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# Quality parameters of maize hybrids intended for silage production

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**A B S T R A C T**

This study's primary objective was to assess the quality parameters of fifteen different genotypes of Serbian maize hybrids in order to establish whether they were suitable for making high-quality silage for ruminant feed. The study was carried out as a two-year field experiment at the Maize Research Institute in Zemun Polje, Serbia. Laboratory analyses included the yield structure of the examined maize hybrids, the evaluation of the composition of lignocellulosic fiber and the percentage of their ratios, as well as the *in vitro* dry matter digestibility and NDF digestibility of the whole plant samples. The Fisher's LSD test was used to statistically analyse the data, which were then reported as the mean and standard deviation of at least three independent replicates. The graphical representation of correlations among the observed parameters used the principal component analysis (PCA) method. All maize hybrids have displayed high-quality characteristics, which are required for the production of high-quality silage.

**Keywords**: maize hybrids, lignocellulosic fibers, *in vitro* dry matter digestibility, silage, animal feed.

**И З В О Д**

Основни циљ овог истраживања био је да се испитају својства петнаест различитих генотипова хибрида кукуруза из Србије како би се утврдила њихова погодност за производњу висококвалитетне силаже за исхрану преживара. Истраживање је спроведено у двогодишњем пољском огледу на локацији Института за кукуруз у Земун Пољу, Србија, а лабораторијске анализе су обухватиле структуру приноса испитиваних хибрида кукуруза, процену састава лигноцелулозних влакана и њихових процентуалних односа, као и *in vitro* сварљивост суве материје и сварљивост NDF-а узорака целе биљке. Подаци су статистички обрађени коришћењем ANOVA анализе варијансе са Фишеровим LSD тестом и приказани као средња вредност ± стандардно одступање од најмање три независна понављања. Анализа главних компонената (PCA) примењена је за визуелно представљање корелација између посматраних параметара. Сви хибриди кукуруза су показали повољна својства која су предуслов за производњу висококвалитетне силаже.

**Кључне речи**: хибриди кукуруза, лигноцелулозна влакна, *in vitro* сварљивост суве материје, силажа, храна за животиње.

## 1. Introduction

Maize (*Zea mays* L.) is the predominant cereal crop in the Republic of Serbia grown on 53% of the harvested area under cereals, followed by wheat (*Triticum aestivum* L.) grown on 38% of this arable land (Grčak et al., 2020). The total 2021 world production of maize reached 1125.03 million metric tons (Shahbandeh, 2021). The first double-cross inbred maize, developed in 1918 by D. F. Jones and experimentally released in 1924 by H. A. Wallace, was the beginning of maize hybrids cultivation (Sutch, 2011). The earliest attempts to ensile maize for feed were made in the late nineteenth century; nevertheless, it wasn't until decades later, after the development of flint x dent hybrids resistant to cold temperatures, that silage maize became widely used in cattle feeds (Barrière et al., 2018). Nowadays, silage maize is one of the most significant annual forage crops regarded as the primary energy source for ruminant feeding around the world, mostly for lactating dairy cows, due to high energy requirements of the feed for maximum milk production (Marsalis et al., 2010). It is simple to manufacture and preserve silage maize, and it can be consumed every day all year round (Barrière et al., 2018). Thanks to the growing progress of silage technology, whole maize plant is one of the most crucial feedstuffs for animals in the world today (Ayaşan et al., 2020). Developing hybrids with enhanced whole plant yield, nutritional value, and agronomic features that give superior ensiling quality has recently received a lot of attention in the breeding of silage maize (Terzić et al., 2020). Studies have revealed that after the plant reaches its physiological peak, the *in vitro* digestibility of forages declines as plant maturity grows (Johnson, 1999).

Plant biomass intended for ensiling needs to contain a certain amount of dry matter composed of different nutrients such as crude protein, crude fat, crude fiber, crude ash, and nitrogen-free extract. Otherwise, the feed that contains more than 60 to 67% moisture is harder for handling and more expensive, prone to producing mucous, moldy silage due to the presence of butyric acid and other undesirable short chain fatty acids, which can lead to the loss of valuable nutrients (Stanaćev and Vik, 2002). Higher starch content in the silage can be obtained when ensiling maize with a high dry matter content, which can lead to problems in preventing aerobic spoilage, caused by low lactic acid bacteria activity during fermentation (Ferrero et al., 2019). Silage maize quality is primarily determined by its composition, i.e. dry matter (d.m.), crude protein, starch, fiber content, as well as dry matter digestibility; however, the important factors that also influence silage quality are fermentation parameters during the ensiling process and its aerobic stability (Serva et al., 2021). The nutritional quality of silage maize is by large determined by the quantity and category of plant cell wall material, i.e. lignocellulosic fibers. Because variations in silage quality might affect overall animal performance, maize's nutrient and fiber content should be as desirable as feasible. Given that a high lignocellulosic fiber content in forage maize might result in restricted digestibility and fodder intake in ruminants, forage analysis is the best method for determining if silage maize is suitable for ruminant nutrition (Kokić et al., 2018; Kruse et al., 2008).

The primary goal of this research was to investigate some of the most significant quality traits of the whole plant of fifteen maize hybrids harvested at the physiological maturity stage, in order to ascertain if they were suitable for manufacturing of silage for ruminant feed.

## 2. Materials and methods

### *2.1. Plant material*

Fifteen dent maize hybrids of divergent genetic backgrounds and maturity groups, namely: ZP 173/8, ZP 377, ZP 440, ZP 444, ZP 560, ZP 600, ZP 606, ZP 623, ZP 643, ZP 666, ZP 667, ZP 679, ZP 745, ZP 747, and ZP 749, developed at the Zemun Polje Maize Research Institute were put to the test in field trials over the course of two years (2019 and 2020).

### *2.2. Experimental site and setup*

The field experiment was set up using a randomized complete block design with two replicates in the Maize Research Institute's experimental field in Zemun Polje, Belgrade, Serbia (44°52'N, 20°19'E, 81m asl). The sowing density was 60,000 plants per hectare, and the plot size was 21 m2. Five average plants from each replication were chosen for additional testing after being picked from the two inner rows of the experimental plot (7 m2). The harvest occurred between the quarter and half milk-line kernel stages (about 30-35%), which is the full waxy phase of maize maturity ideal for silage production.

### *2.3. Plant analyses*

In order to achieve constant moisture, samples of whole plants (WP), stover, or plants without ears (S), and ears (E) were chopped and dried at 60 °C for 48 hours in a forced-air drying oven. They were then pulverized in a mill using 1-mm mesh sieves.

### *2.4. Chemical analyses*

The samples were dried at 105 °C in a lab drying oven for 12 hours, or until a consistent mass was attained, in order to determine the total dry matter content. According to the detergent method by Van Soest (1980), with minor modifications, the fiber analysis comprised determination of the lignocellulosic constituents: neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL), as well as hemicellulose and cellulose (Mertens, 1992). The principle of this method is based on the changes in the solubility of individual lignocellulosic fibers in neutral, acidic, and alkaline solutions. The cellulose content was determined as the difference between the NDF and ADF contents, whereas the hemicellulose content was determined as the difference between the NDF and ADF contents. The analyzed maize hybrids' whole plant samples' in vitro dry matter digestibility (IVDMD) was determined using Aufrere's enzymatic pepsin-cellulase technique (2007). The results are displayed as a percentage of dry matter (d.m.). The formula proposed by Barrière et al. (2018) was used to compute the proportion of NDF digested during the in vitro rumen fluid process: NDFD=100 x (IVDMD - (100 - NDF))/NDF.

### *2.5. Statistical procedures*

The data were examined using one-way ANOVA analysis of variance along with Fisher's F test in the Minitab19 Statistical Software, and the results were presented as the mean ± standard deviation of at least three independent replicates. Using the LSD test, differences between the means with probability $P<0.05$ were considered statistically significant. The relationships between the observed values were shown visually using the principal component analysis (PCA) function in Minitab19 Statistical Software. An acute angle between two measures indicated a positive correlation, whereas an obtuse angle indicated a negative correlation.

## 3. Results and discussions

Figure 1 depicts the yield structure of the examined maize hybrids. The dry matter content of the entire plant varied between 33.89% (ZP 666) and 41.34% (ZP 747). Hybrid ZP 749 had the highest dry matter yield of the whole plant (23.25 t ha$^{-1}$), ZP 623 had the highest stover dry matter yield (12.55 t ha$^{-1}$), while the highest dry matter yield of maize ear (11.30 t ha$^{-1}$) was obtained by hybrid ZP 745. According to a study by He et al. (2021), the dry matter content of the entire maize plant ranged from 39.7% to 50.1%, while the average levels of NDF, ADF, and ADL were 40.1%, 22%, and 1.5%, respectively.

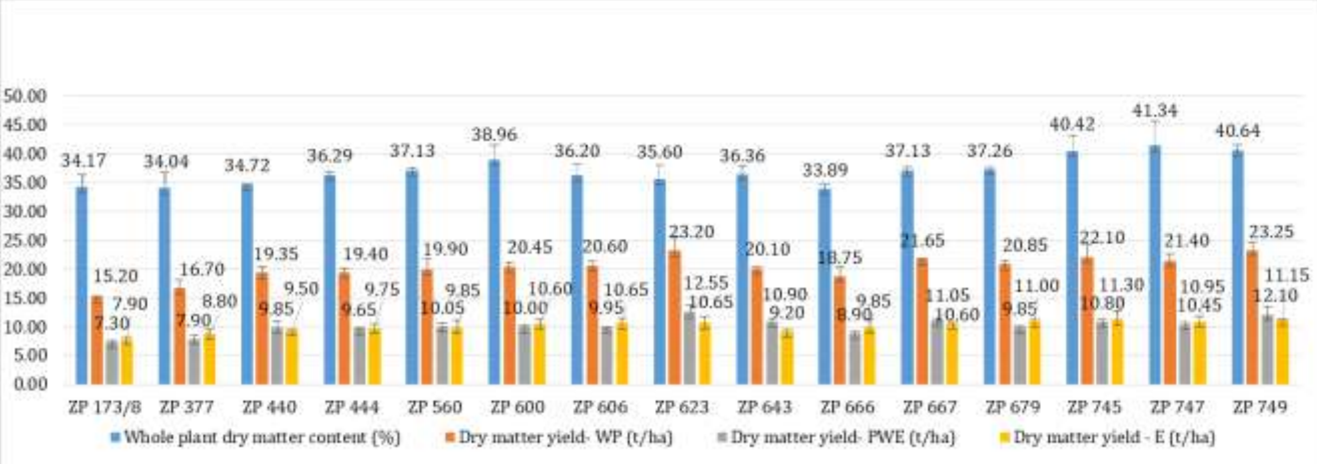


**Figure 1**. Yield structure of the investigated maize hybrids

The type and quantity of lignocellulosic fibers are among the most important parameters in defining the nutritional quality and technological grade of maize biomass for ruminant nutrition. (Terzić et al., 2020). Cell wall components found in NDF include cellulose, hemicellulose, lignin, and silica. The essential roles of the NDF fraction in ruminant nutrition are to supply energy for microbial metabolism, control rumen activity, and maintain animal health by giving the diet more structural value. There are two fractions of NDF: a potentially digestible NDF and indigestible NDF. Lignin is the main barrier to fiber digestion, which prevents enzymes from accessing the cellulose and hemicellulose components of the digestible fiber. Lignin is crucial from a nutritional standpoint as well because it cannot be broken down by rumen microorganisms and is completely indigestible. Lignin's interactions with other components of the cell wall diminish the amount of fiber that may be digested, decrease the availability of the cellulose and hemicellulose fractions of the silage, and thus affect the amount of indigestible NDF (Koukolová et al., 2004). Additionally, NDF is crucial for ruminants even though it can be a bad indicator of silage quality. ADF has a negative relationship with fodder digestibility and is primarily composed of cellulose, lignin, and inorganic silica (Bittman, 2004). According to studies, the plant's dry matter and starch contents rise as it matures further due to the buildup of starch in the ear, whereas the whole-crop fiber content of NDF, ADF, and crude protein tends to decline with time to balance out the rise in stover cell wall content (Kruse et al., 2008; Johnson et al., 1999). Animals choose to eat less forage when the NDF fraction of stover rises with the maize plant's increasing maturity. The variations in the proportions of plant organs as well as their unique ratios of cell wall and content can be seen in the lignocellulosic fractions' whole plant contents. The analysis of the lignocellulosic fibers of the whole plant showed that the NDF content ranged from 43.49% (ZP 600) to 49.06% (ZP 667); ADF from 23.44% (ZP 747) to 26.29% (ZP 679); ADL from 3.29% (ZP 560) to 4.19% (ZP 679); hemicellulose from 20.15% (ZP 600) to 23.45% (ZP 667), and cellulose from 19.78% (ZP 600) to 23.33% (ZP 444), respectively (Figure 2).

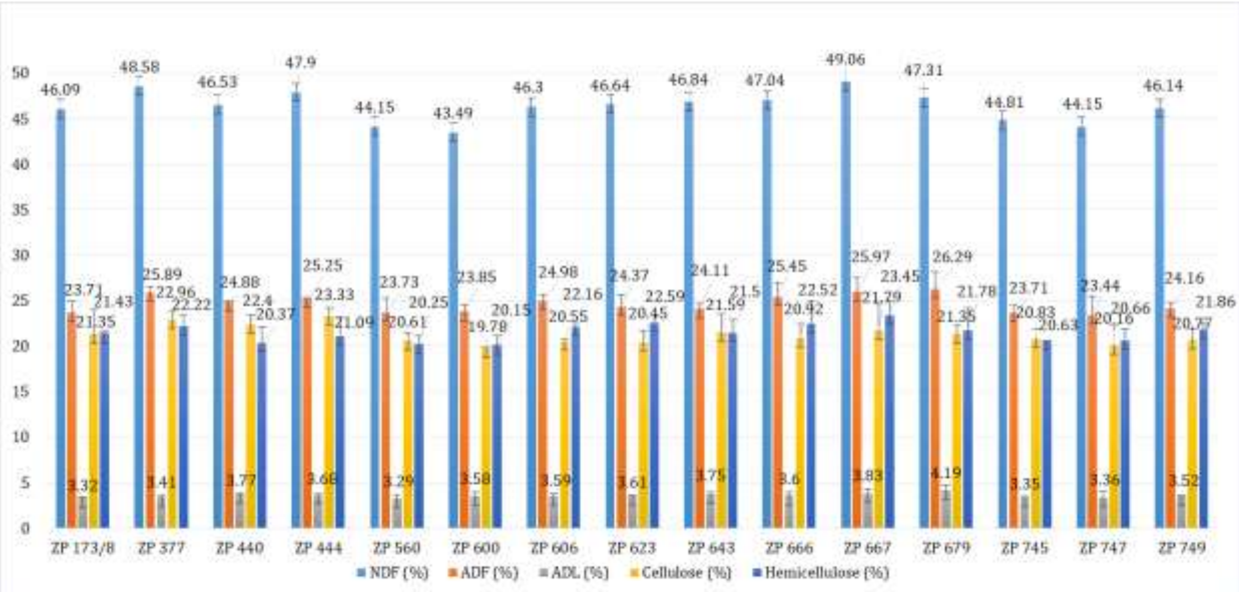


**Figure 2.** Content of the lignocellulosic fibers of the whole maize plant

The average ratios of the various lignocellulosic fibers of the entire maize plant, including ADL/NDF, ADL/ADF, ADF/NDF, hemicellulose/NDF, cellulose/NDF, cellulose/hemicellulose, and hemicellulose/cellulose, ranged from, respectively: 7.02–8.85%, 13.17–15.92%, 51.43–55.57%, 43.77–48.44%, 43.85–48.71%, 90.53–110.62% and 90.40–110.46% (Table 1). These results are in agreement with those of earlier studies by Nikolić et al. (2020), Milašinović-Šeremešić et al. (2017), and Khan et al. (2015). The ADF/NDF ratio is used for the prediction of the concentration of indigestible NDF (iNDF) (Koukolová et al., 2004).

**Table 1.**

Average ratios of the lignocellulosic fibers of the whole plant of maize hybrids

| Hybrid | ADL/NDF | ADL/ADF | ADF/NDF | HC/NDF | C/NDF | C/HC | HC/C |
|---|---|---|---|---|---|---|---|
| ZP 173/8 | 7.19 | 13.98 | 51.43 | 46.50 | 46.32 | 99.63 | 100.37 |
| ZP 377 | 7.02 | 13.17 | 53.29 | 45.73 | 47.25 | 103.33 | 96.78 |
| ZP 440 | 8.09 | 15.14 | 53.46 | 43.77 | 48.14 | 109.99 | 90.92 |
| ZP 444 | 7.68 | 14.57 | 52.71 | 44.03 | 48.71 | 110.62 | 90.40 |
| ZP 560 | 7.45 | 13.86 | 53.75 | 45.87 | 46.68 | 101.78 | 98.25 |
| ZP 600 | 8.22 | 14.99 | 54.84 | 46.32 | 45.47 | 98.16 | 101.87 |
| ZP 606 | 7.74 | 14.35 | 53.95 | 47.85 | 44.38 | 92.76 | 107.81 |
| ZP 623 | 7.74 | 14.82 | 52.25 | 48.44 | 43.85 | 90.53 | 110.46 |
| ZP 643 | 8.01 | 15.55 | 51.47 | 45.90 | 46.09 | 100.42 | 99.58 |
| ZP 666 | 7.64 | 14.13 | 54.10 | 47.87 | 44.47 | 92.90 | 107.65 |
| ZP 667 | 7.80 | 14.73 | 52.94 | 47.80 | 44.42 | 92.92 | 107.62 |
| ZP 679 | 8.85 | 15.92 | 55.57 | 46.04 | 45.13 | 98.03 | 102.01 |
| ZP 745 | 7.48 | 14.13 | 52.90 | 46.04 | 46.49 | 100.97 | 99.04 |
| ZP 747 | 7.60 | 14.31 | 53.09 | 46.80 | 45.66 | 97.58 | 102.48 |
| ZP 749 | 7.63 | 14.57 | 52.35 | 47.37 | 45.00 | 95.01 | 105.25 |
| Min | 7.02 | 13.17 | 51.43 | 43.77 | 43.85 | 90.53 | 90.40 |
| Max | 8.85 | 15.92 | 55.57 | 48.44 | 48.71 | 110.62 | 110.46 |
| Mean | 7.74 | 14.55 | 53.21 | 46.42 | 45.87 | 98.97 | 101.37 |
| SD | 0.44 | 0.69 | 1.14 | 1.34 | 1.42 | 5.94 | 5.91 |

C – cellulose; HC – hemicellulose

The qualitative characteristics of maize hybrids are determined by the plant's morphology and structure (Bertoia & Aulicino, 2014). Different hybrids have different levels of various maize plant components' digestibility. According to Barrière et al. (2005) findings, modern hybrids have an average 5.5% lower *in vivo* cell wall digestibility than older hybrids, despite a slight but considerable increase in the grain share in the overall maize plant. Ayaşan et al. (2020), for instance, showed statistically significant differences in IVDMD between various portions of the maize plant, with the kernel having the highest IVDMD (79.06%) and the lower stalk having the lowest IVDMD (38.13%).

However, silage digestibility is constrained by the percentage of lignification of the cell wall components. According to a study by Crevelari et al. (2018), the genetic variations across maize hybrids affect how digestible a crop is in the rumen. The IVDMD value for the hybrid ZP 173/8 was the highest (63.87%), followed by ZP 606 (61.37%), and ZP 444 (61.00%) (Figure 2). Of all the genotypes of maize, the hybrid ZP 377 has the lowest *in vitro* dry matter digestibility (58.38%) (Figure 3).

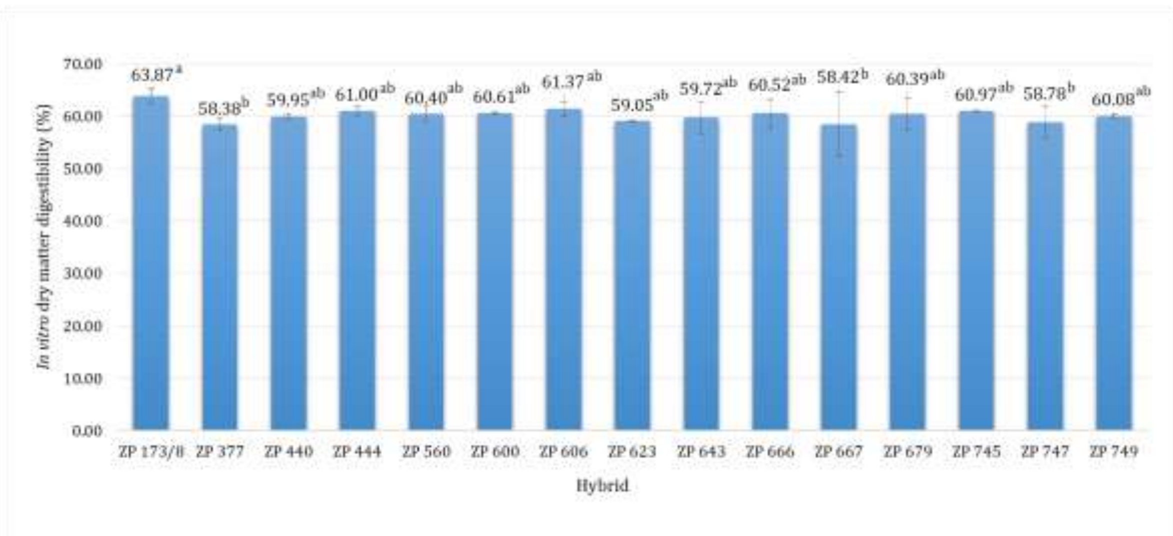


**Figure 3.** *In vitro* dry matter digestibility of the maize hybrids (%)

In recent years, some researchers have begun evaluating potential NDF digestibility as a critical quality indicator in new feed evaluation systems in addition to the presence of lignocellulosic fibers (Koukolová et al., 2004). Moore and Jung (2001) concluded that high IVDMD resulting from a higher grain share due to the more advanced maturity stage the hybrids were harvested at can be distinguished from those which have genetically better fiber quality by comparing NDFD values. Decreased NDF digestibility indicates poor dry matter digestibility, and as a result, feed's energy value and animals' performance are both decreased (production of meat and milk) (Milašinović-Šeremešić et al., 2017). The average NDF digestibility obtained in our study ranged from 6.64% (ZP 747) to 21.61% (ZP 171/8) (Figure 4). They are consistent with levels that have previously been reported by a number of studies (Nikolić et al., 2020; Barrière et al., 2018). Miller et al. (2021) found that higher forage NDFD caused greater NDF and that a higher NDF diet with lower NDFD decreased milk production and dry matter intake in dairy cows. These findings were associated with high levels of ruminal fill, and increased retention times of organic matter and NDF in the rumen.

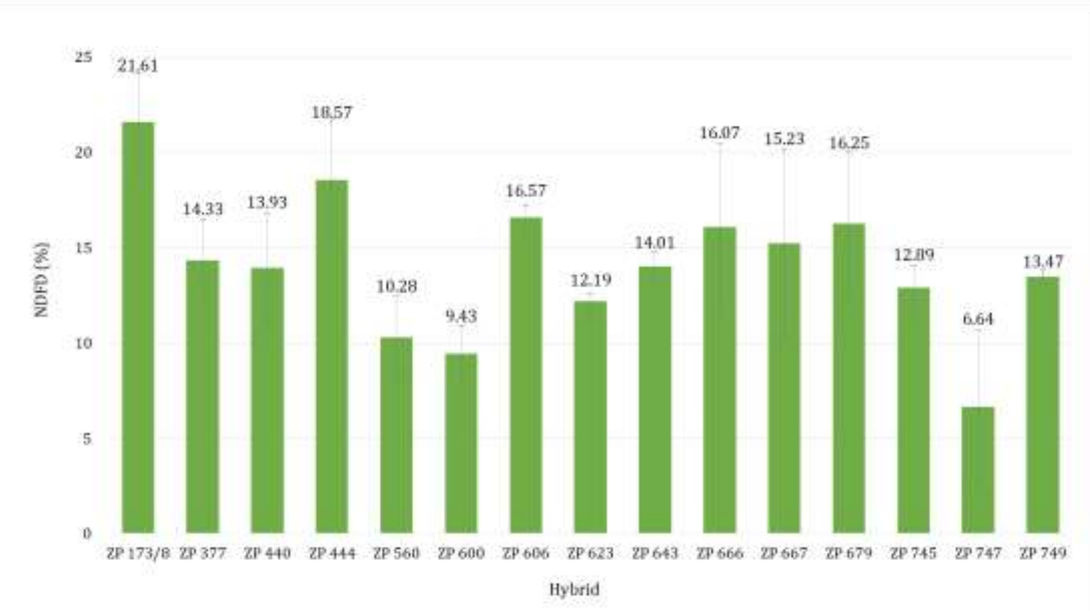


**Figure 4.** Average neutral detergent fiber digestibility (NDFD) (%)

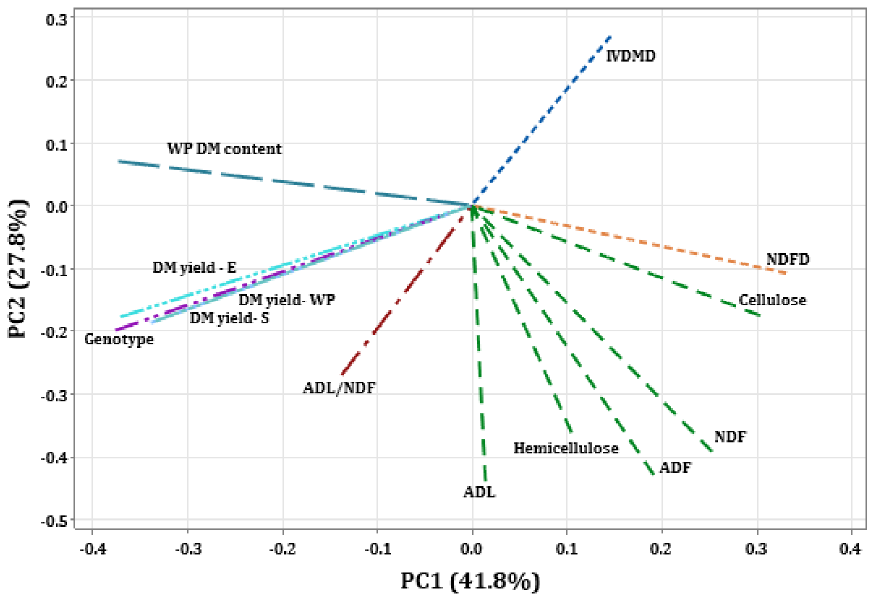


**Figure 5.** Principal components diagram of the analyzed parameters

Furthermore, individual lignocellulosic components were highly correlated, as expected. The ADL/NDF ratio was negatively correlated with the *in vitro* dry matter digestibility. Koukolová et al. (2004) found that the ADL/NDF ratio was highly negatively correlated to NDFD; however, in our study, this correlation was not significant. The dry matter yield of the stover negatively influenced *in vitro* dry matter digestibility the most, followed by the dry matter yield of the whole plant, ADF content and NDF content (Fig.

5). These findings are consistent with earlier research (Terzić et al., 2020; Nikolić et al., 2020).

## 4. Conclusions

The characteristics essential for creating high-quality silage were discovered during this two-year investigation on maize hybrids. The investigated maize hybrids' exceptional lignocellulosic fiber content, adequate dry matter digestibility, and good dry matter yield structure make them appropriate for the production of ruminant feed. The hybrid ZP 173/8 has the highest in vitro dry matter digestibility (63.87%), followed by ZP 606 (61.37%), and ZP 444 (61.00%). The results demonstrated that maize hybrids' intended applications depend on their agronomic qualities, chemical composition, and other genetically predisposed characteristics. The hybrid ZP 173/8 demonstrated its overall advantage over the other hybrids under evaluation by having the highest values of both IVDMD and NDFD. The yield components, such as the dry matter yield of the maize plant, were strongly influenced by genotype, according to the principal components. Future breeding strategies aimed at developing new, enhanced silage maize hybrids may find considerable value in our findings.

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## Declaration of competing interest

The authors have no conflict of interest to declare.

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