed under grazing pressure in sandy areas with that in the wormwood grassland, which was the most species-rich, the average number of species per quadrat was only 14, while in the sandy area it was over 20 in zone "A" and in the most remote areas in zone "C," which was exposed to the least grazing pressure, the number of species was over 40, and even exceeded 50. In addition, the dominant species of *Festuca* taxa were more diverse than in the saline area, where only *Festuca pseudovina* is found.

Although *Festuca wagneri* and *tomanii* differ morphologically, their ploidy levels are identical: both species are diploid, so they cannot be distinguished on the basis of ploidy. More detailed genetic determination will therefore be necessary in the future: for example, distinguishing them on the basis of DNA synthesis should be investigated.

Summary of new scientific findings

- The dry alkaline grassland with wormwood (*Artemisio santonici-Festucetum pseudovinae*) is suitable for grazing and, from a nature conservation perspective, tolerates grazing well in terms of biomass conversion and species diversity. I have confirmed that it is reasonable to examine both aspects on saline pasture: autumn conditions following spring conditions can also be evaluated. The wet alkaline grassland type with foxtail vegetation (*Alopecuretum pratensis*), on the other hand, are sensitive to grazing, resulting in significant deterioration and species impoverishment, but are suitable for mowing.
- The species composition of the alkaline steppe with wormwood and the dry sandy grassland is degraded along the spatial gradient of grazing pressure, away from the corral and livestock farm. In both cases, the vegetation closest to the pens is the most degraded. I have shown that vegetation exposed to lower grazing pressure develops in completely different ways in salt marsh and sandy vegetation.
- Different dominant *Festuca* species appear in pasture areas. *Festuca pseudovina* was the dominant species in the saline area. In sandy pastures, *F. wagneri*, *F. pseudovaginta*,

F. tomanii, and *F. javorkae* occur alongside *F. vaginata*. The occurrence of these species was first reported in these sandy areas. I was the first to analyze the coenological records of the vegetation types formed by these species

- We examined the ploidy of all *Festuca* species along the Danube. We clarified the ploidy status of *Festuca vaginata* and *F. pseudovaginata*. We determined the ploidy level of all collected samples. We separated the *Festuca javorkae* taxon and clarified the ploidy level of *Festuca vaginata* and *Festuca pseudovaginata*.

Publications related to the topic of the dissertation

Journal articles:

in foreign languages

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