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Comparative Analysis of Agronomic Traits, Seed Color Variability and Protein Content in *Pisum sativum* L. Cultivars Across Diverse European Environments

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ABSTRACT

Background: Pea (*Pisum sativum* L.) provides one of the best solutions for the lack of plant-based protein. The different agroecological conditions and the difference in seed color and type can affect seed protein content, composition and agronomic traits.

Methods: A two-year trial on two European sites was done using an augmented block design. Agronomic traits and seed protein content were determined for each plot. Pearson's correlation coefficients and multivariate analysis were done to analyze the structure and pea traits. Electrophoresis was done to investigate variations in protein composition.

Result: The results of multivariate analysis showed the separation of pigmented seeds from non-pigmented seeds, with no clear grouping concerning seed type. The protein composition differed between seeds of different colors. Environmental factors had a significant impact on the duration of flowering, the number of pods and seeds and seed weight per plant.

Key words: Agronomic traits, *Pisum sativum* L., Protein content, Seed.

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INTRODUCTION

Pea (*Pisum sativum* L.) is a legume rich in digestible protein and holds a prominent position as a crucial nutritional source across the globe. Depending on genotype and growth conditions, pea seeds can contain up to 33% protein (Dahl *et al*., 2012; Lam *et al*., 2018; Daba and Morris, 2021). Also, pea contains low levels of anti-nutritional compounds, providing a rich supply of amino acids and fiber (Clemente *et al*., 2015), with a unique ability for biological nitrogen fixation and mobilization of insoluble soil nutrients, bringing about qualitative changes in soil properties (Choudhary *et al*., 2023).

In the last fifty years, Europe has seen a significant rise in protein demand due to population growth. This has led to a decrease in animal husbandry, prompting the adoption of legumes as substitutes for animal proteins both in Europe and globally (Maurya *et al*., 2015). While pea cultivation has increased recently (Faostat, 2021), the demand for new protein sources continues to grow (Ismail *et al*., 2020). This increase is driven by high protein consumption in Europe, particularly plant-based protein, which exceeds recommended levels by an average of 70% (Westhoek *et al*., 2011). Peas, unlike soybeans, can be grown across a wider geographical range, including colder climates. Importantly, pea grain can be used directly in animal feed without prior heat treatment, which streamlines usage and reduces processing costs (Anderson *et al*., 2007).

Pea breeding aims to boost seed yield and protein content for wider applicability in diverse agricultural systems. To ensure stable and adaptable yields in peas, it's important to evaluate how different pea varieties interact with their environment (G×E) across various agricultural conditions (Nassir and Ariyo, 2011; Haile 2020; Isik *et al*., 2017). Pea genotypes typically have two seed forms: smooth and wrinkled. While protein content and composition may differ between these forms, specific information remains inconclusive. Pea seeds vary in color, including brown, light and dark green, cream, or yellow, with pigmented, non-pigmented, or mixed varieties, depending on the genotype (Stjepanović *et al*., 2012). The protein content and composition may not be directly influenced by seed coat color, but there could be some differences related to morphological and physiological properties, such as seed surface or color (Guindon *et al*., 2021).

To address the growing demand for plant-based protein sources and recognize the paramount role of peas, this study investigates the protein content of pea cultivars with varying seed colors. The goal is to determine how different agroecological conditions impact pea seed protein content and agronomic traits related to seed yield.

MATERIALS AND METHODS

Field trials were established in 2019 and 2020 at two distinct locations, the experimental field of the Institute of Field and Vegetable Crops, Rimski šančevi, Republic of Serbia (45°20′N, 19°51′E) and the experimental field of Agro Seed Research, Kessenich, Belgium (51°08′N, 5°48′E). The analyzed panel included research lines and varieties of dry and forage pea from Southern Europe. All material was kindly provided from germplasm collection from the Institute of Field and Vegetable Crops. The experiments were set up on 64 pea (*Pisum sativum* L.) genotypes, using the augmented design methodology within a row-column system (Cullis *et al*., 2006; Kumar *et al*., 2024) with four experimental blocks. Trials were implemented using the same experimental plan, with the difference of vegetative period from March until July for trial in Serbia and from April until August for trial in Belgium. The data for analyzed traits was measured and collected from ten randomly selected plants from each plot, avoiding marginal rows, or from a seed sample from the whole plot.

Weather conditions

Meteorological data from the Serbian trial plot (RHMZ, Annual Bulletin for Serbia in 2019 and 2020) revealed climate variations for both years. In 2019, the pre-sowing period had higher temperatures and less precipitation, balanced by above-average rainfall in April. May saw decreased temperatures and increased precipitation during pea bloom, while June experienced above-average precipitation during pod development, followed by an unusually dry July. In 2020, both temperature and precipitation exceeded averages during the pre-sowing period and March. April and May had below-average precipitation, while June saw significantly higher levels. The average temperatures from January to December 2020 were 0.6°C cooler than in 2019, with 61.1 mm precipitation, 8.4 mm more than in 2019, both above the long-term average (Fig 1, a). Meteorological data from the Belgian trial plot (Data Access Viewer - Agroclimatology) showed that, in 2019, March's pre-sowing period had above-average temperature and precipitation, with below-average precipitation in April. June and July witnessed below-average temperatures during flowering and pod filling, respectively. Precipitation during the pea vegetative season (April to August) was below average, except in March and June. From May to August 2020, temperatures were below average, except for April, with above-average temperatures in the pre-sowing period (January and February). Precipitation variations occurred, with below-average levels in April and May. The average temperatures from January to December 2020 were 0.5°C warmer than 2019, with 47 mm precipitation, 2.2 mm less than 2019, both below the multi-year average (Fig 1,b).

Seed yield components

Analyzed agronomic traits were determined for each plot. Time to flowering (FT) and plot seed yield (PSY) were determined for the whole plots; pods per plant (PP), pod length (PL), seed weight per plant (SWPP) and seeds per pod (SPP) were determined on a random sample of 10 plants from each plot; a thousand seed weight (TSW) was determined regarding the method of seed weight determination (ISTA, 2015) and seed protein content (PC) was determined by Fourier transform near-infrared spectroscopy (FT-NIRS) on an Antaris™ II FT-NIR Analyzer.

Protein composition

The protein composition was determined by using SDS-polyacrylamide gel electrophoresis by Laemmli (1970). The results obtained by electrophoretic analyses were then quantified by gel scanning and densitometric analysis using BioVision (Wilber, Germany) software. The identification of 18 individual subunits was conducted based on molecular weight and information from previous studies by Barać *et al*. (2010) and Barać *et al*. (2011). The representation of each genotype's subunits in the sum amounts to 100%.

Seed color and seed type

The determination was done by observing seed color within one month after harvest and seed type was observed immediately after harvest. Categorization of the seeds was done based on their color, resulting in the following groups: yellow (non-pigmented), green (pigmented and non-pigmented), mixed green (with varying shades of yellow), mixed non-pigmented (consisting of different shades of non-pigmented), mixed pigmented (consisting of different shades of pigmented) and mixed (a combination of pigmented and non-pigmented). Furthermore, the seeds were sorted into different types, namely smooth, wrinkled, a mixture of dimpled and smooth and a mixture of wrinkled and smooth (UPOV *Pisum sativum* TP TG/7/10 Rev. Pea).

Statistical analysis

Results from a two-year study were used to model spatial variation using a first-order autoregressive structure, while values for measured traits were predicted based on the spatial variation, using Best Linear Unbiased Predictions (BLUP). A mixed model analysis, following Demidenko *et al*. (2005), was conducted in Progeno software (Maenhout, 2018). Significant differences between average values across the two years were determined using a T-test. Descriptive statistics for both trials were computed using XLStat. Principal component analysis (PCA) was performed on traits from both sites, using mean values across both years, to examine the population structure of 64 pea genotypes, considering seed color and type, using Minitab (Minitab 17 trial version). Pearson's correlation coefficients between BLUPs for quantitative traits were analyzed in R program version 4.2.2 (R Package "corrplot", Wei and Simko, 2017), following Benesty *et al.* (2009). For non-parametric statistics comparing multiple groups of pea seeds based on color and type, a Kruskal-Wallis test was performed, according to Corder *et al.* (2009), with ggplot2 package in R program version 4.2.2 used for visual exploration of differences and relationships between seed groups (Wickham, 2016).

RESULTS AND DISCUSION

Agroecological impact on pea traits

In order to evaluate the impact of agro-ecological conditions on pea traits, the significance of the differences through the T-test was evaluated between the average values of two years for FT (days), PP, PL (cm), SPP, SWPP (g), TSW (g), SY (kg/ha) and PC (%). Based on the descriptive statistics shown in Table 1, it can be seen that FT had mean values of 22.3 and 14.9 days, mean PP values were 11.5 and 8.1, PL had similar values for both trials (5.8 cm for Serbian and 5.1 cm for Belgian trial); SPP was 8.7 and 6.6 in average, while SWPP was 7.2 g and 6.2 g for Serbia and Belgium, respectively. These traits showed significant differences between two different agroecological environments. Traits TSW, with average values of 182.6 g and 185.3 g and SY with averages of 104.9 kg/ha and 106.4 kg/ha showed no statistical differences between the two trial sites. Mean values for PC at the Serbian site exhibited an average protein content of 27.3%, while at the Belgian site, it was 27.4%, showing no impact of the environmental conditions on this trait. The selection of two geographically distinct locations aimed to enhance the study's scope by considering diverse agroecological conditions. Findings suggest that imbalances in rainfall during the flowering phase can significantly affect interactions between plants and pollinators, thereby regulating flowering time (Kuppler and Kotowska, 2021). Insufficient rainfall, leading to heat stress as observed in the Belgian trial, results in pod rejection and reduces the number of pods per plant, in line with findings by Atung (2018) and Mohapatra *et al*. (2020). Additionally, unfavorable agroecological conditions reduce flowering time, contributing to decreased seed mass (Tawaha and Turk, 2004). This study concludes that considerable variations in agroecological conditions between the two investigated localities, particularly in rainfall patterns, significantly impact these characteristics. Aside from the differences in precipitation quantity and distribution, these features may also be influenced by the timing of sowing, according to Bozoglu *et al*. (2007). The research conducted by Barcchiya *et al*. (2018) and Saxesena *et al*. (2014) suggests that SPP is mainly influenced by environmental factors rather than genotype, although it does exhibit significant heritability in a broader sense, while PL is a heritable trait, meaning that genetic factors, more than agroecological conditions, have a major influence on this trait (Avci and Ceyhan, 2006), which can be seen from similar results on two localities. By comparing

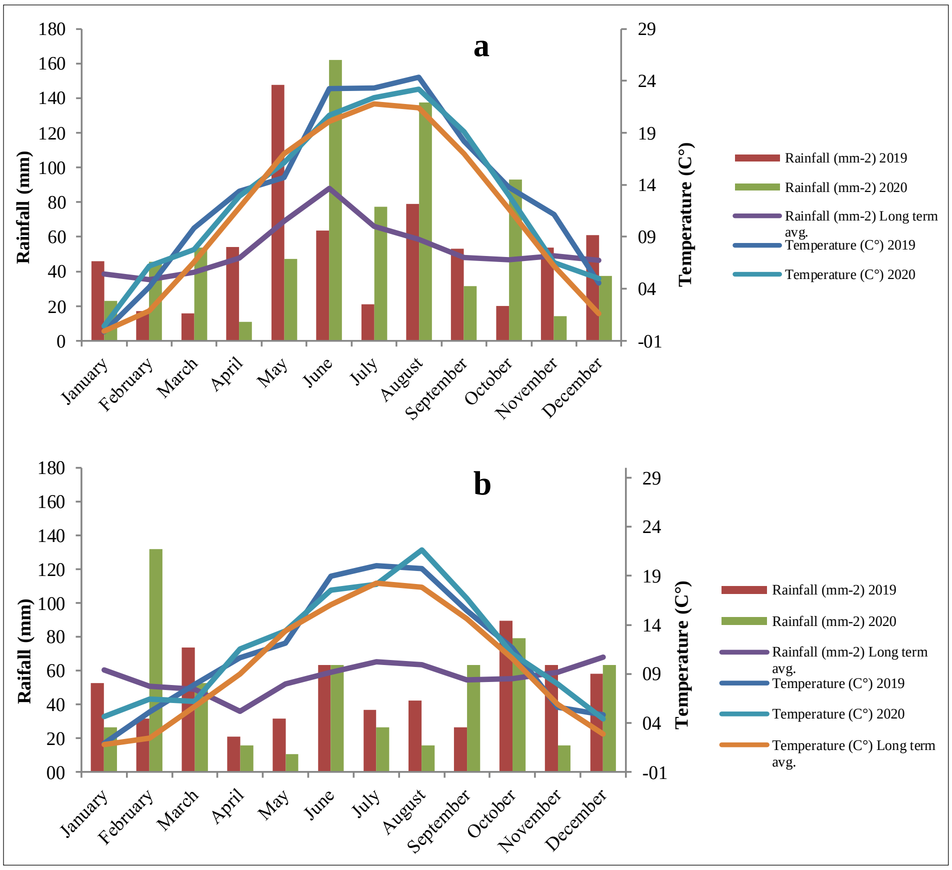
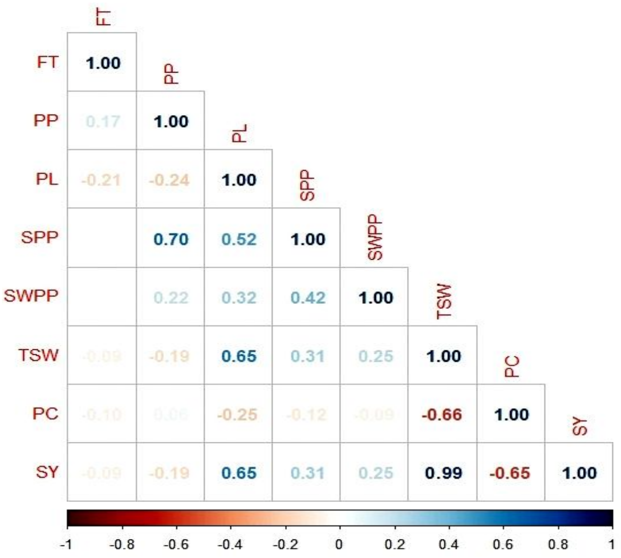


Fig 1: Monthly temperatures and precipitation in 2019 and 2020 in Serbian (a) and Belgian trial (b).

the average of the two-year results at both locations, very small differences were observed in the TSW, which confirms that this trait is highly heritable and genetically determined (Burstin *et al*., 2015; Georgieva *et al*., 2016; Singh *et al*., 2017). Similar can be said for seed protein content, which aligns with the previous research conducted by Crosta *et al*. (2021). The yield is significantly influenced by the interaction of genetic and environmental factors, where cultivation at high temperatures caused by climate change, as well as the amount and distribution of precipitation, contribute to low yield (Acikgoz *et al*., 2009; Atung, 2018).



FT- Time to flowering (days), PP- Pods per plant (#), PL- Pod length (cm), SPP- Seeds per pod (#), SWPP- Seed weight per plant (g), TSW- Thousand seed weight (g), PC- Protein content (%), SY- Plot seed yield (kg/ha).

Fig 2: Correlations between pea traits.

Pearson's correlation coefficients were calculated to evaluate the relationship between traits, using average values from two years (Fig 2). A significant positive correlation was expressed between SY and TSW (0.99) and SPP expressed a positive correlation with PP (0.70) and PL (0.52). Also, a positive correlation was expressed between PL and two traits, TSW and SY (both 0.65). A significant negative correlation was expressed between PC and two traits, TSW (0.66) and SY (0.65). The correlations between the rest of the pairs were of weak or no significance, indicating possible non-linear interactions. A strong positive correlation between SY and TSW is to be expected, this trait is directly related to seed yield and they are mostly positively correlated (Khan *et al*., 2017). On the other hand, TSW is negatively correlated with PC content and the same was observed between SY and PC, which is negative in most cases (Dhama *et al*., 2010; Mohanty *et al*., 2020; Asha *et al*., 2020). TSW had a positive influence on SY, possibly because heavier seeds provide a more favorable condition for the growth of plants that are highly adaptable to agroecological growing conditions. PC, on the other hand, was negatively correlated with SY, indicating a trade-off between protein content and yield. The results of the correlation of PL, which was positively correlated with SPP, TSW and SPP, were similar to Naeem *et al*. (2020), while the positive correlation between SPP and PP were contrary to the findings of Mukherjee *et al.* (2023).

Table 1: The impact of agroecological conditions on pea traits for two locations.

Trait	Trial	Mean	St.Dev.	Min	Max	CV
*FT	Serbia	22.3	2.8	14.6	27.5	12.7
	Belgium	14.9	1.5	11.4	18.8	10.0
*PP	Serbia	11.5	1.3	7.8	15.6	10.9
	Belgium	8.1	0.9	6.5	11.4	11.4
*PL	Serbia	5.8	0.8	4.0	8.8	13.4
	Belgium	5.1	0.6	2.7	7.4	12.1
*SPP	Serbia	8.7	0.6	7.0	10.1	7.2
	Belgium	6.6	0.5	5.4	8.0	7.9
*SWPP	Serbia	7.2	0.6	6.0	9.3	7.8
	Belgium	6.2	1.0	4.1	9.7	16.4
TSW	Serbia	182.6	50.4	57.5	256.4	27.6
	Belgium	185.3	41.9	109.2	288.4	22.6
PC	Serbia	27.3	1.1	25.0	29.8	4.0
	Belgium	27.4	1.0	25.1	30.2	3.6
SY	Serbia	104.9	24.8	43.5	141.5	23.7
	Belgium	106.4	20.7	69.0	156.7	19.5

FT- Time to flowering (days), PP- Pods per plant (#), PL- Pod length (cm), SPP- Seeds per pod (#), SWPP- Seed weight per plant (g), TSW- Thousand seed weight (g), PC- Protein content (%), SY- Plot seed yield (kg/ha).

Note. Statistically significant if p≤0.05 (*).

Understanding protein composition variability in pea genotypes

PCA analysis has been used to gain insight into the similarities and differences among pea varieties based on their physicochemical composition (Guindon *et al*., 2021), basic composition variability (Santos *et al*., 2019), genetic diversity (Ouafi *et al*., 2016) and bioactive compounds (Han *et al*., 2023). Multivariate analysis was performed based on the examination of mean values of all traits for both locations, in order to investigate the population structure of 64 pea genotypes differing in color and seed type. The PCA analysis of the tested pea genotypes for the first two main components is graphically represented (Fig 3). The results of the multivariate analysis show the separation of pigmented seeds from mixed non-pigmented seeds by the first axis (35.9%) and yellow non-pigmented seeds by the second axis (17.9%), with no clear grouping concerning seed type. The results obtained from multivariate analysis in this study indicate a clear distinction between yellow pea seeds and both pigmented and non-pigmented green seeds, with a noticeable separation of mixed pigmented genotypes. In contrast, there was no clear structure to the PCA plot in the analysis based on the division of genotypes by seed type. Gixhari *et al*. (2014) have supported these outcomes with their research on pea, employing PCA to demonstrate that traits such as seed number per pod, weight of 1000 seeds and genotype yield account for a significant portion of the observed variability. Also, this could be related to morphological and physiological properties, similar to the findings of Guindon *et al*. (2021), where yellow varieties showed superior values for weight and seed size, which affect the number of seeds per pod, while wrinkled varieties showed higher protein content.

An additional analysis was carried out to investigate variations in protein composition by categorizing pea genotypes based on color (Fig 4, a) and seed type (Fig 4, b). There were variations in the ratio of vicilin/legumin among seeds of various colors, although they were not statistically significant (p-value = 0.9963 according to the Kruskal-

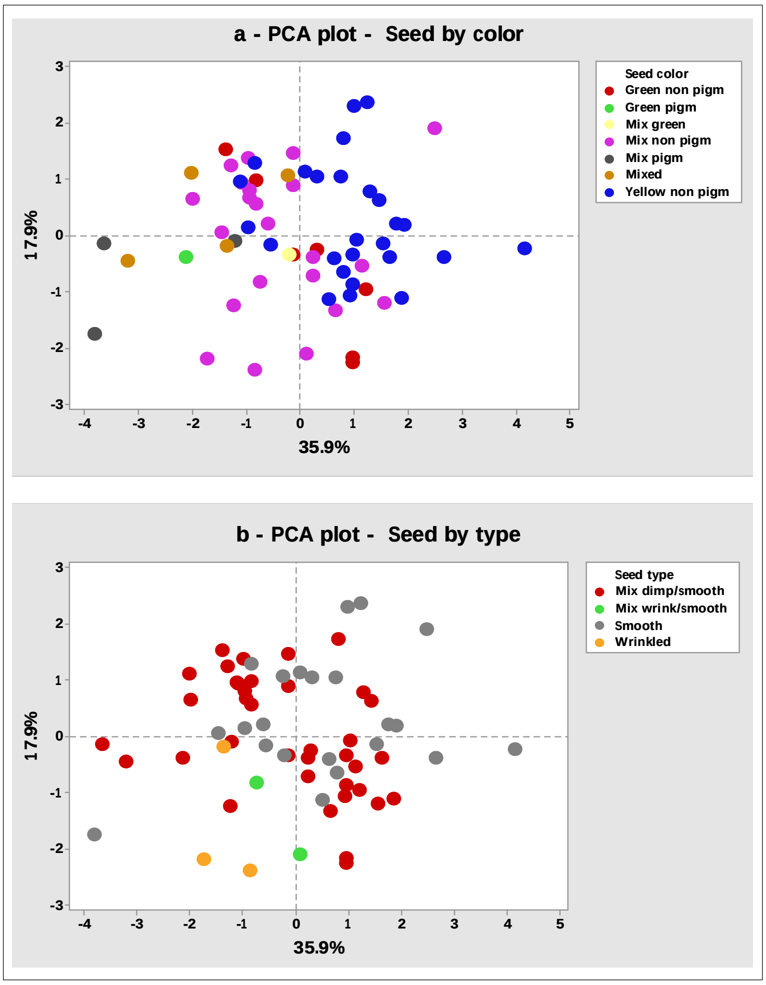


Fig 3: Principal component analysis (PCA) for 64 pea genotypes, grown in two years (2019 and 2020) at two locations (Rimski šanèevi, Serbia and Kessenich, Belgium), classified according to seed color (a) and seed type (b).

Wallis test). According to the visual representation of the results (Fig 4, a), the green non-pigmented seed exhibited the highest concentration of vicilin and the green pigmented seed exhibited the highest concentration of α legumin. The content of convicilin and α legumin was similar across all varieties of seeds, except for genotypes with mixed pigmented seed, which exhibited the lowest level of convicilin content. Concerning seeds of various types, there were no significant variations in protein content among them (p-value = 0.9778 according to the Kruskal-Wallis test), except for genotypes with mixed seed (smooth/ wrinkled), which exhibited the highest concentration of convicilin and the lowest concentration of α legumin (Fig 4, b). The protein composition differed between seeds of different colors, showing more similarities between seeds of different types. Mainly, yellow seeds, which are mostly associated with dry pea genotypes, are considered to have different protein content, compared to green or pigmented seeds, which most often belong to forage or vegetable peas, as well as wild pea relatives. Genotypes with dark seed color exhibited the lowest mean vicilin content compared to other genotypic groups, as indicated by the percentage distribution of protein subunits. Conversely, genotypes with green pigmented seeds displayed the highest α legumin content compared to the observed groups. These findings could bear significant implications for both industrial processes and food production. This is especially relevant due to the emulsification properties of vicilin, which render pea genotypes with higher vicilin content valuable for technological processing (Barać *et al*., 2010). Moreover, genotypes with elevated legumin content offer considerable nutritional value, as confirmed by Villa

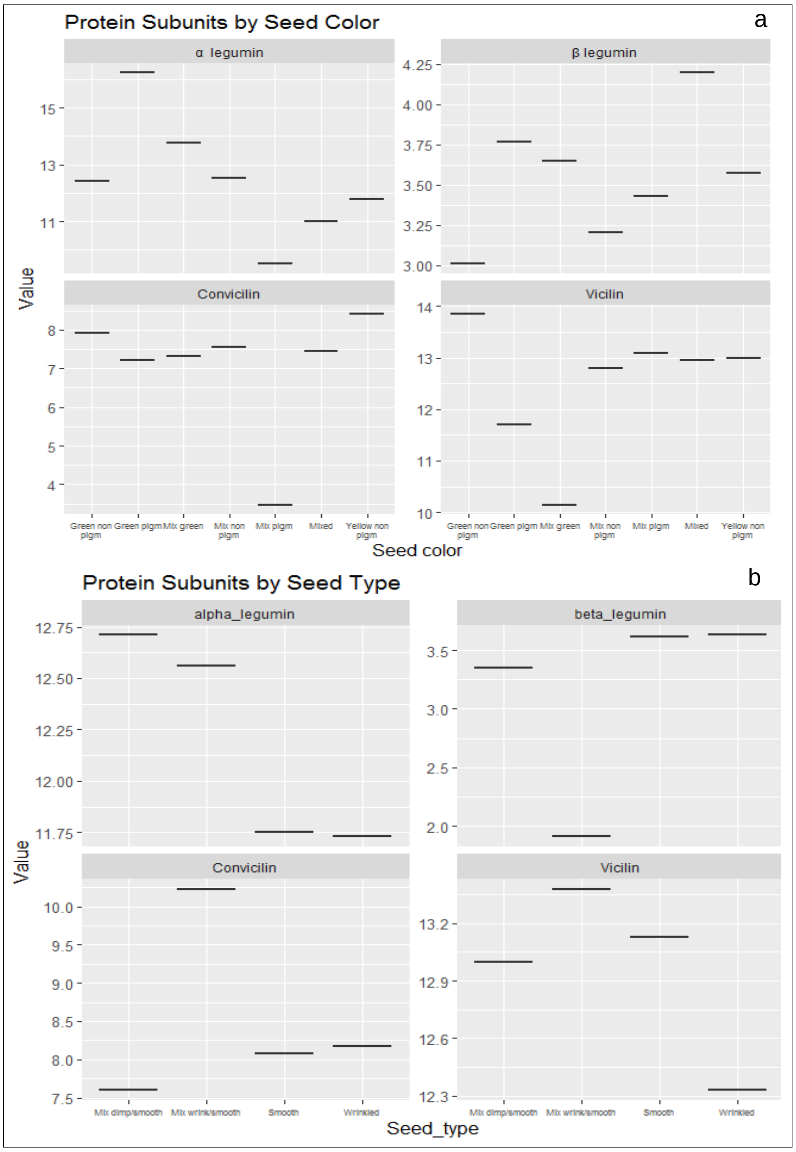


Fig 4: Content of four significant subunits (%) in pea protein, in genotypes differing in seed color (a) and type (b).

et al. (2018). Notably, Gabriel *et al*. (2008) reported a negative correlation between amino acid digestibility and legumin levels, while Rubio *et al*. (2014) found that vicilin fractions are rich in arginine, isoleucine, leucine and lysine compared to the leguminous fraction. It is worth mentioning that genotypes with darker seed color (mixed pigmented) have the lowest representation of α and β legumin subunits.

CONCLUSION

In conclusion, our research emphasizes the profound influence of agroecological conditions on pea traits. We discovered that variations in rainfall and other environmental factors between the two examined localities had a significant impact on the duration of flowering, the number of pods and seeds and also seed weight per plant. The findings underscore the importance of understanding the interplay between environmental conditions and plants, which in turn offer insights and valuable guidance for agricultural practices and the optimization of crop yields in diverse agroecological settings.

Shifting the focus to protein composition, our research shed light on notable disparities among pea seeds of different colors and types. Particularly, genotypes with dark seed color exhibited the lowest vicilin content, while those with green pigmented seeds displayed the highest á legumin content. These variations carry significant implications for the food industry and nutrition. Vicilin content enhances technological processing, while legumin content enriches nutritional value. Our findings hold promise for informing the potential use and diverse applications of different varieties of pea genotypes.

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Conflict of interest

All authors declare that they have no conflicts of interest.

REFERENCES

Acikgoz, Å., Ustun, A., Gul I., Anlarsal, E., Tekeli, A.S., Nizam, I., Avcıoglu, R., Geren, H., Cakmakci, S., Aydinoglu, B., Yucel, C., Avci, M., Acar, Z., Ayan, I., Uzun, A., Bilgili, U., Sincik, M., Yavuz, M. (2009). Genotype × environment interaction and stability analysis for dry matter and seed yield in field pea (*Pisum sativum* L.). Spanish Journal of Agricultural Research. 7(1): 96-106.

Anderson, V.L., Lardy, G.P., Ilse, B.R. (2007). Field pea grain for beef cattle. The Professional Animal Scientist. 23(1): 1-7.

Asha, A.B., Srinivasa, V., Hanumantappa, M., Aghora, T.S., Ganapathi, M., Chithra, K. (2020). Correlation and path analysis in garden pea (*Pisum sativum* L.). Journal of Pharmacognosy and Phytochemistry. 9(5): 1728-1731.

Avci, M.A., Ceyhan, E. (2006). Correlations and genetic analysis of pod characteristics in pea (*Pisum sativum* L.). Asian Journal of Plant Sciences. 5(1): 1-4.

Atung, C.K. (2018). Effects of high temperature on pea (*Pisum sativum*) seed quality and attributed traits: A thesis presented in partial fulfilment of the requirements for the degree of Master of Science. Plant Breeding at Massey University, Manawatu, New Zealand (Doctoral Dissertation, Massey University).

Barać, M., Pešić, M., Stanojević, S., Macej, O., Ristić, N., Èabrilo, S., Žilić, S. (2010). Profile and functional properties of seed proteins from six pea (*Pisum sativum*) genotypes. International Journal of Molecular Sciences. 11(12): 4973-4990.

Barać, M., Čabrilo, S., Pešić, M., Stanojević, S., Pavlićević, M., Maćej, O., Ristić, N. (2011). Functional properties of pea (*Pisum sativum* L.) protein isolates modified with chymosin. International Journal of Molecular Sciences. 12(12): 8372-8387.

Barcchiya, J., Naidu, A.K., Mehta, A.K., Upadhyay, A. (2018). Genetic variability, heritability and genetic advance for yield and yield components in pea (*Pisum sativum* L.). International Journal of Chemical Studies. 6(2): 3324-3327.

Benesty, J., Chen J., Huang, Y., Cohen, I. (2009). Pearson Correlation Coefficient. Springer Topics in Signal Processing Book Series. STSP, vol 2.

Bozoglu, H., Peksen, E., Peksen, A., Gulumser, A. (2007). Determination of the yield performance and harvesting periods of fifteen pea (*Pisum sativum* L.) cultivars sown in autumn and spring. Pakistan Journal of Botany. 39(6): 2017-2025.

Burstin, J., Salloignon, P., Chabert-Martinello, M., Jean-Bernard Magnin-Robert, J., Siol, M., Jacquin, F., Chauveau, A., Pont, C., Gregoire Aubert, G., Delaitre, C., Truntzer, C., Duc, G. (2015). Genetic diversity and trait genomic prediction in a pea diversity panel. BMC Genomics. 16: 1-17.

Clemente, A., Arques, M.C., Dalmais, M., Le Signor, C., Chinoy, C., Olias, R., Rayner, T, Isaac, P.G, Lawson, D.M, Bendahmane, A., Domoney, C. (2015). Eliminating anti-nutritional plant food proteins: The case of seed protease inhibitors in pea. Plos One. 10(8): e0134634.

Choudhary, R., Singh, B.K., Choudhary, A., Choudhary, J.S., Jat, S.K. Kumar, R. (2023). Influence of Plant growth regulators on growth, yield and yield components in garden pea. Legume Research. 46(10): 1366-1369. doi: 10.18805/LR-4880.

Corder, G.W., Foreman, D.I. (2009). Nonparametric Statistics for Non-Statisticians. Hoboken: John Wiley and Sons. pp. 99-105.

Crosta, M., Nazzicari, N., Ferrari, B., Pecetti, L., Russi, L., Romani, M., Cabassi, G., Cavalli, D., Marocco, A., Annicchiarico, P. (2021). Pea grain protein content across italian environments: Genetic relationship with grain yield and opportunities for genome. enabled selection for protein yield. Frontiers in Plant Science. 12: 718713-718713.

Cullis, B.R., Smith, A.B., Coombes, N.E. (2006). On the design of early generation variety trials with correlated data. Journal of Agricultural, Biological and Environmental Statistics. 11: 381-393.

Daba, S.D., Morris, C.F. (2021). Pea proteins: Variation, composition, genetics and functional properties. Cereal Chemistry. 00: 1-13.

Dahl, W.J., Foster, L.M., Tyler, R.T. (2012). Review of the health benefits of peas (*Pisum sativum* L.). British Journal of Nutrition. 108(S1): S3-S10.

Data Access Viewer, Prediction of Worldwide Energy Resource (https://power.larc.nasa.gov/data-access-viewer/).

Demidenko, E., Mandel, I. (2005). Yield analysis and mixed model. ASA Section on Nonparametric Statistics. 1636-1643.

Dhama, S.K., Tyagi, N.K., Singh, P.B. (2010). Interrelationship and path analysis for seed yield and its component characters under eight environments in pea (*Pisum sativum* L.). Legume Research: An International Journal. 33(2): 87-94.

Faostat Statistics, (2021). Crop and livestock products-statistics on area harvested of dry pea in European region. https://www.fao.org/faostat/en/#data/QCL).

Gabriel, I., Quillien, L., Cassecuelle, F., Marget, P., Juin, H., Lessire, M., Sève, B., Duc, G., Burstin, J. (2008). Variation in seed protein digestion of different pea (*Pisum sativum* L.) genotypes by cecectomized broiler chickens: 2. Relation between *in vivo* protein digestibility and pea seed characteristics and identification of resistant pea polypeptides. Livestock Science. 113(2-3): 262-273.

Georgieva, N., Nikolova, I., Kosev, V. (2016). Evaluation of genetic divergence and heritability in pea (*Pisum sativum* L.). Journal of BioScience and Biotechnology. 5(1): 61-67.

Ghixari, B, Vrapi, H, Hobdari, V. (2014). Morphological characterization of pea (*Pisum sativum* L.) genotypes stored in Albanian genebank. Albanian Journal of Agricultural Sciences. (special edition).

Guindon, M.F., Aguero, M.G., Gatti, I., Cointry, E. (2021). Comparative analysis of the physicochemical composition of pea cultivars. Ciencia y Tecnología Agropecuaria. 22(2).

Haile, S. (2020). Genotype × environment interaction and grain yield stability of finger millet [*Eleusine coracana* (L.) subsp. Coracana] varieties in eastern and western Åthiopia. Doctoral Dissertation, Haramaya University.

Han, X., Akhov, L., Ashe, P., Lewis, C., Deibert, L., Zaharia, L.I., Forseille L., Xiang D., Datla R., Nosworthy M., Henry C., Zou J., Yu B., Patterson, N. (2023). Comprehensive compositional assessment of bioactive compounds in diverse pea accessions. Food Research International. 165: 112455. https://doi.org/10.1016/j.foodres.2022.112455.

Isik, F., Holland, J., Maltecca, C. (2017). Genetic Data Analysis for Plant and Animal Breeding, Vol. 400. Cham, Switzerland. Springer International Publishing.

Ismail, B.P., Senaratne-Lenagala, L., Stube, A., Brackenridge, A. (2020). Protein demand: Review of plant and animal proteins used in alternative protein product development and production. Animal Frontiers. 10(4): 53-63.

ISTA, (2015). International Rules for Seed Testing. International Seed Testing Association.

Khan, R.A., Mahmud, F., Reza, M.A., Mahbub, M., Shirazy, J.B., Rahman, M. (2017). Genetic diversity, correlation and path analysis for yield and yield components of pea (*Pisum* sativum L.). World Journal of Agricultural Sciences. 13: 11-16.

Kumar, A., Kumar, H., Sharma, V. and Kamaluddin (2024). Estimation of genetic parameters, selection indices and association analysis of seed yield and its component traits in chickpea (*Cicer arietinum* L.). Legume Research. 47(3): 352-360. doi: 10.18805/LR-4506.

Kuppler, J., Kotowska, M.M. (2021). A meta analysis of responses in floral traits and flower-visitor interactions to water deficit. Global Change Biology. 27(13): 3095-3108.

Laemmli, U.K. (1970). Cleavage of structural proteins during the assembly of the head of bacteriophage T4. Nature. 227(5259): 680-685.

Lam, A.C.Y., Can Karaca, A., Tyler, R.T., Nickerson, M.T. (2018). Pea protein isolates: Structure, extraction and functionality. Food Reviews International. 34(2): 126-147.

Maenhout, S. (2018). Progeno. Gent: Ghent University, (https://progeno.net/).

Maurya, P., Angom, B., Kumar, D. (2015). International conference on role of agri-science, forestry, food technology and participatory natural resource management for mitigation of climate change. New Delhi, Jawahar Lal University. 2(1): 40-44.

Mohanty, A.T., Verma, A., Pandey, B. (2020). Evaluation of correlation and path coefficients analysis for yield attributing traits in garden pea (*Pisum* sativum L.) under tarai tract of Uttarakhand. International Journal of Current Microbiology and Applied Sciences. 11: 1315-1322.

Mohapatra, C., Chand, R., Tiwari, J.K., Singh, A.K. (2020). Effect of heat stress during flowering and pod formation in pea (*Pisum sativum* L.). Physiology and Molecular Biology of Plants. 26(6): 1119-1125.

Mukherjee, E., Chaudhary, L. and Kumar, M. (2023). Morphological characterization and genetic variability study on early maturing pigeonpea [*Cajanus cajan* (L.) Millsp.] lines. Legume Research. 46(9): 1117-1125. doi: 10.18805/LR-5153.

Naeem, M., Ghani, M.A., Atif, R.M., Amjad, M., Ahmad, T., Batool, A., Jahangir, M.M., Ali, B., Abbas, M.M., Akram, M.N. (2020). Estimation of correlation and path coefficient for morphological and quality related traits in pea (*Pisum sativum* L.). Bangladesh Journal of Botany. 49(3): 549-555.

Nassir, A.L., Ariyo, O.J. (2011). Genotype × environment interaction and yield-stability analyses of rice grown in tropical inland swamp. Notulae Botanicae Horti Agrobotanici Cluj-Napoca. 39(1): 220-225.

Ouafi, L., Alane, F., Rahal-Bouziane, H. Abdelguerfi, A. (2016). Agro-morphological diversity within field pea (*Pisum sativum* L.) genotypes. African Journal of Agricultural Research. 11(40): 4039-4047. doi: 10.5897/AJAR2016.11454.

Republic Hydrometeorological Institute of Serbia-RHMZ (URL http://vvv.hidmet.gov.rs/indek_eng.php).

Rubio, L.A., Perez, A., Ruiz, R., Guzman, M.A., Aranda-Olmedo, I., Clemente, A. (2014). Characterization of pea (*Pisum sativum*) seed protein fractions. Journal of the Science of Food and Agriculture. 94(2): 280-287.

Santos, C.S., Carbas, B., Castanho, A., Vasconcelos, M.W., Vaz Patto, M.C., Domoney, C., Brites, C. (2019). Variation in pea (*Pisum sativum* L.) seed quality traits defined by physicochemical functional properties. Foods. 8(11): 570. doi: 10.3390/foods8110570.

Saxesena, R.R., Vidyakar, V., Vishwakarma, M.K., Yadav, P.S., Meena, M.L., Lal, G.M. (2014). Genetic variability and heritability analysis for some quantitative traits in field pea (*Pisum sativum* L.). The Bioscan. 9(2): 895-898.

Singh, B.K., Sutradhar, M., Singh, A.K., Singh, S.K. (2017). Evaluation of genetic variability, correlation and path coefficients analysis for yield attributing traits in field pea [*Pisum sativum* (L.) var. arvense]. Research on Crops. 18(2): 316-321.

Stjepanović, M., Čupić, T., Gantner, R. (2012). Grašak. Poljoprivredni Fakultet Osijek.

Tawaha, A.M., Turk, M.A. (2004). Field pea seeding management for semiarid Mediterranean conditions. Journal of Agronomy and Crop Science. 190(2): 86-92.

UPOV, (2022). Guidelines for the conduct of test for distinctness, uniformity and stability. Pea (*Pisum saivum* L.). International Union for The Protection of New Varieties of Plants.

Villa, C., Costa, J., Oliveira, M.B.P. (2018). Bovine milk allergens: A comprehensive review. Comprehensive Reviews in Food Science and Food Safety. 17(1): 137-164.

Westhoek, H., Rood, T., van den Berg, M., Janse, J., Nijdam, J., Reudink, M., Stehfest, E. (2011). The protein puzzle, The consumption and production of meat, dairy and fish in the European Union. PLB Netherland Environmental Assessment Agency. 2011. https://www.pbl.nl/en.

Wei, T., Simko, V. (2017). R Package “corrplot”: Visualization of a Correlation Matrix. R package version 0.84. 2017. https://cran.r-project.org/package=corrplot.

Wickham, H. (2016). ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York. ISBN 978-3-319-24277-4, URL https://ggplot2.tidyverse.org/.