

HIERARCHICAL CLUSTER ANALYSIS OF CULTIVAR × SOWING DENSITY COMBINATIONS FOR WHEAT YIELD AND GRAIN QUALITY ON CHERNOZEM SOIL

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Abstract. The identification of cultivar-specific responses to sowing density is important for simultaneously optimizing wheat productivity and grain quality. This study applied hierarchical cluster analysis to classify nine winter wheat (*Triticum aestivum* L.) cultivar × sowing density combinations on the basis of two-year mean values obtained on chernozem soil. Three cultivars, Glosa, Anapurna and Biharia, were evaluated at 780, 650 and 530 viable grains m^{-2} . Separate dendrograms were constructed for grain yield, protein content, wet gluten content, Zeleny sedimentation value, thousand-kernel weight (TKW) and sowing density, using single linkage and Euclidean distances. The cluster structures demonstrated that the similarity among treatments was trait-dependent. For grain yield, Biharia at 780 and 650 viable grains m^{-2} formed a close pair, whereas the remaining treatments showed a different aggregation pattern and Biharia at 530 viable grains m^{-2} was positioned distinctly in the hierarchy. For protein, the three Biharia treatments formed a coherent group that was clearly separated from the Glosa and Anapurna combinations. Wet gluten produced three principal cultivar-related branches, with the closest similarities occurring within specific density combinations. Zeleny sedimentation and TKW also displayed cultivar- and density-dependent clustering, confirming that technological quality cannot be inferred from yield behavior alone. The dendrogram based on density produced three expected groups corresponding to 780, 650 and 530 viable grains m^{-2} , independently of cultivar, validating the treatment coding and emphasizing the contrast among plant populations. Overall, hierarchical clustering revealed both cultivar identity and sowing density as important sources of phenotypic differentiation. The results support the use of cluster analysis as a complementary multivariate tool for identifying treatment similarities and for developing cultivar-specific sowing-density recommendations when yield and grain quality are considered together.

Keywords: winter wheat; hierarchical cluster analysis; sowing density; grain yield; protein; wet gluten; Zeleny; thousand-kernel weight

INTRODUCTION

Winter wheat (*Triticum aestivum* L.) production requires technological decisions that maintain high yield while preserving grain quality. Genotype, crop management and environmental conditions jointly regulate stand establishment, tillering, spike density, kernel development and the accumulation of storage proteins. Sowing density is particularly important because it modifies competition among plants and may alter both yield components and technological quality (Schmitz and Ransom, 2021; Lachutta and Jankowski, 2024).

The response to plant population is strongly genotype-dependent. A high density may increase the number of spikes per unit area but can reduce kernel weight or modify grain protein and gluten traits, while moderate densities may allow cultivars with strong tillering capacity to compensate through individual-plant productivity. Recent studies have therefore emphasized that optimum density should be defined for specific cultivars and production environments rather than treated as a fixed technological constant (Zhang et al., 2025; Lachutta and Jankowski, 2024).

Grain quality is itself multidimensional. Protein content and wet gluten are closely related to baking performance, whereas the Zeleny sedimentation value reflects characteristics of the protein complex and TKW describes an important physical component of grain formation. Research on diverse wheat germplasm has shown strong relationships among protein, gluten and sedimentation traits, but also substantial genotypic variation in their joint expression (Singh et al., 2022; Paunescu et al., 2023).

When several correlated traits and treatment combinations are evaluated, multivariate approaches provide information that cannot be obtained from isolated mean comparisons. Hierarchical cluster analysis groups observations according to their similarity and has been successfully used to classify wheat genotypes, grain-quality profiles and management responses (Salamon et al., 2024; Zhang et al., 2026). In the present experiment, clustering was performed separately for yield and each quality indicator, allowing the stability or reorganization of cultivar $\times$ density similarities to be visualized across traits.

The objective of this study was to characterize the hierarchical similarity of nine combinations obtained from three winter wheat cultivars (Glosa, Anapurna and Biharia) and three sowing densities (780, 650 and 530 viable grains m^{-2}), using two-year mean data from chernozem soil. The analysis focused on grain yield, protein, wet gluten, Zeleny sedimentation value, TKW and density itself, with the aim of identifying cultivar-specific and density-specific grouping patterns relevant to technological optimization.

MATERIAL AND METHODS

The study included three winter wheat cultivars: Glosa (s1), Anapurna (s2) and Biharia (s3). Each cultivar was evaluated at three sowing densities: D1 = 780, D2 = 650 and D3 = 530 viable grains m^{-2} . The nine combinations were coded C1_s1d1 through C9_s3d3. Thus, C1–C3 represented Glosa at the three densities, C4–C6 represented Anapurna, and C7–C9 represented Biharia. The present analysis used mean values across two experimental years for soil type II, classified in the source dataset as chernozem.

The traits subjected to hierarchical cluster analysis were grain yield, grain protein content, wet gluten content, Zeleny sedimentation value and thousand-kernel weight (TKW; M.M.B. in the original statistical output). Sowing density was also clustered as a control variable for treatment structure. The source statistical output did not contain replicate-level raw observations; consequently, this paper interprets the topology and relative linkage structure of the supplied dendrograms rather than recalculating inferential statistics.

Hierarchical agglomerative clustering was carried out separately for each trait using the single-linkage method and Euclidean distance, as specified in the statistical output. In single linkage, the distance between two clusters is defined by the smallest distance between observations belonging to the two clusters. Lower linkage distance therefore indicates greater similarity between treatment combinations for the trait under consideration. Dendrograms were interpreted by examining the earliest pairings, the formation of larger branches and the relative distance at which major groups were merged.

Table 1

Code	Experimental combinations used in the cluster analysis			Viable grains
	Cultivar	Density	m^{-2}	
C1	Glosa	D1		780
C2	Glosa	D2		650
C3	Glosa	D3		530
C4	Anapurna	D1		780
C5	Anapurna	D2		650
		500		

C6	Anapurna	D3	530
C7	Biharia	D1	780
C8	Biharia	D2	650
C9	Biharia	D3	530

RESULTS AND DISCUSSION

The dendrogram for grain yield showed a strong separation among treatment combinations and a particularly close association between C7 and C8, corresponding to Biharia at 780 and 650 viable grains m⁻². These two treatments joined at a comparatively low linkage distance, indicating similar two-year mean yield behavior. The Glosa and Anapurna treatments formed several closer subgroups within the opposite major branch. C9 (Biharia × 530 viable grains m⁻²) occupied a more distinct position in the hierarchy. This pattern suggests that reducing density to 530 viable grains m⁻² changed the yield expression of Biharia more strongly than the difference between 780 and 650 viable grains m⁻². Density-dependent compensation through tillering and kernel formation is well documented in wheat and may differ considerably among cultivars (Schmitz and Ransom, 2021; Zhang et al., 2025).

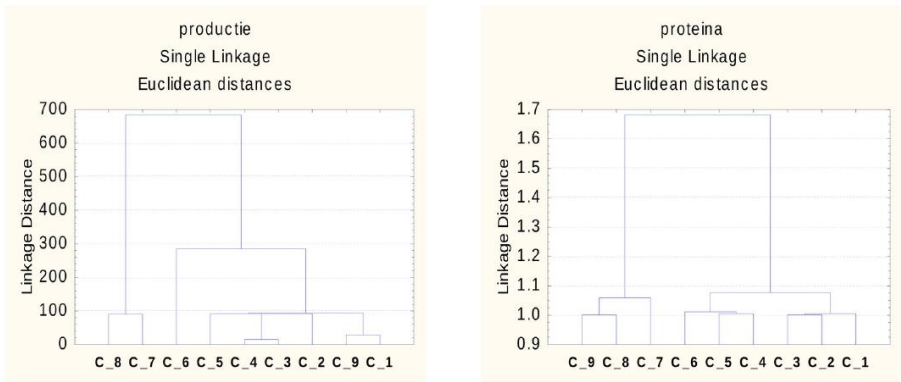


Figure 1. Hierarchical clustering of the nine cultivar × density combinations for grain yield (left) and protein content (right). Single linkage, Euclidean distances.

Protein content produced a more cultivar-oriented structure. C7, C8 and C9, representing the three Biharia densities, formed a coherent branch that was separated at a relatively high linkage distance from the remaining treatments. Within the Glosa and Anapurna groups, several density combinations joined at short distances. This indicates that, for protein accumulation, cultivar identity exerted a strong organizing effect in the similarity structure. Such behavior is consistent with studies showing that grain protein is strongly genotype-dependent but remains responsive to stand density, nitrogen supply and environmental conditions (Landolfi et al., 2021; Paunescu et al., 2023; Lachutta and Jankowski, 2024).

The wet-gluten dendrogram displayed three major zones of similarity. The Biharia combinations C7–C9 were grouped on one side, with C7 and C8 showing close similarity before joining C9. Anapurna treatments C4–C6 formed another compact region, while Glosa combinations C1–C3 occupied a third branch. The clustering was not perfectly identical to the protein dendrogram, demonstrating that protein concentration and wet-gluten content, although related, do not provide redundant information. Similar multivariate differentiation has been

reported in wheat collections in which protein, wet gluten and Zeleny values were positively associated but still contributed differently to genotype classification (Singh et al., 2022).

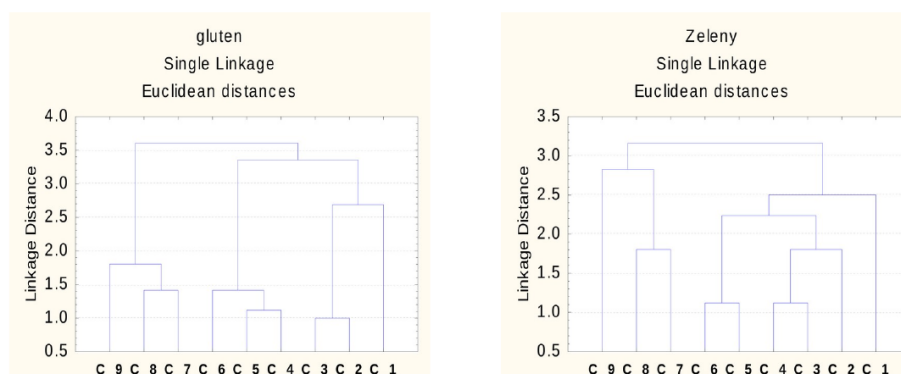


Figure 2. Hierarchical clustering for wet gluten content (left) and Zeleny sedimentation value (right). Single linkage, Euclidean distances.

The Zeleny sedimentation dendrogram revealed a more complex reorganization of treatments. C5 and C6 formed a close pair, as did C3 and C4, whereas other combinations were joined at progressively larger distances. The fact that some clusters crossed cultivar boundaries indicates that sedimentation behavior was influenced by the cultivar $\times$ density combination rather than cultivar identity alone. The Zeleny test integrates aspects of protein quantity and gluten strength; therefore, treatments with similar protein concentrations may still differ in sedimentation behavior. Recent quality studies have likewise shown that sowing strategy and plant density can modify protein, wet gluten and sedimentation indices to different extents (Lachutta and Jankowski, 2024).

For TKW, the dendrogram again revealed a combination of cultivar and density effects. C7 and C8 were relatively similar, while C4 and C5 formed another close subgroup. C1–C3 also showed short-distance associations before joining the larger structure. TKW is determined by genotype and post-anthesis assimilate supply and can respond to stand density because increasing competition modifies grain number and individual-kernel filling. Studies of wheat seeding rate commonly report density effects on yield components and kernel mass, although the magnitude depends on cultivar and environmental productivity (Schmitz and Ransom, 2021; Zhang et al., 2025).

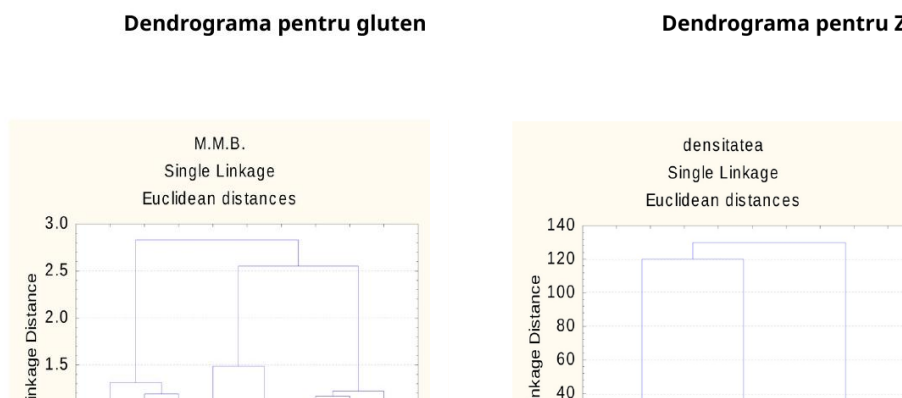


Figure 3. Hierarchical clustering for thousand-kernel weight (TKW; left) and sowing density (right). Single linkage, Euclidean distances.

The density dendrogram provides an internal check on the treatment structure. The nine cases separated into the three expected density classes, each containing one treatment from Glosa, Anapurna and Biharia. This confirms that the coding used in the cluster output correctly represents the three plant populations and highlights the large numerical separation among 780, 650 and 530 viable grains m^{-2} . In contrast, the yield and quality dendrograms did not simply reproduce these density classes, demonstrating that the biological responses resulted from interactions between cultivar characteristics and plant population.

Across traits, the most important result is the instability of treatment similarity from one variable to another. A pair of treatments that clustered closely for yield was not necessarily equally similar for Zeleny or TKW. This supports a multivariate interpretation of wheat technology. Cluster analysis is useful because it identifies phenotypically similar treatment groups without imposing a priori categories. Recent multivariate wheat studies have used hierarchical clustering, PCA and related approaches to separate genotypes according to combined yield, quality and adaptation profiles (Salamon et al., 2024; Zhang et al., 2026).

The results also complement the PCA-based evaluation of the same cultivar $\times$ density system. PCA summarizes covariance among several variables simultaneously, whereas the present trait-by-trait cluster analysis shows which treatment combinations are similar for each individual agronomic or quality indicator. Used together, these approaches can distinguish general multivariate performance from trait-specific similarity. For practical recommendations, both should ultimately be integrated with ANOVA of the complete multi-year dataset, particularly because year, soil and genotype $\times$ environment effects can substantially modify grain yield and processing quality (Mitura et al., 2023; Paunescu et al., 2023).

CONCLUSIONS

Hierarchical cluster analysis clearly differentiated the nine Glosa, Anapurna and Biharia $\times$ sowing density combinations, but the grouping pattern depended on the trait analyzed.

For grain yield, Biharia at 780 and 650 viable grains m^{-2} showed high similarity, whereas Biharia at 530 viable grains m^{-2} was more distinct. Protein and wet-gluten clustering showed a stronger cultivar-related structure, particularly for Biharia, while Zeleny sedimentation and TKW produced more complex cultivar $\times$ density groupings.

The density dendrogram recovered the three experimental plant-population classes independently of cultivar, while the biological traits did not reproduce these classes exactly. This confirms that yield and grain quality responses cannot be explained by sowing density alone.

The cluster approach is therefore useful as a complementary tool for identifying treatment similarity and for supporting cultivar-specific density recommendations. The final technological recommendation should integrate cluster analysis with PCA and inferential analysis of the complete multi-year, multi-soil dataset.

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