

Comparative analysis of SPAD, NDVI, phenological and generative parameters of maize hybrids (*Zea mays* L.)

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Abstract

Maize is a versatile multi-purpose crop that plays an essential role in the global food security. Meeting the needs of a growing population places the arable lands under the stress of intensive crop production. As the variations in climate conditions pose additional threats to maize production, the challenge focuses now on sustaining the crop yield, ensuring maximum yield and safeguarding crop protection.

The experiment was carried out in 2023 at the Látókép Plant Production Experimental Site of the University of Debrecen (Hungary). The authors aimed to monitor the changes in spectral reflectance at leaf and canopy level at several growth stages of two different maize hybrids. Another objective was to evaluate their field agronomic performance and eventually compare between their results. Thus, SPAD, NDVI and LAI indices were recorded at five phenological stages (6-leaf, 12-leaf, silking, dough, maturity) of the development of two maize hybrids. Other agronomic characteristics of maize had been measured, including plant height, cob length, stalk diameter, grain moisture content, nitrogen accumulation and grain yield.

The results demonstrated that Mv 352 hybrid had higher potential in terms of yield produced (13.64 t/ha) comparing to Fornad (12.93 t/ha), in addition to higher plant height, cob length and stalk diameter. The SPAD, NDVI and LAI recorded values showed higher values as the different growth stages developed and then decreased reaching the maturity phase. It was found that both hybrids had slight variation in their performance in this regard. However, in terms of nitrogen accumulation, Mv 352 had higher nitrogen content accumulated during the whole growing period in

comparison with Fornad, highlighting its great performance in terms of nitrogen absorption and translocation.

This study highlights the importance of monitoring SPAD, NDVI, and LAI values, as well as the plant nitrogen accumulation that provides insights into the physiological conditions of maize plants during the different growth stages, allowing for the early detection of stress factors, therefore enabling timely interventions. Additionally, this work emphasises the crucial aspect of performing these measurements for the selection and breeding of high-performing maize hybrids.

Keywords: maize hybrids, SPAD, NDVI, LAI, phenological stage, yield

Introduction

Maize is a versatile multi-purpose crop, has played a vital role in global agri-food systems since its initial recognition. In terms of production volume, maize is the current leading cereal and most widely used crop. At present, the global average yield of maize is 5.64 t/ha (*Zhang et al. 2023*), and it provides an essential source of nutrition for humans and livestock alike. Maize is considered the basis for food security in some of the poorest regions of the world, such as Africa, Asia, and Latin America, consuming around 80–90% of the maize produced (*Horváth et al. 2021*). By 2050, food demand is expected to rise by 70 percent due to the growth of the world's human population to 9 billion (*Godfray et al. 2010, Randive et al. 2021*). However, maize production faces several challenges including soil degradation, water scarcity and increasingly adverse climate events due to climate change (*Nagy 1997, 2010, 2021*). Extreme weather events are anticipated to frequently occur, with much longer and more prolonged droughts expected in the near future by 2100 (*Gombos and Nagy 2019, Buzási et al. 2021*). The vulnerability of maize to climate change has been witnessed through the decline in yield under stressed scenarios (*Khatibi et al. 2022, Shojaei et al. 2022ab*). For instance, a study done by *Li et al. (2022)* highlighted that the limited of water supply for maize growth due to increased evapotranspiration and more intense drought and heat waves can lead to a decline in yield under global warming scenarios. Another research work presented by *Zhang et al. (2022)*

pointed out the significant effects of temperature, precipitation, and sunshine hours on maize yield, indicating, therefore, the substantial impact of climate change on maize production. Due to high temperatures, the duration and rate of grain filling can be altered which in turn affect the harvested yield (*Wilhelm et al. 1999, Dupont and Altenbach 2003, Gombos and Nagy 2022*). It was reported by *Zhou et al. (2016)* that high temperatures or low radiation from silking to maturity under early sowing, and low temperatures under late sowing conditions with low radiation during grain filling decreased the post-silking plant growth rate, thereby resulting in reduced grain yield. Meanwhile, soil fertility issues are another major constraint to crop productivity that can be aggravated due to climate change. For instance, high temperatures and reduced moisture levels may lead to fewer nutrient uptake by maize plants, affecting their growth and productivity (*Szabó et al. 2022*). Nitrogen is a key element for crop production, especially for maize production (*Laskari et al. 2022*). It affects maize leaf area development and maintenance and the photosynthetic efficiency (*Muchow 1988, Muchow and Davis 1988, Srivastava et al. 2018*), as well as the dry matter production (*Guitman et al. 1991, Prystupa et al. 2004*). Given the importance of maize in global food security, fertilisers have been used in order to increase crop yield to meet the country's self-sufficiency in food production. However, the extensive use of fertilisers has had negative impact on both the environment and living organisms. Therefore, improving nitrogen management decisions for maize production has become essential to overcome environmental problems and low nutrient use efficiency (*Bojtor et al. 2021, Nagy et al. 2023*). Research studies have revealed that the influence of the rate and timing of nitrogen application through improving yield and its quality traits (*Davies et al. 2020, Illés et al. 2020, Mousavi et al. 2020, 2021; Kizilgeci et al. 2021*). Precision agricultural technology, including NDVI, SPAD and LAI measurements has been demonstrated to be efficient tools for monitoring maize yield. Normalised difference vegetative index (NDVI) is a widely used indicator for plant health and biomass and has shown a good relationship with nitrogen status in maize plant (*Rambo et al. 2010*). The Soil Plant Analysis Development (SPAD) value is a measure of chlorophyll content, most popular as it is user-friendly and non-destructive, is essential for photosynthesis and overall plant growth.

Another key parameter for assessing maize growth and development is the Leaf Area Index (LAI) that reflects the photosynthetic area of the crop. Monitoring changes in LAI values, indicate the variations in the growth stage and overall crop development.

The main aim of the present study was to assess the changes in spectral reflectance at leaf and canopy level at several growth stages of two different maize hybrids. In addition, the other objective was to evaluate the field agronomic performance of the studied maize hybrids and eventually compare the obtained results between the maize hybrids.

Materials and Methods

Description of the experimental area, design, treatments, and measurements

The experimental field trial was carried out in 2023 at the Látókép Plant Production Experiment Site of the University of Debrecen, Hungary. The experimental design consisted of one plot containing two hybrids under irrigated conditions. Fornad (FAO 420) and Mv 352 (FAO 350) were the test crops that are characterised by the difference in stress-tolerance levels. On 20th April, the seeds were sown in each row with spacing the plantings 15.6 cm apart and a total plant population of 65 000 plants per ha. The emergence of the studied hybrids occurred 13 days after sowing and they were harvested on 28th September. A drip irrigation system was installed in the experiment from 18th June and lasted until 10th August providing 374 mm of irrigation water during the growing season (12 applications in total).

Data collection was performed in conformity with the maize phenological stages classification system proposed by *Hanway* (1963) who established the developmental stages before silking based on the number of leaves. He defined subsequent stages based on grain development and divided the growing season of maize into 11 stages. During the growing season, particularly at four different phenological stages, the nitrogen content of the studied hybrids were evaluated. Thus, at the 12-leaf, silking, dough and maturity stages, four randomly selected plants were collected from each hybrid, partitioned into leaves, stalk with tassel, ear with silk, cob, kernels and husk. The plant parts were placed in paper bags, which, at first were oven-dried at 60 °C for 72 h and then weighed to obtain the dry matter

(DM) of the plants. Subsequently, they were sent to the laboratory in order to analyse their nitrogen concentration. The pretreated (dried, grinded) samples were analysed as the following: N-content was measured with Kjeldahl method according to the *MSZ-08-1783-6:1983* Hungarian standard: 1 g of the samples were measured out into N-free paper and were hot digested in $\text{H}_2\text{SO}_4 - \text{H}_2\text{O}_2$ using selenium as catalysator (VWR International). The NH_4 -content of the residue was distilled in automated distillatory equipment (VELP UDK 149) into 4 (m/m)% boric acid and was titrated with 0.2 N H_2SO_4 using TITROLINE 5000 automated titrator (SI Analytics).

Moreover, SPAD, NDVI and LAI indices were recorded at five phenological stages (6-leaf, 12-leaf, silking, dough, maturity) of the development of two maize hybrids. Other agronomic characteristics of maize were measured, including plant height, cob length, stalk diameter, grain moisture content and grain yield.

Agrometeorological characteristics

In the winter half-year preceding the 2022 growing season, the deeper soil layers could not be replenished with water, and the initial spring water availability of maize was particularly low. April was cool with average rainfall. Thereafter, the weather was warm or very warm, sunny and extremely dry practically until the second half of August. Monthly mean temperatures in all three summer months were relatively uniformly 2–3 °C above the multi-year average, with 44 heat days. In total, only 56 mm of rain fell during the summer, with only two daily rainfall events above 10 mm, which did not contribute significantly to the water supply of maize due to the very high evaporation capacity of the air. From the beginning of the summer, the soil drought severely limited water uptake and transpiration, and maize growth was very poor.

The performed research has shown that maize is most sensitive to water deficit during the phenological stages of flowering, yield formation and early crop development, starting from the period immediately preceding the tasseling. Prior to this stage, during the period of intensive vegetative development, the plant is less sensitive to rainfall deficiency. The critical month for drought damage is July, which, in addition to plant factors, is usually the warmest month, with the greatest difference between potential

evaporation and precipitation, and by which time soil moisture is usually already significantly reduced. There is a lot of research showing that high temperatures have a yield-reducing effect. This is becoming increasingly common in Hungary. Assessing the consequences is further complicated by the fact that in drought years, water shortages in the soil and high air temperatures tend to reinforce each other, and that cool, dry and very hot, wet summers are not common.

In 2023, the meteorological factors and each agrotechnical element were assessed chronologically, analysing the weather mainly in relation to the critical phenophases. During the growing season, temperature, precipitation and solar radiation conditions were assessed primarily on a monthly and then a 10-day basis. For the 10-day temperature data, the anomaly was assessed primarily, and for precipitation, the actual values were considered. Soil temperature is important during the phenological phase of maize emergence – initial development. Consequently, daily soil and air temperature data were included in the study for a two-month period between 19th April (the time of setting up the soil thermometers) and 18th June.

For a reliable analysis of crop years, it is necessary to take into account the weather parameters of the previous one or two years. In 2022, the growing season preceding 2023, there was an exceptional drought in the region and by August the water content of the soils in the deeper layers had fallen to around the level of permanently bound water. Subsequently, however, September saw near-record rainfall (152 mm), which triggered an earlier than usual and significant recharge of soil water. This favourable trend continued in the winter half-year. The total precipitation over the six months was 334 mm, 120 mm above the multi-year average (for comparison, it is worth noting that in the winter of 2021/2022 only 144 mm precipitation fell) (*Table 1*). Together with the precipitation in September, there was 486 mm precipitation, which, including some evaporation loss, was sufficient to saturate the soils to their water capacity. The mean temperature for the half-year was 5.5 °C, i.e. 1.3 °C above the multi-year average. Solar radiation conditions are considered average with 694 hours of sunshine per half-year.

Table 1. *Monthly and semi-annual patterns of temperature, precipitation and solar radiation in 2023, with deviations from 1981–2010 averages in parentheses*

Period	Mean temperature (°C)	Precipitation (mm)	Sunshine duration (hours)
Winter half-year (months X-III)	5.5 (+1.3)	334 (+120)	694 (+22)
Summer half-year (months IV-IX)	18.4 (+0.9)	336 (-10)	1512 (+28)
April	9.3 (-1.9)	48 (-5)	151 (-64)
May	16.5 (-0.1)	33 (-31)	247 (+3)
June	19.5 (+0.2)	82 (+16)	275 (+13)
July	22.5 (+1.2)	46 (-20)	306 (+28)
August	22.8 (+2.0)	86 (+37)	286 (+4)
September	20.0 (+3.8)	41 (-7)	247 (+44)

In June, the weather continued to be free of extreme events, with even, moderately warm temperatures, in line with the multi-year average. Precipitation patterns were favourable, with most of the 82 mm monthly total falling in the first and last thirds. The number of sunny hours (275 hours) was slightly above average. The vegetative development of maize continued this month under favourable meteorological conditions. The average temperature in July (22.5 °C) is higher than usual, but 1–2 °C below the average temperature of the hottest months of recent summers. The month was characterised by few clouds and a lot of sunshine (306 hours). The middle of the month was the warmest, but there were no extreme heatwaves, with temperatures reaching 34–35 °C on only two days. By the last third of July, temperatures had returned to multi-year averages. The water demand of maize is highest during the initial period of silking and yield development. This phenological period has again took place mainly in July. Monthly rainfall of 46 mm (20 mm below average) was not sufficient to ensure optimal water availability, but no significant water stress developed in the region's soils which have favourable water management.

In 2023, the weather conditions for the growing season were also generally favourable. It should be stressed that the dominant periods were characterised by

moderately warm, evenly distributed temperatures, with no periods of extreme heat or cold. Precipitation during the growing season was around average. However, within this period, the amount of precipitation during May and July was only half of the usual values. It has to be noted that there was no significant stress due to water shortages as a result of the water stored in the deeper layers of the soil and the favourable temperature conditions.

Results

During the cropping season of 2023, Mv 352 produced the highest yield of 13.64 t/ha, 70 kg/ha higher than that obtained by the Fornad hybrid (*Table 2*). The weather conditions in 2023 did not witness any extreme events of heat or cold and were favourable for maize production. Despite the studied hybrids being grown under the same weather conditions with the same soil characteristics, it can be observed that overall, Mv 352 expressed better agronomic physiologic and vegetative performances than Fornad. For instance, the highest plant height (326.6 cm), cob length (156.1 cm) and LAI (4.5) values were obtained by Mv 352. At the same time, Fornad produced strong stalks with a 0.5 mm larger diameter than that of Mv 352 (19.81). The difference in maize performances could be explained by the difference in the genetic behavior of the examined maize hybrids.

Table 2. *Agronomic characteristics of maize*

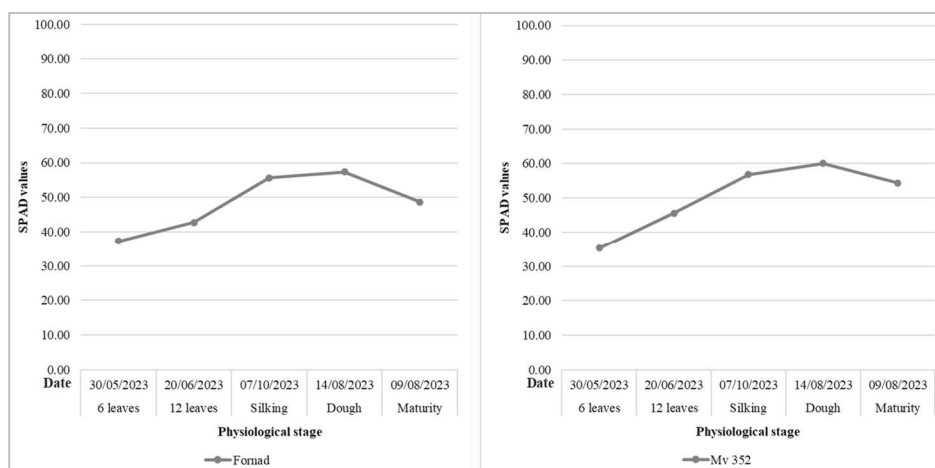
	Plant height (cm)	Cob length (cm)	Stalk diameter (mm)	LAI	Yield (t/ha)
Fornad	313.9	127.4	20.35	4.4	12.93
Mv 352	326.6	156.1	19.81	4.5	13.64

SPAD

During the growing season, the SPAD values varied from 37.30 to 57.40 and from 35.30 to 60 for Fornad and Mv 352 hybrid (*Figure 1*). Based on the obtained results, it can be noticed that the trend of SPAD index for Fornad is similar to the one of Mv 352 hybrid with slight variation before the silking

stage. At the 6-leaf stage, the recorded SPAD value of the Fornad hybrid was higher than that of Mv 352 by two SPAD values. The highest observed SPAD values were 57.40 and 60 corresponding to Fornad and Mv 352, respectively and both were recorded at the dough stage. Following this phase and progressing towards the maturity phase, a decrease can be observed in SPAD readings for both hybrids. Based on the obtained results, post-silking and grain filling phases detected higher SPAD values compared to the vegetative phase.

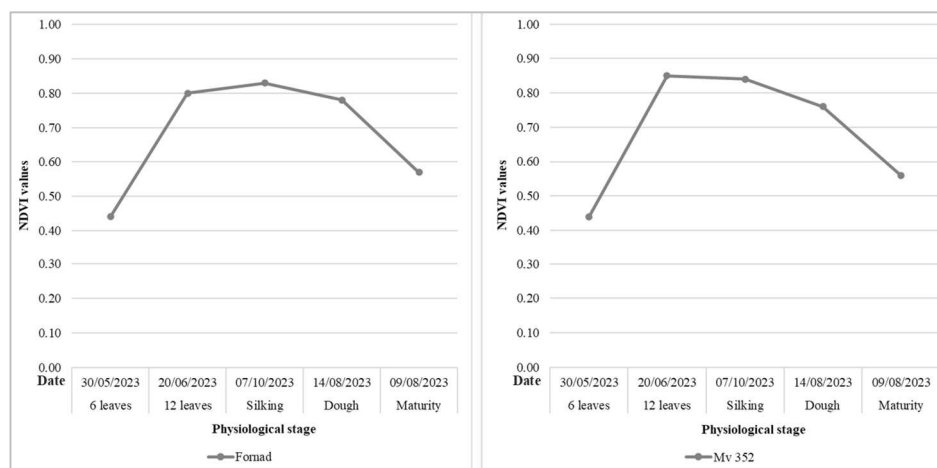
Figure 1. *SPAD values of Fornad and Mv 352 during the different growth stages*



NDVI

The recorded NDVI values during the growing season showed similar trends for both maize hybrids with slight variation between the 12-leaf stage and the dough stage (*Figure 2*). The NDVI values varied from 0.44 to 0.83 and from 0.44 to 0.85 for Fornad and Mv 352 hybrid. At the 6-leaf stage, the recorded NDVI value was 0.44 for both studied hybrids. The highest NDVI value observed was 0.83 by Fornad and recorded at the silking stage, whereas it was 0.85 verified at the 12-leaf stage by Mv 352. Between the silking and the maturity stage, a decrease in NDVI reading for both hybrids can be observed in the graph below. According to the obtained results, the highest NDVI values were detected during the 12-leaf and silking stage.

Figure 2. NDVI values of Fornad and Mv 352 during the different growth stages



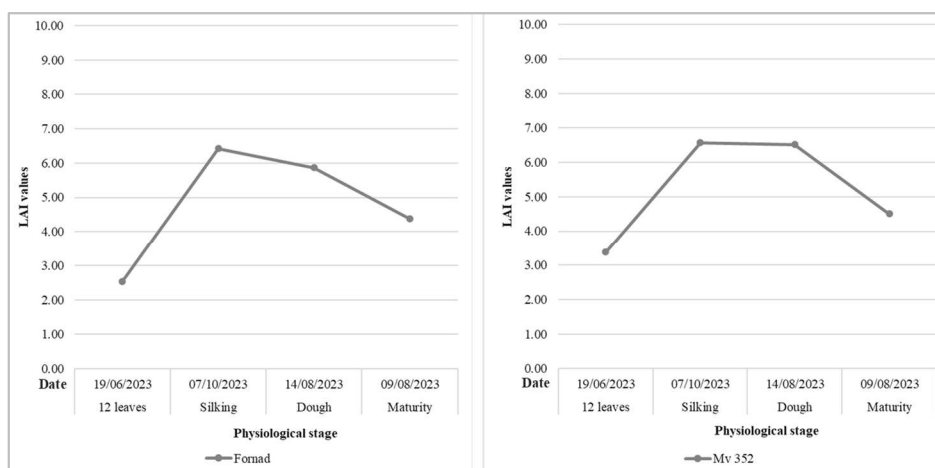
LAI

A variation of the LAI trends can be observed in relation to both hybrids, varying from 2.53 to 6.42 and from 3.38 to 6.57 for Fornad and Mv 352 (Figure 3). At the 6-leaf stage, the recorded LAI value of Mv 352 hybrid was slightly higher than that of Fornad. The highest LAI values observed during the growing season were 6.42 and 6.57 corresponding to Fornad and Mv 352, respectively, registered at the silking stage. Between silking and the maturity phase, a drop in LAI values can be observed regarding the Fornad hybrid, however, the decrease in LAI values registered in Mv 352 was progressive.

Grain moisture content and dry matter

The plant's life cycle consists of three primary stages that include the vegetative phase, the reproductive phase and grain filling phase. Thus, with the progress in plant growth, the dry matter accumulation increases to reach its maximum value at the silking stage, before shifting to the reproductive organs such as the ear, tassel and silks. During the grain filling phase, the dry matter accumulation in kernels occurs reaching its maximum weight at physiological maturity.

Figure 3. LAI values of Fornad and Mv 352 during the different growth stages

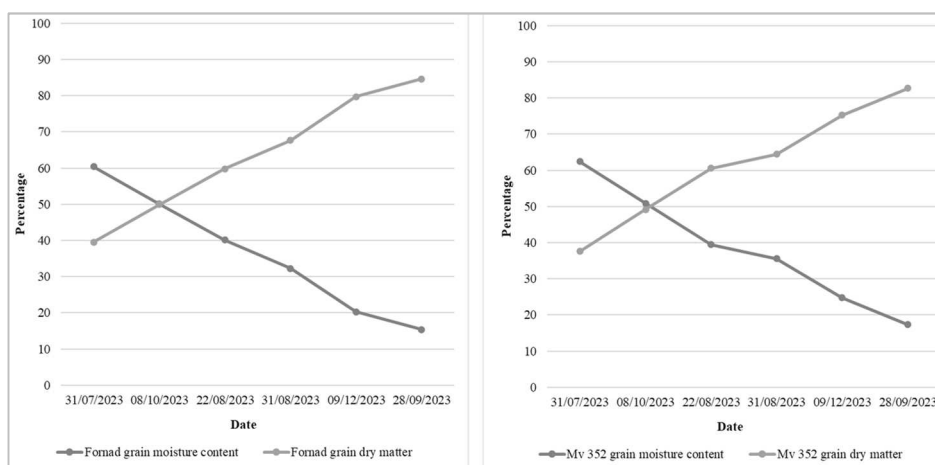


According to the obtained results, the silking stage has been reached on the 10th of July, therefore, the post-silking and grain filling phases occurred and the plant started to store nutrients and carbohydrates in the kernels (*Figure 4*). As the accumulation of dry matter in the kernels increases, grain moisture decreases. From post-silking stage until the maturity stage (3rd of September), the rate of the moisture loss in Fornad grains was 0.91%/day which is the equivalent of 14.42 heat unit (HU) per 1%, while it was 0.56%/day which is the equivalent of 18.82 HU per 1% after the maturity stage. Regarding Mv 352, the rate of grain dehydration was 0.97%/day which is the equivalent of 13.65 HU per 1% from post-silking stage until the maturity stage. However, the rate continued to decrease after the maturity stage with 0.49%/day, the equivalent of 21.46 HU per 1%.

Nitrogen accumulation

From the early stage of maize development, the demand for nitrogen was relatively small and increased in the subsequent stages. It can be noticed that Mv 352 had higher nitrogen content accumulated during the whole growing period in comparison with Fornad, highlighting its great performance in terms of N absorption and translocation.

Figure 4. Maize grain moisture content and dry matter

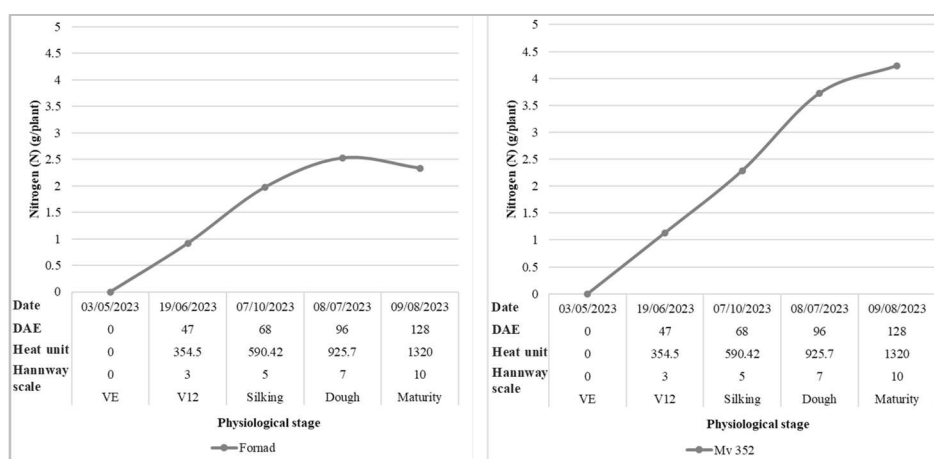


Maize leaves had a higher N demand compared to other organs until reaching the dough stage when the N mobilisation into the kernels occurred and the highest N contents were reached, accumulating respectively 1 g/plant and 1.80 g/plant for Fornad and Mv 352. Up to V12, Fornad accumulated 0.91 g/plant of nitrogen with a rate of 0.019 g/plant/day, while Mv 352 accumulated a bit higher amount of nitrogen of 1.13 g/plant with a rate of 0.024 g/plant/day (Figure 5). Subsequently, the N absorption mainly by the plant vegetative organs increased significantly where the leaves accumulated the highest nitrogen contents at the silking stage, of 0.95 g/plant and 1.20 g/plant for Fornad and Mv 352 respectively. The stalk was the second to accumulate a higher concentration of nitrogen at this stage, with 0.74 g/plant and 0.88 g/plant accumulated respectively by Fornad and Mv 352. At the dough stage, as the ear development increased, the N mobilisation switched to the grains, cob, and husk. Meanwhile, the accumulation of nitrogen by leaves and stalk progressively decreased with a rate of 0.05 g/plant/day and 0.6 g/plant/day for leaves, 0.04 g/plant/day and 0.04 g/plant/day for the stalk, regarding Fornad and Mv 352 hybrids.

At the end of the cycle, the nitrogen accumulation by Fornad, in the kernels, leaves, stalk, husk, and cob was 1.61 g/plant, 0.35 g/plant, 0.24 g/plant, 0.08 g/plant, 0.06 g/plant, respectively, totaling 2.33 g/plant. As for

Mv 352, the accumulation of nitrogen content in the kernels, leaves, stalk, husk, and cob was 2.68 g/plant, 0.73 g/plant, 0.65 g/plant, 0.09 g/plant, 0.08 g/plant, respectively, totaling 4.23 g/plant. The accumulation rate of nitrogen at this phase was 0.07 g/plant/day for Fornad, when it was 0.13 g/plant/day for Mv 352.

Figure 5. Nitrogen uptake by both maize hybrids as a function of phenological stages



Discussion

Given the difference in yield between the examined hybrids despite the favourable weather and soil conditions for crop development, this suggests that the genetic variation between the two hybrids can result in a large yield difference. For instance, in terms of agronomic physiology and vegetative growth, overall, Mv 352 outperformed Fornad. For decades several studies have addressed the importance of maize breeding topic for yield improvement along with enhancing its genetic resistance to biotic and biotic stresses (*Hake and Ross-Ibarra 2015, Fadhli et al. 2020, Hossain et al. 2022*), considering that combining the developed new varieties with appropriate cultivation technology is crucial step for achieving optimal yield. Grain moisture is another important element for assessing maize quality since it influences the physical characteristics of the kernels, their susceptibility and

deterioration, storage and stability. It was advised of harvesting the grains when reaching around 23% and 24% of moisture, and they are considered suitable for storage when their moisture is less than 23% (Mousavi et al. 2018). In the present study, the grain moisture content of Mv 352 reached 24.27% at R4, suggesting that it might be the proper time of harvesting, while Fornad grains were mostly suitable for storage at that time (20.26%). The cultivation, growth, and yield of maize are influenced by numerous agricultural practices alongside weather conditions, with fertilisation being a key factor. Maize heavily relies on nitrogen as it is the key element for photosynthesis, enhancing leaf pigmentation, and contributing substantially to grain yield (Asare et al. 2023, PDA 2024). Exposure to nutrient deficiency may hinder the crop from proper development. Therefore, ensuring adequate and enough nourishment particularly in the early stages of vegetative growth could increase production. Studies have shown correlations between SPAD, NDVI and LAI readings and plant nitrogen content highlighting the usefulness of on site precise and effective assessment of nitrogen in maize (Din et al. 2017, Yang et al. 2017).

Conclusion

This study concluded that it is essential to obtain insights into maize growth and development through the use of precision agricultural technologies that allow for efficient and precise assessment, contributing to better-informed decision-making in crop production. The obtained results demonstrated that Mv 352 hybrid had better potential in terms of agronomic performance and in the ability of N absorption and translocation, which eventually resulted in a higher yield, compared to the Fornad hybrid.

Acknowledgments

Project no. TKP2021-NKTA-32 has been implemented with the support provided by the Ministry of Culture and Innovation of Hungary from the National Research, Development and Innovation Fund, financed under the TKP2021-NKTA funding scheme. This paper was supported by the János Bolyai Research Scholarship (BO/00068/23/4) of the Hungarian Academy of Sciences.

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