60 Years of Hungarian Hybrid Maize

1953–2013

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Zoltán Bedő[1], Beáta Barnabás[2]

Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Science
[1]Cereal Breeding Department,
[2]Plant Cell Biology Department,
2462 Martonvásár, Brunszvik u. 2.
e-mail: zoltan.bedo@agrar.mta.hu

Over the six decades that have expired since the first hybrid maize was born, the research concept in Martonvásár plant breeding has changed on a number of occasions in line with the priorities set in Hungarian agriculture and in international agrarian research. Sixty years ago an increase in cereal yields was the principal aim, as Hungary was forced to import cereals for nearly two decades after the Second World War. The first modification in the Martonvásár research strategy was linked with the construction of the phytotron, which provided excellent facilities for basic research connected with plant breeding and for the analysis of adaptability. The next alteration in the concept behind the scientific research was made possible by the change of regime and the reform of the Hungarian Academy of Sciences in the first half of the 1990s. It has now become clear that, in addition to increasing yields, the world's agricultural scientists need to pay far greater attention to improving yield stability, the quality of the processing industry and the sustainability of production.

The reputation of the Martonvásár research institute was established 60 years ago on the basis of the results achieved in maize breeding. It was here that Endre Pap developed the first inbred maize hybrid in Europe, registered in 1953 under the name Mv 5. During the last 60 years over a hundred registered hybrids have been bred in Martonvásár, with a further fifty or more in cooperation with other breeding units, the total sowing area of which exceeded 20 million hectares in Hungary. Martonvásár was also the first to elaborate a field technology for the production of hybrid maize and its processing in seed plants. The testing basis was broadened to improve the efficiency of the breeding programme, and the whole breeding process was mechanised, from sowing through harvesting to data processing. New basic breeding materials were developed and the frequency of agronomically favourable genes was increased by several cycles of recurrent selection. A protocol for the analysis of polymorphism in the maize inbred lines bred in Martonvásár was elaborated on the basis of morphological descriptions, isoenzyme patterns and DNA-based methods. In order to speed up the breeding process, a winter nursery was established in the southern hemisphere in Chile 22 years ago, thus reducing the time required for line development by half. In addition to fodder maize hybrids, hybrids with high starch content, ideal for fermentation, were developed for the purpose of bioethanol production. The Martonvásár maize breeding team was the first in Europe to breed high-yielding leafy (Lfy) hybrids, which have a new type of plant architecture, involving a greater leaf number above the ear, and excellent digestibility, making them ideal for silage production.

The breeding programme has always been characterised by wide-ranging international cooperation, which promoted both the breeding of hybrids for use in Hungary and the development of hybrids intended for export. In this framework, over 50 hybrids were granted state registration in many countries in Europe. Hybrids bred in Martonvásár alone or in cooperation are now grown on more than 12 million ha outside Hungary.

Nature never stands still: plants need to adapt themselves to the environment throughout their whole life cycle, from emergence to the harvesting of the yield. Five generations of maize breeders in Martonvásár have also adapted to this constant change, from Endre Pap through breeding teams headed by István Kovács, Tamás Szundy and Csaba Marton to the young PhD students working in the department today, with the excellent support of technicians, field and laboratory staff. Congratulations are due to them all for 60 years of excellent results, which make up one of the most memorable chapters in the history of Hungarian plant breeding.

Hybrid maize in Hungary is 60 years old

L. Csaba Marton

Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
2462 Martonvásár, Brunszvik u. 2.
e-mail: martoncs@mail.mgki.hu

Abstract

On the 60th anniversary of the birth of Mv 5, the first hybrid maize developed in Europe, we honour the memory of both Endre Pap, the breeder of Mv 5, and the establishment of hybrid maize breeding and seed production in Hungary. The birth of Hungarian hybrid maize was the life work of Endre Pap, and is recognised throughout the world as the greatest achievement of Hungarian plant breeding.

The development of the maize hybrid Martonvásári 5 gave an enormous boost to the research institute, founded only a few years earlier. For several decades Hungarian maize breeders dominated their field, achieving one success after another in terms of both basic science and practical results. In addition to breeding, the agronomic research required to ensure the success of maize production also reached a high standard. Martonvásár was the first to elaborate and introduce into Hungary the field technology required for the production of hybrid maize seed and its processing in seed plants.

These results, important in the history of science, happily coincided with the demand for modernisation in Hungarian agriculture, so within a few years the whole of the national maize-growing area was sown with hybrids from Martonvásár.

Sixty years after the registration of the Martonvásári 5 hybrid, the institute's maize breeders still occupy first place among Hungarian breeders as regards their share of the Hungarian market, where they face stiff international competition, and are ranked 3rd or 4th in comparison with multinational companies.

Introduction

Hybrid maize breeding in America can look back on a history of around 100 years (*Shull* 1909, 1910; *East* 1909). The first hybrid maize seed was sold in the USA for a dollar a pound in 1924 (*Troyer* 1995). The history of hybrid maize breeding in Hungary, and also in Europe as a whole, began with the registration of Mv 5 in 1953. From 1964 onwards, 100% of the maize-growing area in Hungary was occupied by hybrid maize (*Berko and Horváth* 1993).

Thanks to the use of hybrids, there was an enormous increase in yield averages throughout the world, and this tendency is still continuing. Various hypotheses were raised to explain the phenomenon of heterosis, but the real reasons are still not known. The process has been analysed in experiments by numerous scientists – breeders, geneticists, crop producers and physiologists, and analysis at the molecular level is now underway in many laboratories.

Analysis at the plant (phenotype) level did not answer the question of how heterosis works, but it did provide a great deal of useful information that helped improve the methodology of breeding programmes.

On the basis of experimental data, *Frey* (1971) demonstrated that the first hybrids yielded 7–11% more than open-pollinated varieties, while compared to the first hybrids the surplus yield achieved by those developed in the 1960s was 49%.

In experiments performed by *Russell* (1984) the yield of hybrids developed in the 1980s was 66.4% (4.21 t/ha) higher than that of open-pollinated varieties and 27.5% (2.28 t/ha) higher than that produced by hybrids from the 1930s.

In another experiment, *Russell* (1985a) proved that if the plant density was increased the superiority of hybrids compared with open-pollinated varieties was even greater.

Duvick (1977) established that the increase in the yield potential of hybrids amounted to 50–53 kg/ha/year. These values represented 57–60% of the increase in average yields in Iowa. In a later experiment *Duvick* (1984) estimated the increase in yield potential to be 73–92 kg/ha/year, while in similar investigations *Castleberry et al.* (1984) reported a yield increase of 82 kg/ha/year, equivalent to 75% of the increase in the US yield average.

Heterosis breeding began in the 1930s in Hungary, and Hungarian breeders were the first to produce variety hybrids, which yielded 10–15% more than varieties (*Szundy and Kovács* 1991).

Variety hybrids did not achieve wide popularity because of the appearance of inbred hybrids, which yielded 10–15% more than the variety hybrids (*Kovács and Szundy* 1991).

Győrffy (1976) evaluated the effect of production factors on the increase in maize yield averages in a polyfactorial experiment and found that the most important yield-enhancing factor was the nutrient supply, followed by the variety, which made a 26% contribution to the yield increase. Based on the data of the same experiment, *Berzsenyi and Győrffy* (1995) later estimated the role of the variety to be 30%. Both estimates are considerably lower than those reported in the American literature. *Marton et al.* (1997) suggested that the reason for this discrepancy was the different rate of increase in national yield averages. While the rate of increase amounted to 100 kg/ha/year during the given period in the USA, this value was considerably more than 100 kg/ha/year in Hungary, even reaching a figure of 200 kg/ha/year over a short period. According to *Troyer* (1995), American agriculture achieved a surplus income of 300 million US$ a year as the result of maize breeding.

The birth of Martonvásári 5 (Mv 5)

The development of the maize hybrid Martonvásári 5 (Mv 5) was primarily the intellectual achievement of *Endre Pap* from Mindszentpuszta. The hybrid maize known as Martonvásári 5 (Mv 5) was registered by the Variety Registration Council on 16 December 1953. Mv 5 was the first hybrid maize developed by crossing inbred lines, not only in Martonvásár and in Hungary, but in the whole of Europe.

For the sake of historical accuracy it should be noted that hybrid maize had been developed in Hungary by crossing varieties at an earlier date. *Rudolf Fleischmann* developed 12 variety hybrids in 1933, while *László Berzsenyi-Janosits* developed 171 in

1948. As the result of trials, four variety hybrids were granted state registration in 1953 (Óvári 1, Óvári 3, Óvári 4, Óvári 5).

The seed of these variety hybrids was produced on more than 10,000 hectares by 1957, which would have been enough to plant the whole maize-growing area of the country, but variety hybrids never got a foothold in commercial production, because Mv 5 was registered in the same year (1953) as the variety hybrids. While the variety hybrids yielded 10–15% more than the varieties, Mv 5, like inbred hybrids in general, was capable of achieving a 20–30% yield surplus. The trials carried out in 1953 therefore decided not only the fate of Mv 5, but also the outcome of the competition between varieties, variety hybrids and inbred hybrids, in favour of inbred hybrids.

The performance of the Mv 5 hybrid in the 1952–1953 national variety trials was characterised by *Herbert Taróczi* as follows:

> *"It was not by chance that our best hybrid at present, Martonvásári 5, was classified in the top group at all the 17 plant variety testing stations in the country, without exception, and was ranked first in 16 cases (94%). A similar case has never yet been reported in the history of maize breeding and experimentation, either in Hungary or abroad. This hybrid proved to be unbeatable in the most diverse regions and soils."*

Jánossy et al. (1957) gave a detailed description of the Martonvásári 5 hybrid, noting that it was granted state registration "*on the basis of its substantially greater yield potential compared with the other maize varieties*" and the fact that "*due to its good adaptability and mid-early ripening it could be sown anywhere in the country.*" Martonvásári 5 performed outstandingly not only in Hungary, but also in state trials in Austria.

The Martonvásári 5 hybrid occupied 1% of the national maize sowing area in 1956, 28% in 1959 and 56% in 1960, after which its sowing area gradually declined and it was no longer grown commercially by the early 1970s.

This unexpected, virtually 'ready-made' success provided an enormous boost for the young institute, founded only a few years earlier. The scientific abilities and responsible thinking of the research staff is illustrated by the fact that after Endre Pap left the country in 1956, maize breeding continued without a break and delivered one success after another over the next few decades, playing a dominant role both in scientific life and in the field of practical results.

From the 1980s onwards work was carried out for more than ten years on the introduction and popularisation of foreign hybrids selected in the framework of a special project set up for this purpose. During this period more than 40 of the hybrids introduced in this way were granted state registration. In the meantime, a large proportion of the available resources were dedicated to modernising and renewing the Martonvásár maize breeding programme.

Breeding aims from the 1980's onwards

The priorities in the new programme were chosen on the basis of the intellectual and technical infrastructure inherited from our predecessors, the lessons learnt from the 1970s, and on adaptable components from competitive foreign programmes. Special emphasis was placed on the avoidance of line monocultures as a protective measure against genetic vulnerability and with the aim of breeding strong-stalked hybrids with rapid drying down, and also in order to conform with the new legal and economic situation with regard to the patenting of breeding materials.

The priorities in the present breeding programme were determined according to the altered requirements facing maize production and maize utilisation. Attention was also given to the ecological conditions in Hungary. The increasingly frequent occurrence of climatic extremes and the very varied agronomic standard of maize production justified improvements in the adaptability of the hybrids, including resistance to both abiotic (cold, drought) and biotic stress factors.

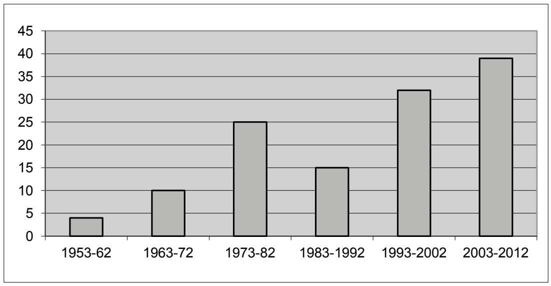

Fig. 1. State registered Mv hybrids (1953–2012)

The hybrid testing facilities were broadened, with an almost five times increase in the number of experimental plots. Hybrids from Martonvásár are tested in comparative trials at around 100 foreign locations. In addition, the hybrids are evaluated in systematic agronomic experiments.

Research on chilling tolerance has always been part of the *60* years of maize breeding in Martonvásár (*Kovács* 1958; *Herczegh* 1978; *Szundy* 1981; *Marton* 1991). Chilling tolerance is of special importance for the early (FAO 200–240) and extra-early (FAO 150–190) hybrids grown for silage or grain purposes on areas north of latitude 55° (*Pintér* 1994). The improvement of resistance to diseases and pests is an integral part of the breeding programme (*Kizmus and Marton* 1986; *Szőke et al.* 2009). The resistance of large numbers of breeding materials (populations, segregating material, lines and hybrids) to common smut, head smut, Fusarium stalk rot and ear rot is examined every year. In recent years the evaluation of infection with Fusarium ear rot has been commenced in various growing areas of the country. In addition to the level of infection, the toxin content of mouldy ears is also analysed. Healthy food and fodder can only be produced from toxin-free maize.

The corn rootworm has been present in Hungary since 1995. In the years since its appearance it has spread throughout the country. Particularly heavy losses are suffered by farmers in regions where maize is grown in a monoculture.

There are two paths available to maize breeders attempting to improve the resistance of hybrids to corn rootworm (*Marton et al.* 2009). One is the use of conventional methods to increase the tolerance of the hybrids to larval damage. The other is the development of transgenic maize.

In studies on the ripening dynamics of maize, knowledge on the nature of ripening and drying down has been obtained for the most important lines and sources, and successful selection has led to the development of new inbred lines and hybrids with rapid drying down (*Hadi* 1982; *Pók* 2002). In recent years the labelling of the breeding materials with genetic markers has been commenced (*Nagy et al.* 1999). By simultaneously evaluating various types of markers (isoenzymes, PCR, microsatellites) the genetic background and breeding value of unknown breeding materials can be detected with great precision (*Nagy et al.* 2003). The digestibility of the hybrids is tested with the aim of improving the internal quality of silage maize (*Zsubori et al.* 2002). An extensive population improvement programme was set up to broaden the genetic basis (*Herczegh et al.* 1986). The principal aim of seed biological analysis is to elaborate a technology ensuring high quality seed under stress conditions in Hungary (*Berzy et al.* 2003).

The time required to develop new lines has been reduced to half by establishing a winter nursery in the southern hemisphere, first in Argentina and currently in Chile. The broadening of the genetic basis is also served indirectly by the joint breeding programmes set up with 20 foreign institutes and companies, in the framework of which the partners have free access to each other's breeding lines.

The correct choice of aims is proved by the fact that more and more Martonvásár maize hybrids have been state registered in recent years and the number of hybrids included in the national list at the same time has never been as high as it is now (*Fig. 1*). Over the last 10 years 39 new hybrids have been granted state registration. Of these, 6 belong to the extra-early group, 21 to the early, 8 to the mid-season and 4 to the late group. The majority of the hybrids are grain hybrids (32), while seven have been registered for silage utilisation and one is a sweetcorn. In terms of the commercial growing area occupied by Mv hybrids, Martonvásár ranks 3rd or 4th among all the market participants, and 1st among the Hungarian variety owners. The quantity of Martonvásár hybrid maize seed marketed abroad exceeds that sold in Hungary. Over the last few decades, Martonvásár hybrids have been registered in Russia, Ukraine, Croatia, Romania, Bulgaria, Turkey and Iran.

Seed production

Martonvásár was the first to elaborate or introduce field technologies for hybrid maize seed production and for its processing in seed plants.

The results achieved in maize breeding, which made their mark in the history of science, happily coincided with the demand for modernisation in Hungarian agriculture, so within a few years almost the whole of the national maize-growing area was sown with hybrids from Martonvásár. The spread of hybrids and the complete dominance of hybrids in maize production took place in a fifth of the time required in the 'land of opportunity', the innovative and market-oriented USA (*Fig. 2*). The yield-enhancing effect of these hybrids can be expressed in millions of tonnes on a national scale. The average yield in the five years before the spread of hybrids was 2.15 t/ha, which rose to 2.97 t/ha for the first five years after hybrids occupied 100% of the growing area. This increase (38%) could be attributed in great part of the hybrids, though the improvements in the technology also played a role in the rise in average yields.

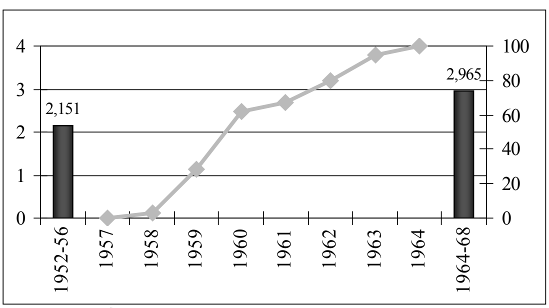

Fig. 2. National maize yield averages in the five years before and after the spread of hybrids

The hybrid programme adopted in 1954 promoted the spread of the hybrids. In summer 1956 Martonvásár was given the first diesel-fired *Campbell* grain dryer fitted with an automatic thermostat and in the same year the first six-chambered maize seed dryer in the country was built. From 1957 onwards the research institute set up a team specialising in hybrid maize seed production and took the production of basic hybrid maize seed into its own hands. The first six hybrid maize seed plants in state farms were built in 1958 (*Baja, Boly, Mezőhegyes, Mezőnagymihály, Debrecen, Murony*). Later, between 1959 and 1964, further seed plants were constructed in Mezőfalva, Dalmand, Mosonmagyaróvár, Szenttamás, Hódmezővásárhely and Cegléd, providing the foundations of the Hungarian hybrid maize seed industry, the capacity of which, taking into consideration the 65-day working season, reached a figure of 36,000 t a year.

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Research promoting the sustainability of maize production in Martonvásár

Tamás Árendás, Zoltán Berzsenyi, Péter Bónis, Györgyi Micskei*, L. Csaba Marton

Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
2462 Martonvásár, Brunszvik u. 2.
e-mail: arendas.tamas@agrar.mta.hu

Abstract

Agronomic research began in Martonvásár after the first inbred maize hybrid, Mv 5, entered commercial production in the mid-1950s and took the form of up-to-date long-term experiments and small-plot field experiments on technological adaptation. Thanks to their suitability, a considerable proportion of these experiments still continue to serve the sustainable development of maize production. In the initial stages of the research the aim was to obtain knowledge on factors related to an increase in the intensity of field crop production and to reducing the need for manual labour. Nowadays the long years of data, phenological observations, ecological and eco-physiological measurements promote the use of models and are increasingly linked to the precision techniques made possible by intensive technological development.

Introduction

Maize breeding and crop production research have been inseparably linked in Martonvásár from the very beginning. The results of the polyfactorial experiment set up in autumn 1959 very quickly proved that the genotype was one of the most decisive factors influencing yield enhancement in maize. If the genetic potential latent in the hybrids was to be manifested, however, it became essential to examine and precisely quantify the correlations and interactions existing between individual production factors.

The aim expressed by Béla Győrffy almost half a century ago is still valid today:

> *"When making recommendations on crop production, we should not be looking for the very best solution, but should be presenting a number of possible good solutions on the basis of the experimental data."*

Materials and methods

Maize research in the Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences is based on many decades of long-term experiments and technological adaptation trials, which provide a unique opportunity to examine the agronomic responses of maize hybrids.

The long-term experiments set up by *Béla Győrffy* in Martonvásár at the end of the 1950s, which are among the oldest in the country, still completely satisfy the methodological criteria raised for such experiments today. The long-term experiments consistently maintained in Martonvásár, which are known on an international scale, consist of over 700 plots on an area of around 15 hectares. These long-term experiments represent living field laboratories. Changes in soil fertility, the efficiency of crop production systems and the factors that influence them can only be adequately investigated with the help of data series obtained under such conditions.

In these bi- and multi-factorial technological experiments, studies are made on the agronomic responses of maize, and on interactions between the genotype, the production technology and the variable environmental factors. The results serve as the basis for continuous improvements in maize production, which are directly exploited in practice.

Results and discussion

Crop rotation experiments

One of the basic questions raised during the early years of intensive development in Hungarian maize production was whether the success of cereal production could be enhanced by increasing the ratio of monocultures. Right from the start, the results of the crop rotation vs. monoculture experiment made it clear that alfalfa was a worse forecrop for maize than winter wheat and that under Hungarian conditions a maize-winter wheat diculture was competitive with a maize monoculture even in terms of total yield (*Győrffy* 1975).

Comparative analysis based on longer series of data confirmed that the yield of maize in a monoculture, even before the spread of the corn rootworm, was always lower than in a crop rotation, and that the yield-enhancing effect of rotation was inversely proportional to the ratio of maize in the crop sequence. Averaged over the fertiliser treatments, the yield-enhancing effect was greatest in a Norfolk rotation (peas-winter wheat-maize-spring barley) (NF: 0.904 t ha^{-1}). This was followed in decreasing order by a maize-wheat-alfalfa triculture (MWA: 0.853 t ha^{-1}), a maize-wheat diculture (MW: 0.490 t ha^{-1}) and a maize-alfalfa sequence (MA: 0.376 t ha^{-1}). The yield-enhancing effect of a crop rotation was significantly greater without mineral fertilisation than on fertilised plots (MW: 0.15, MA: 1.254, MWA: 1.401, NF: 1.357 t ha^{-1}). In maize crop rotations, fertilisation reduced the rotation effect by almost 50% (*Berzsenyi et al.* 2000).

Mineral fertilisation experiments

In the early stages, both in Martonvásár and elsewhere, this research was aimed at discovering whether mineral fertilisers, either in combination with farmyard manure or alone, were able to increase the yield of maize and maintain the fertility of the soil (*Győrffy* 1958).

Averaged over 44 years of a long-term fertilisation experiment on the effect of macroelement combinations (*Árendás et al.* 2010), the greatest, significant yield increment in maize was achieved in Martonvásár with N fertiliser (with wheat as forecrop: N vs. 0: 0.87, NP vs. P: 0.75, NPK vs. PK: 1.09 t ha^{-1}; with maize as forecrop: 1.64, 1.60 and

1.91 t ha^{-1}). In dry years nitrogen only had a positive effect on the yield of maize sown after the phosphorus-demanding wheat crop when combined with PK fertilisers. In wet years there was no significant difference between the maize yields achieved on plots poorly or well supplied with phosphorus, but in the case of drought, better P supplies had a yield-increasing effect.

In fertiliser dose experiments, N fertilisers had a significant influence on the grain yield in a monoculture. Averaged over the years 1970-2002 the maize yields (t ha^{-1}) in various N treatments were as follows: N_0: 3.69; N_{80}: 6.97; N_{160}: 8.33; N_{240}: 8.34. The yield stability was greatest for the 160 kg ha^{-1} N rate. As regards the year effect, the yield increase (t ha^{-1}) at each N rate in wet years, compared with dry years, was as follows: N_0: 1.29; N_{80}: 1.96; N_{160}: 1.87; N_{240}: 1.65. The correlation between kernel number and grain yield was close at low N rates (stress environment) and loose for favourable N supplies. The opposite trend was observed between the grain yield and the thousand-kernel weight (*Berzsenyi* 2009).

The yield level was higher in a crop rotation, where smaller N fertiliser doses were required for maximum yield. A comparison of the yields achieved with the same hybrids in monoculture and crop rotation between 1995 and 2006 gave the following results (t ha^{-1}): in monoculture: N_0: 3.876; N_{80}: 7.128; N_{160}: 8.463; N_{240}: 8.556; in crop rotation: N_0: 8.288; N_{80}: 9.556; N_{160}: 9.561; N_{240}: 9.422. Irrespective of the year, the greatest yield in the crop rotation was recorded for N doses of 80 and 120 kg ha^{-1}.

Plant density experiments

Simultaneously with the appearance of inbred maize hybrids a national experimental network was set up, coordinated by the Martonvásár institute, to investigate whether the varieties and hybrids in commercial production could be sown more densely (*I'só* 1958). Thanks to the results of these experiments and those set up in Martonvásár with the methodology used by Béla Győrffy, optimum plant density values rose from the 20.000 plants ha^{-1} recommended in the 1940s (70 × 70 cm) to 35-40 thousand in the 1950s, to around 50 in the following decade and to 55-60 thousand plants ha^{-1} in the 1970s (*Győrffy* 1979). Experiments on plant density also investigated the importance of the shape of the area available to the plant, supplemented by measurements on the water and nutrient regime (*Győrffy* 1958).

The results of experiments performed over the last three decades on maize hybrids bred in Martonvásár prove that the relative water and nutrient deficits caused by excessive plant densities have an unfavourable effect on the quantity and stability of the yield. Yield losses can be attributed to an increase in variability per plant and to a higher ratio of barren plants. When the results of 22 years were analysed using the regression method of stability analysis, the yield was found to be the most stable at a plant density of 60 000 plants ha^{-1} under non-irrigated conditions, based on the yield responses of the maize hybrids. At an environmental mean of less than 4.6 t ha^{-1}, a plant density of 40 000 plants ha^{-1} was more stable, while 80 000 plants ha^{-1} can be expected to be more stable when the environmental mean is over 7.9 t ha^{-1}. The stability of 100 000 plants ha^{-1} will only be greater than that of 80 000 at an environmental mean of over 13.6 t ha^{-1}.

Analysis of the period between 1981 and 2010 showed that the year had a substantial effect on both the grain yield and the optimum plant density. In dry years the optimum plant density was 64 630 plants ha^{-1}, with a maximum yield of 6.639 t ha^{-1}, while in wet years these figures were 80 790 plants ha^{-1} and 9.864 t ha^{-1} (*Berzsenyi and Tokatlidis* 2012). The stability of plant density ranges also changed as a function of the maturity group (*Berzsenyi et al.* 2011). The stable plant density range was widest (50-90 thousand plants ha^{-1}) for the FAO 200-299 maturity group, narrowing as the vegetation period lengthened and shifting towards thinner stands (50-70 thousand plants ha^{-1}).

Sowing date experiments

Investigations on the sowing date of the hybrid Mv 5 made it clear by the end of the 1950s that the weather had a substantial influence on plant responses (*I'só* 1962), leading to contradictions between the results of individual experiments. For instance, in 1958 this hybrid gave the highest grain yield when sown between May 1st and 20th, while in 1959 it yielded better when sown between April 11th and 30th. Analysis of variance on the results of a sowing date experiment set up with several genotypes in 1960 revealed that the effect of genotype was almost three times as great as that of the sowing date.

Based on the results of a long-term three-factor experiment (N rate × sowing date × hybrid) in the years 2008–2011, the yield fluctuation due to the year was also shown to be considerable (37.8%) in terms of the extreme values of the environmental mean (2008: 10.84 t ha^{-1} vs. 2009: 6.74 t ha^{-1}) (*Árendás* et al. 2012). The effects of these factors on the grain yield was significant in all the years, but in every year the effect of N fertilisation was greater than that of the sowing date or the genotype. These results confirm the findings of *Berzsenyi and Dang* (2003), who reported the greatest effect for the year, followed by N fertilisation, hybrid and sowing date.

On the basis of N responses averaged over sowing dates and hybrids, the highest yields over the three years were obtained on plots fertilised with 180 kg ha^{-1} N active ingredients, while a rate of 120 kg N ha^{-1} had a significantly smaller effect. In years favourable for maize development the 180 kg ha^{-1} N rate had a significant positive effect compared with 120 kg ha^{-1}, but a further increase in the N rate had no significant influence on the yield.

In terms of the productivity of the hybrids averaged over the other two factors, the genetic gain achieved for the Martonvásár hybrids could be detected, in addition to the yield potential as a function of the vegetation period.

Averaged over N treatments and hybrids the grain yield was highest in one year when sown in the last ten days of April, while in the other three years sowing on April 20th had the most favourable effect. In three of the four years, sowing in May resulted in significantly lower maize yields than sowing 10 days earlier, at the end of April. A delay of 20 days compared with the optimum sowing date led to a yield reduction equivalent to 46.9 kg ha^{-1} day^{-1}.

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The success continues with us

Dénes Oross
Bázismag Ltd.
2464 Martonvásár-Erdőhát, Lot No.: 096/36
e-mail: oross.denes@bazismag.hu

Abstract

In 2005 I was challenged as the managing director of Bázismag Ltd. to stop several years of market share downtrend of hybrid corn and turn the company around to achieve profitable growth. Although the reorganisation of the company took several years, the market share of hybrid corn from Martonvásár due to the methods applied has amounted to almost 7% of the total Hungarian corn market in 2013. At the same time, the company has displayed a steady and profitable growth.

Introduction

The agro-industrial sector is the major arena for multinational companies, particularly in Hungary due to our outstanding agricultural output in European terms and our leading position in seed production in Europe. The seed business line (especially hybrid plants) is still quite profitable compared to other business lines. The barriers of the business line to entry and exit are high, and although the number of competitors dropped as a result of company consolidations all around the world, competition remains to be strong due to the struggle of the resulting multinational companies for the acquisition of higher market shares.

Something else seems to be perceptible in the background. The market is immense and profitability is adequate, so why this fierce fight? The answer becomes clear if we take a closer look at the strategic business lines of multinational companies active in the seed market: chemical and insecticide production and distribution, pharmaceutical and genetic research.

Seeds have become strategic carriers and a global race afoot for acquiring markets and the genetic bases.

Materials and methods

We have to develop and implement a marketing strategy for our company in the above outlined competitive environment that leaves nothing, but the only possible alternative – growth.

I have scrutinised and analysed the external and internal environment of the company, performed a SWOT analysis and reformulated the process of target market selection and segmentation. It was important to evaluate which of the competitive strategies and growth strategies may be applied to a Hungarian company, which is one of the minor players in the Hungarian market.

The identification and elaboration of the product strategy are key elements to market development. It is also much easier to communicate distinctive competitive advantages than finding them in the case of homogeneous products. Positioning with the time series data requires a lot of time and perseverance, but it may also help us to find the weak spots of our competitors.

The prices reflect the value of the products and the application of progressive pricing facilitates the increase of profitability.

The new brand strategy of the company is key for development and for the future. Particularly, because there is no future without strong commercial brands in the competition of branded products.

Results and discussion

The market share of improved hybrid corn from Martonvásár together with that of the other Hungarian breeders had dropped to a few per cent by the beginning of the millennium. At the same time, this situation gave an opportunity for the new management to analyse the situation and market opportunities of the company freely and make brave decisions concerning operational and strategic matters.

The development and implementation of a new marketing strategy was obviously imperative, nonetheless, several changes had to be effected beforehand that were prerequisites to the company's survival and, in the case of success, were naturally essential for the adoption and application of the new marketing strategy. First of all we had to begin building our own company brand as part of that. Although we are aware it may take quite some time to perform, the resulting success nevertheless may be the key to creating long-term value.

Products

The product lines of hybrid plants are considerably homogeneous, because all varieties provide the same usefulness although there are clearly dissimilarities in terms of product properties (growing cycle, plant size, colour and resistance to diseases).

On the other hand, it is difficult to determine the value of specific products on the basis of the performance test data as well as whether its usefulness reaches or surpasses that of competitive products. The reason thereto is that performance tests usually last 2-3 years focusing primarily on the variety effects and are less suitable to determine the "variety – production field – crop year" effects. The production site and soil conditions as well as the temperature and precipitation conditions (drought) result in much greater differences of the attainable yield than the yield differences between individual varieties. The applied production technology (cultivation, fertilisation, pest control) also add further discrete factors to the matter at hand, which almost clearly leads to the conclusion that the value of a variety is assessed along a multitude of discrete and subjective elements. It is hardly possible to examine all factors and even the most accurate field tests in most cases fail to present any significant differences.

Products demonstrating the best general adaptability are the most successful in the market. This property, however, is the most difficult to measure and the most prolonged tests are required, by definition, to find the hybrids with good adaptability. Also, in most cases, the adaptability of a product to the conditions of our smaller or broader agricultural environment shows only after its market launch.

Sales methods and market access opportunities

Prior to deciding on the applicable sales methods, we had to identify the target markets in terms of their geographical location. Since the headcount and resources of our company did not facilitate to cover large areas and our commercial experience and knowledge was limited principally to Hungary and the areas of the neighbouring countries inhabited by Hungarians, we focused chiefly on domestic operations. Consequently, we set up three geographical areas of operations:

- Hungary and the areas of Slovakia and Romania inhabited by Hungarians
- other EU countries (Czech Republic, France, Germany, Spain, Bulgaria)
- countries outside the EU (Russia, Ukraine, Iran)

In terms of market access, we had to consider that the products we market have numerous competitors and our we have limited information about most markets (*Kotler and Keller, 2006*).

In order to develop a strategy, we had to identify the company's strengths we could build on as well as the external environmental or market opportunities we could potentially capitalise on.

The co-owner of our company is a breeding institute with the tangible assets, genetic background, basic seeds for breeding and intellectual capacity required for continuous breeding activities. Breeding is indeed successful, because over 20 new hybrids have received state recognition in recent years, and our proven and new products can cover the overall hybrid corn market. Our products have good producibility, production is reasonably cost-effective and good in comparison with our competitors. We have increased the number of our international partnerships, while our significance and market value as efficient breeder and independent trading partner have improved.

There is an increasing demand for new, adaptable and drought tolerant products, which also applies to our products. The energy industry has specified new product properties we can respond to by utilising our research basis. The information pertaining to new demands will support us to market our products on a wider scale. Environmental effects and industrial demands make customers more receptive to try new products, therefore, we have better prospects for introducing our products and encourage our customers to try them. The production of GMO products remains to be unauthorised in Hungary, consequently, this segment continues to be inaccessible to our competitors as well.

Environmental effects (such as lack of irrigation, crop loss due to drought, hectic fluctuation of corn prices), however, exacerbate producers' fears who begin looking for products that satisfy the demand posed by the energy industry and, by way of their good adaptability, tolerate increasingly frequent droughts caused by climate change.

Considering the two opportunities helps us identify what customers want and develop the most effective marketing strategy for the upcoming sales seasons on the basis thereof.

The analysis of the external and internal environment, the competition and competitors demonstrates that intra-sectoral competition constitutes the greatest hazard of the five factors discussed by Porter (*Kotler and Keller, 2006*). There are either no new competitors aspiring to enter this segment or they face an extremely high entry barrier. There is no need to be concerned about threats by substitute products at the moment, nevertheless, the bargaining position of suppliers is very strong. Substantial capital is required to maintain business operations, stock rotation is slow and strategic stock levels must be kept high. Customers are also in a strong bargaining position and brand commitment indeed is a difficult barrier to break. At the same time, market potential may grow, the economic and political environment is in general favourable and we may benefit from the otherwise negative impact of climate change. Our breeding activities are impressive and we have successfully introduced our new products.

I developed the company marketing strategy to achieve the objectives set (increase of corn market share, profit growth), some elements of which deserve to be highlighted.

Our company follows a fundamental strategy based on differentiation endeavouring to distinguish our products with an advantage for customers from those of our competitors and thereby achieve outstanding performance. The company focuses on its own strengths (broad product line, excellent adaptability properties of products, high seed quality and fast service) and communicates them to customers efficiently.

Our company is positioned as an active player of the European seed industry with the ability to breed top quality products, produce the seeds thereof and serve broad target markets. Conventional breeding is combined with active and dynamic trading activity. We strive to produce results through products with excellent adaptability providing the best answer to producers' demands by way of mutually beneficial partnerships with agricultural producers and distributors.

A fundamental aspiration to increase our share in the corn market in a profitable way. Considering the novelty of the products and the market, we also applied the Ansoff product-market expansion matrix to explore growth opportunities (*Kotler and Keller, 2006*).

In terms of the whole company we were forced to apply a combination of diverse strategies. We pursue market development strategy in EU countries entering new markets with our existing products (hybrid corn). We also use the market development strategy in Western European countries with the difference that we deliver the seeds. Our operations focus on the product development strategy in Hungary. We continuously improve our corn product line and increase the number of product varieties within the product line to deliver the most perfect solution to our customers. We expand our product range with new product lines adding rape and sunflower to our product portfolio.

A successful brand may be the cornerstone of a company's mission, long-term growth and the increase of company value in general (*Doyle, 2002*).

We have deemed it necessary to establish our own brand in the market, particularly because hybrid corn, our product is a considerably homogeneous product and the best means of distinction is brand (*Randall, 2000*).

We launched the introduction of the Marton Genetics brand in view of the above principles and have used it in most of our target markets. The introduction of the logo as the image representation of the brand was successful and had a favourable reception. We develop a new image for the new brand in the coming period and begin brand building to create added value.

We have set Bázismag Ltd. on a growth path in recent years in a very strong international competitive environment. The company's market share has increased considerably in Hungary and the Marton Genetics (MG) hybrid corns of Martonvásár are ranked at the 4th place in 2014 behind international competitors. The MG sunflower and rape seeds are becoming recognised in an increasingly wider scope in Hungary. The corn seed export volume of Bázismag Ltd. has increased continuously in the past three years with seeds exported to 7 countries outside Hungary. Our relations with distributors and customers are thriving, sales revenues and profits are growing.

The company has entered its profitable growth phase.

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Maize in Iran

Amir Ansari, Mohammad Reza Sharifi
Adineh Group
IR- Tehran No.2, Saba Alley, Sheikh Bahaee Ave, Mollasadra Ave
e-mail: info@adinehgroup.com

Iran is situated in the south-western part of Asia, in the region known as the Middle East. It has a total area of 1,648,195 square kilometres, making it the 18th largest country in the world. According to the census made in 2011, the population is 76,091,000.

Iran is bordered on the north by Azerbaijan and Turkmenistan, on the east by Afghanistan and Pakistan and on the west by Turkey and Iraq. In addition, part of the northern border is formed by the Caspian Sea, while it is bordered on the south by the Persian Gulf and the Makran Gulf.

Due to its size, climatic location and geographical conditions, several very diverse types of climate may be experienced in Iran at the same time.

The characteristic weather conditions are as follows: freezing; cold; humid and wet; moderately humid and wet; moderately dry; warm and dry; hot and dry; and hot and humid.

Agriculture is the third most profitable branch of the economy after the production and sale of oil and gas. Iran's most important agricultural exports are pistachio and saffron.

Cereal is produced on 8.5% of the total 14,717,500-hectare arable area of Iran. The most important crops are wheat, barley, rice and maize.

Maize is the third most important food and fodder crop. Grain maize is produced on 265,000 ha, which amounts to 2.2% of the arable land, and silage maize on 164,056 ha, or 1.3% of the total arable land.

Maize is produced in the dry and moderately dry areas of the Khuzestan, Fars, Kermanshah, Kerman and Qazvin provinces, generally on irrigated farms.

Maize has a long tradition of cultivation on Iranian farms. Thanks to the farmers' experience, yield averages are 7200 kg/ha for grain maize and 57,346 kg/ha for silage maize on irrigated areas, while on non-irrigated land these figures are 2380 kg/ha and 18,023 kg/ha, respectively. The very dry weather experienced in recent years has proved a great challenge to growers.

The seed requirements of the country amount to 15,000–16,000 t, of which 13,000 t is produced in Iran, while the remaining 2000–3000 t is imported. The varieties sown can be divided into three main groups: late (FAO 600–700), medium late (FAO 500–600) and medium early (FAO 450–500). Late varieties make up 58% of the market, medium late varieties 30% and medium early varieties 12%.

The attention of large-scale growers and breeders in Iran was attracted by the results achieved by breeders in the countries of Eastern Central Europe, where the climate is favourable and there is a long tradition of scientific excellence, several decades ago. Based on long-term contacts, the major importers of maize seed into Iran are Hungary, Serbia and Croatia.

Thanks to its favourable agricultural conditions, almost 60% of the sowing area in Hungary is suitable for seed production. As far as we know, wheat and maize are grown on 80% of the total arable land in the country. However, apart from these two crops, the country is also known to have great experience in the production of other crops, such as sunflower.

Hybrid seed breeding began in Hungary in the 1930s and in most places hybrid seed has been sown since the 1960s. According to our information, 25% of the seed produced each year is exported. In recent years, Iran has become one of the major markets for maize seed and Hungary is now the 3rd ranked exporter.

The company Arman sabz Adineh has the exclusive rights to Hungarian maize seed in Iran and plays a key role in introducing and popularising Hungarian varieties on the Iranian market.

The first import transactions were made in 2010 and involved a total of 118 mt maize seed. With the help of the company's extensive trading network, the attention of farmers was quickly drawn to the advantages of Hungarian seed, so the first transaction was a great success.

The maize variety traditionally grown in Iran on the widest scale is Sc 704. However, thanks to its shorter ripening period and lower water requirements, Maxima has become a popular alternative. It is ideally suited for sowing as a second crop or in places where sowing is delayed for any reason.

Maxima and Siloking are now grown widely under cooler conditions in the Gazvin, Hamedam, Chaharmaha and Bakhiari provinces, where they produce outstandingly high yields. Under the warm conditions in Fars and Ilam provinces these hybrids can also be successfully grown, thanks to their good stress tolerance and low water requirements.

As the result of constant efforts, synchronised with the progress made by Hungarian breeders, our company is gradually increasing the exports of Hungarian maize seed to Iran, and this rise is expected to continue in a dynamic manner.

The Arman sabz Adineh Company has invested over 5 million dollars in the seed industry. The investments include the construction of a seed processing plant, and the establishment of seed dryers, warehouses and logistics centres. The developments made by the company represent a national project. At present the Arman sabz Adineh seed processing plant is the only such facility in private hands in Iran.

In summary, it can be said that the dynamically increasing demand for food and fodder on the Iranian market has led to a rapid expansion of imports and a considerable rise in local production. Due to the rise in maize cultivation, Iran is a promising potential market for Hungarian seed. The aim of the Arman sabz Adineh Company is to expand the Hungarian seed market by increasing the volume of seed for the varieties already grown in the country, and by introducing new varieties into Iran. This work is still in the initial phases, and we are still far from the limits of market capacity. The key to successful cooperation in the long term is for our Hungarian partners to develop hybrids with good adaptability to Iranian weather conditions and with a satisfactory production technology.

Our philosophy and our opinion on the breeding of Lfy hybrids

Francis Glenn[1], Sietse Pen[2]
[1]Glenn Seed Ltd
RR#1, Blenheim, ON, Canada, NOP 1 A0
[2]Glenn Maize France BV,
81 Rte de la chapelle de Rousse, 64290 Gan, France

The development of Lfy silage maize varieties was the result of developments in thinking and genetic materials that led to a revolution in silage maize production. The era of dual-purpose hybrids is coming to an end, as growers have experienced what Lfy silage maize hybrids are capable of in terms of yield and fodder value.

Maize breeding basically means the breeding of inbred lines (parental components). In a crossed population we look for sublines that carry the multitude of traits we would like to see in the end-product and that have the best genetic background. In the course of inbreeding selection is made for the desired traits in an effort to find the genotype that incorporates the largest number of these traits. It is amazing to see the results of this work. What we are looking for in silage breeding is good germination ability, good initial development under both warm and cold conditions, rapid spring development to produce a closed canopy as soon as possible, a high yield with large and extra leaves, a large ear with healthy kernels, soft kernels with favourable starch composition, flexible but strong stalks with a thin epidermis, good disease resistance in the mature leaves so that the plant stays green and fertile for a long time, early flowering, a long grain-filling period and seeds that are still soft at silage maturity.

Selection was carried out over more than twenty generations for families with normal and higher leaf number, satisfying the above criteria, and the result was the development of a number of stable inbred lines that were suitable for the creation of leafy (Lfy) hybrids.

The product bought by the farmer is the hybrid. So crosses are made between lines originating from various families, which can be expected to produce robust, high-yielding hybrids. Based on many years of experiments and the responses of farmers it can be said that our efforts have produced hybrid 'ideotypes' that are exactly what the farmers want. When prospective hybrids are tested the aim is to find genotypes that have all the traits of the 'ideotype' and perform better than those previously used by silage growers.

As the ear of Lfy types is attached lower down the stalk, the harder stalk section below the ear makes up a smaller proportion, the photosynthesising leaf area that feeds the ear is greater, and there is less stalk breakage and root lodging, as the centre of gravity of the plant is lower. Selection for more flexible stalks and the stay-green trait has resulted in lower lignin content in the stalks than in that of grain-type hybrids. Lignin inhibits digestion, so stalks with low lignin content are easier to digest.

Lfy hybrids have more young leaves above the ear (*Fig. 1*), with a consequent rise in the sugar content in both the stalks and the leaves at harvest. Farmers have reported that Lfy hybrids ferment more rapidly, giving a sweet smell to the silage. Cows love it, and the farmers say they eat more of it, thus increasing the milk yield.

Selection was consistently made for hybrids with softer seed tissue at harvest. Due to the softer starch the kernels are more easily decomposed during chopping, so the starch is more rapidly converted into sugar.

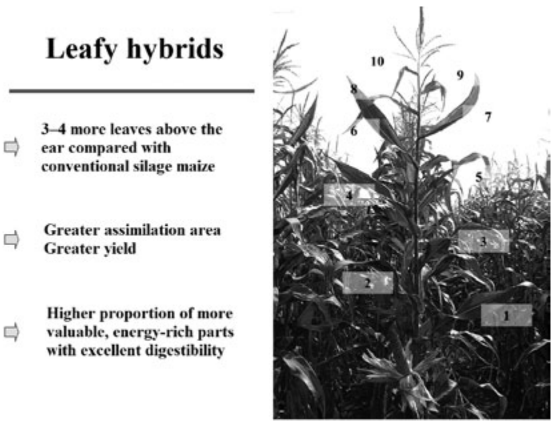

Fig. 1. Main characteristics of Lfy hybrids

Hybrids were selected with a longer grain-filling period and with leaves that remained healthy right up to harvest. These plants take longer to reach the 65% moisture content in the whole plant that is indicative of silage maturity, and the moisture content stays in the ideal 60–70% range for a longer time than in the case of grain-type hybrids. This protracted harvesting period is greatly appreciated by farmers.

A high proportion of grain in the silage is a desirable trait. Selection is made for large ears and high grain yield, but not on the basis of experiments aimed at producing grain-type hybrids, so Lfy hybrids are not recommended for dual-purpose utilisation. The hybrids are also selected for stalk quality, so that the quality remains good until silage maturity, but as a criterion this is only important for grain hybrids. Importance is also attached to slow maturing, which is probably linked to slow drying down after the black layer has formed.

The field experiments are performed with a plant density of 69,000 plants/ha and selection is made for types with intensive growth, a high grain ratio and high yield. Research has shown that higher plant density gives a higher yield under optimum conditions, but under average or worse conditions a high plant density leads to a reduction in the grain ratio, while the stalk portion of the whole silage has poorer digestibility. The experiments demonstrate that the rival grain hybrids used as standards do not perform better in an average environment at high plant density. At a density of 69,000 plants/ha the Lfy hybrids form a large canopy, thus ensuring a satisfactory quantity and quality of silage even on dry soil.

According to the farmers, Lfy hybrids produce larger ears in dry years than grain hybrids at the same location. We have also observed this on countless occasions. Despite the fact that the Lfy plants have a greater canopy and leaf area, they still perform better under dry conditions. This is only true of the varieties selected by us, not of Lfy hybrids that do not possess drought tolerance. The hybrids are tested at numerous locations until we can establish the limits to where they can be grown and have a good idea of how they perform under stress conditions. Last, but not least, we make sure that they carry the best possible combination of the desired traits.

Breeding for use as silage thus has different aims to those targeted by grain hybrid breeders. Marketing experts who work for companies selling ‘dual-purpose’ grain/silage hybrids simply call hybrids with slow drying down silage hybrids, and the ‘dual-purpose’ label is just an advertising trick.

Harvested whole plants stored as silage form a complex product and it is difficult to express its value for growers. Its value can best be measured in terms of milk yield per hectare. But how can this value be determined, and how can the value of different hybrids be compared?

The fresh yield can be measured using various experimental techniques by testing the hybrids over several years at several locations. An experimental harvesting, sampling and drying protocol has been elaborated that allows the experiments to be managed precisely and efficiently and means that four people can harvest up to 550 plots a day.

The determination of quality is another story. In recent years we have consulted a great number of livestock feeding experts and fodder chemists, and have come to the final conclusion that the most important parameter is neutral detergent fibre (NDF) and its digestibility. Although the acid detergent fibre (ADF) is used as a parameter to denote milk yield per hectare and to determine relative nutritional value, it is not actually correlated to anything. We started analysing volatile fatty acids to see how the sugar is fermented to form the volatile fatty acids that cause such a great difference between Lfy and dual-purpose hybrids in terms of flavour and the initial stages of digestion. Farmers have often told us that they notice a sweet smell when storing and feeding silage made from Lfy hybrids. This better quality silage results in greater nutrient uptake and thus higher milk production.

Fig. 2. Silóking, the most popular Lfy hybrid

So how should selection be made for silage hybrids with excellent quality? Based on our personal convictions, we work on the following hypothesis:

1. We work with inbred lines that have extra leaves and that result in hybrids with at least two extra leaves above the ear, so that the total leaf number above the ear is at least 11.
2. Varieties are tested at a maximum plant density of 69,000 plants/ha.
3. Emphasis is placed on agronomic traits, the growth rate, and resistance to root and stalk lodging.
4. Selection is made for even more flexible stalks with a thinner epidermis.
5. Selection is made for large ear size and soft seed structure.
6. We check that large ears are produced at all the locations in the trials, as this is the guarantee of yield stability.
7. Slow-maturing varieties are sought for, as these provide farmers with a wide harvest period, together with ideal moisture content for storage and feeding.
8. Among the hybrids meeting the above criteria, the one with the highest yield is chosen.
9. Attention is paid to new digestibility data from feeding experiments to ensure that the hybrid with the best digestibility is chosen from among those that meet the above criteria.

Lfy hybrids have changed the way farmers think. Those who choose Lfy hybrids are quite clear about the fact that they want to grow silage maize. They also know that with these silage hybrids they can produce feed with better quality than with grain hybrids. So they are prepared to change their production technology so that the better quality latent in leafy silage hybrids can be fully manifested. At present the most popular Lfy hybrid in Hungary is Silóking (*Fig. 2*).

At present we can witness a revolution in silage maize production, in which Lfy hybrids play an outstanding role.

On the 60th anniversary celebration

Oszkár Gyulavári
Cereal Research Non-Profit Ltd. Co., Szeged-Táplánszentkereszt

First and foremost, I would like to warmly congratulate on the achievements of maize breeding in Martonvásár in the past 60 years and I feel obliged to add a few words to what has been said before. I am lucky that I can say that I had already been a maize breeder when Mv 5 was registered and not only did I personally know its breeder Endre Pap but also I had the chance to work with him in different projects. I would like to mention only one of these projects, in which he was the chairman of a maize breeding collective and the evaluation of results and the elaboration of the action plan for the following year was managed by a team of three people I had the opportunity to be part of with him and László Berzsenyi.

Endre Pap's inevitable merit was to follow all the new methods which could lead to great success worldwide and he felt capable and ambitious enough to be able to breed high-yielding maize hybrids himself. He did not have a lot of opportunities and he really had to fight an uphill struggle and a lot of people observed his projects very sceptically, but today we really must acknowledge his work and his achievements. I can remember once a US visitor, Mr Garszt expressed his opinion on Endre Pap's work, which was shared by many those days. Mr Garszt said he appreciated his incredible efforts Endre Pap made in his job but he mentioned the example of the dead pig, which could never catch up with the maturity level of the other pigs any more, which was a symbol used for showing the differences between the struggling breeding work done by Endre Pap and the American way. But Endre Pap, despite all these obstacles finally was able to end the battle in triumph.

After he left the country, the maize breeding collective broke up although it could have contributed to the success of the Hungarian maize breeding with its work. Maize breeding was relocated to Keszthely belonging to the Ministry of Agriculture, where we were dealing with creating the basic conditions of research for years. In Martonvásár, the institute could carry on research work undisturbed and could achieve immortal merits in providing the whole country with seeds of high-yielding hybrids.

In the late 1960s László Berzsenyi started the Hungarian-German cooperation in the field of maize breeding, which became trilateral with the joining of the Polish maize breeders. In this collective work maize breeders in Martonvásár also played an important role.

Seed exports of the collective hybrids used to be higher than the demand for hybrid maize seeds in Hungary, yet exports in certain years exceeded the demand several times over. After the change of the political system in Hungary this work unfortunately halted. New ways have had to be found. I am aware of that serious methodological research has been conducted in Martonvásár in the field of in vivo haploid induction method, which has been introduced and used successfully by a lot of companies in the world. There is more and more interest focused on the East and as a result of this a lot of hybrids from Martonvásár are taking part in research abroad and the results are really promising concerning the seed export prospects.

I wish the maize breeder colleagues in Martonvásár to be able to recognize the possibilities of the present and the future and to be able to work persistently to achieve their goals. I hope that after its glorious past, maize breeding in Martonvásár can expect a very promising future and as in the past we will be able to cooperate very successfully in the future, too.

Section lectures

Section 1

Effect of year and preceding crop on the fertiliser response of maize

Tamás Árendás[1], Zoltán Berzsenyi[1], Péter Bónis[1], Györgyi Micskei[1]*, Péter Csathó[2]
[1]*Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences. 2462 Martonvásár, Brunszvik u. 2.*
[2]*Institute for Soil Science and Agricultural Chemistry, CAR HAS*
1022 Budapest, Herman O. u. 15
e-mail: arendas.tamas@agrar.mta.hu

Abstract

The yield responses of maize were investigated in a long-term fertilisation experiment set up in 1959 on chernozem soil with forest residues. A period involving 14 years in which the maize hybrid Norma SC was sown (1988-2013) was analysed in the present work. In the 9 years when mineral fertilisers were shown to have a positive effect, the role of N in enhancing productivity could be demonstrated in all cases, and that of P and K in three each. There were considerable differences in yield responses as a function of year and preceding crop. In dry years macroelements had no significant effect on maize sown after wheat. Averaged over the years in which the rainfall quantity over the vegetation period exceeded 350 mm, N, P and K significantly increased the maize grain yield when the preceding crop was wheat.

Introduction

The effect exerted by mineral fertilisers on crops and the environment is influenced by numerous factors. Soil properties, the state of the atmosphere, the crops grown, the crop sequence and the traits of the individual varieties are all of decisive importance for crop productivity (*Nagy* 2007).

The data from several decades of long-term fertilisation experiments on maize in Hungary have proved that, among the macroelements, the effect of nitrogen is greatest and can be detected most frequently. Like all row crops, maize gives a better response to improved K supplies than cereals. By contrast the P response of maize and the P requirements of the crop are lower than those of wheat (*Csathó et al.* 2009).

The effect of mineral fertilisers on crop productivity is greatly influenced by crop rotation, partly due to the diverse water and nutrient consumption of the successive crops. While the yield-increasing effect of wheat crop rotations is manifested regardless of the fertilisation level, in maize sequences fertilisation reduces the effect of rotation by almost 50% (*Berzsenyi et al.* 2000).

Materials and methods

The effect of N, P and K fertilisers, alone and in combination, on the plant–soil system has been investigated in a maize–wheat diculture since autumn 1959 in a long-term fertilisation experiment set up by Mihály Krámer on chernozem soil with forest residues in Martonvásár. Until 1975 the quantities of active ingredients (kg ha^{-1}) supplied each year were 64 (N), 39 (P_2O_5) and 61 (K_2O), while from autumn 1975 onwards they have been increased to 160, 80 and 80.

The effects of the treatments are examined on plots measuring 6,5 × 7,7 m = 50.05 m², arranged in a random block design. Half the N active ingredients and the whole quantity of P and K are added in autumn, before basic soil cultivation, and the remaining N in spring, prior to sowing. The fertilisers are applied in the form of calcium ammonium nitrate or NH_4NO_3 (27 or 34% N), superphosphate (18% P_2O_5) and potassium chloride (40 or 60% K_2O).

At the start of the experiment the topsoil (0-20 cm) had a pH_{H2O} value of 7.2 and contained 3.0% humus, 0.8% $CaCO_3$, 30-40 mg kg^{-1} AL-soluble P_2O_5 and 150-200 mg kg^{-1} AL-K_2O. After the change in the fertiliser quantities, plots fertilised with phosphorus were placed in the ‘good’ supply category according to the official Hungarian classification system (*MÉM NAK* 1979) from 1984 onwards and those treated with potassium in the ‘very good’ category from 1980 onwards (*Árendás et al.* 2003).

In each four-year fertilisation cycle, the first two crops are maize, followed in the 3rd and 4th years by winter wheat. This paper reports the results obtained for maize in seven fertilisation cycles, from 1988-2013. In all 14 years the genetic background was constant, as the effects of the fertilisation treatments were compared each year based on the yield responses of the Martonvásár hybrid Norma SC (FAO 390). The years were divided into dry and wet years on the basis of the rainfall quantity during the vegetation season (*Fig. 1*) compared with the mean for the 14 years, using the limit values reported by *Harnos* (1993). On this basis the years 1988, 1993, 1997, 2000, 2009, 2012 and 2013 were classified as dry and the years 1989, 1992, 1996, 2001, 2004, 2005 and 2008 as wet.

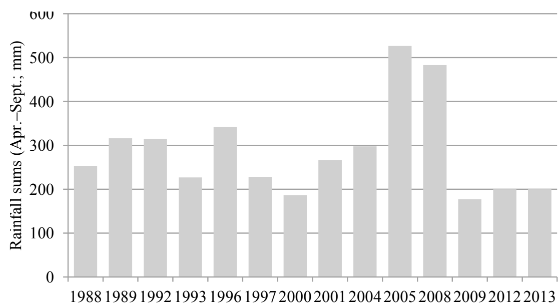

Fig. 1. Rainfall quantities (mm) during the vegetation season of maize (Apr.-Sept.). Martonvásár, 1988–2013.

The data were evaluated using one-way analysis of variance for a random block design according to Sváb (1981).

Results and discussion

Based on the yield responses of maize, significant fertiliser effects could be observed between the treatments in 9 of the 14 years (*Table 1*). The yield-enhancing effect of N was significant in all 9 years, and the positive role of P and K in three years each.

No difference could be observed in the control plots between the average yields achieved after different preceding crops (wheat in the 1st year and maize in the 2nd).

The deviation expressed as a percentage of the mean value (CV%) revealed that mineral fertilisers had a substantial yield-stabilising effect on maize sown after maize, the greatest reduction in fluctuation being achieved by the joint application of N and P.

Averaged over the 14 experiments the NPK treatment resulted in the highest grain yields. Based on differences between treatment pairs, a dose of 160 kg ha^{-1} N increased the yield by 28.6% on chernozem soil with forest residues, which is well supplied with phosphorus and potassium. The magnitude (NPK-NK=0.46 t ha^{-1}) and proportion (100×NK/NPK=94%) of the P effects were smaller than in earlier periods of the experiment (*Árendás et al.* 2003) and in the first 10 years of other field experiments on maize set up under similar conditions (*Csathó* 2002). The mean value of the K effect was the same as that recorded in the earlier period, with values of 0.44 t ha^{-1} (NPK-NP) and 94% (100×NP/NPK) in the period 1988-2013.

During half of the experimental years the nutrient responses of the Norma SC hybrid, which is drought-tolerant and has excellent adaptability, were examined under dry or droughty conditions. The rainfall deficit in years when maize was sown after wheat was 74 mm (-26%) compared with the mean for the whole period (287 mm). The grain

Fertilisation cycle	Year	Preceding crop	Treatment					$LSD_{5\%}$
			0	NP	NK	PK	NPK	
VIII.	1988	Wheat	4.93	5.90	5.08	5.20	5.38	ns
	1989	Maize	4.92	9.11	7.99	5.45	8.37	1.44
IX.	1992	Wheat	2.95	3.67	3.93	4.31	4.39	ns
	1993	Maize	3.68	4.10	4.70	4.38	4.09	ns
X.	1996	Wheat	6.90	8.58	8.28	6.82	10.04	1.34
	1997	Maize	6.58	9.84	8.56	7.15	10.17	1.32
XI.	2000	Wheat	5.53	7.39	7.02	5.82	7.71	0.91
	2001	Maize	6.97	10.32	10.04	8.55	10.17	1.12
XII.	2004	Wheat	5.65	7.86	7.64	5.81	8.05	0.71
	2005	Maize	7.52	9.01	9.51	7.11	9.61	1.68
XIII.	2008	Wheat	6.41	9.54	9.39	6.27	10.13	0.45
	2009	Maize	4.23	5.27	6.07	4.69	6.49	0.68
XIV.	2012	Wheat	5.65	3.83	4.85	5.44	4.72	ns
	2013	Maize	4.91	4.54	5.64	4.79	5.77	ns
Mean (after wheat)			5.43	6.68	6.60	5.67	7.20	
CV% (after wheat)			13.3	13.3	12.0	9.8	13.0	
Mean (after maize)			5.54	7.46	7.50	6.02	7.81	
CV% (after maize)			18.0	8.9	12.3	18.3	9.2	
Mean (VIII-XIV.)			5.49	7.07	7.05	5.84	7.51	

ns = non-significant

Table 1. Effect of NPK mineral fertiliser combinations on the yield responses of maize Martonvásár, 1988–2013

yield recorded on the unfertilised control plots was 5.37 t ha^{-1} in these years. The deficit in the dry years when maize was grown after maize was similar (79 mm), while the yield was 4.85 t ha^{-1}. In wet years the total rainfall for the period April-September averaged 359 mm after wheat and 370 mm after maize, which resulted in grain yields of 5.48 and 6.47 t ha^{-1} in the unfertilised treatments.

Among the macroelements, N caused the greatest increase in the productivity of maize after wheat in dry years and K the smallest, but the effects were not significant for any of the nutrients (*Fig. 1*). In maize grown after maize in rainfall-deficient years the yield-enhancing effect of K was three times greater than when wheat was the preceding crop (0.69 vs. 0.23 t ha^{-1} $year^{-1}$), but was below the value (0.81 t ha^{-1} $year^{-1}$) significant at the P=5% probability level. When maize was sown after maize, 160 kg ha^{-1} nitrogen caused a significant increase in yield of 1.38 t ha^{-1} under dry conditions. This positive effect was considerably greater in wet years (2.35 t ha^{-1}), but P and K responses could not be detected on the basis of maize grain yield. In years when the rainfall quantity was greater than 350 mm no difference could be observed between the N responses of maize grown after maize or wheat. In years favourable for maize development the grain yield of Norma SC sown after wheat was significantly increased by both P and K fertiliser, the positive effect of 80 kg ha^{-1} P_2O_5 being 0.84 t ha^{-1}, while that of 80 kg ha^{-1} K was 0.74 t ha^{-1}.

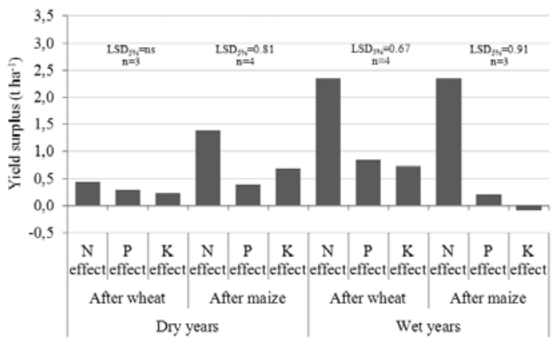

Fig. 1. NPK responses of the maize hybrid Norma SC as a function of year and preceding crop. Martonvásár, 1988–2013

Acknowledgement

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Analysis of maize yield responses from the time of Béla Győrffy to the present

Zoltán Berzsenyi, Tamás Árendás, Péter Bónis, L.Csaba Marton
Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
2462 Martonvásár, Brunszvik u. 2.
berzsenyi.zoltan@agrar.mta.hu

Abstract

The effects of N fertilisation, sowing date and plant density and their interactions on the yield and yield stability of maize hybrids were examined using the data of several decades of long-term experiments in Martonvásár. In comparison with a monoculture, a yield surplus of 2.25 t ha^{-1} was achieved in crop rotation, using 40–80 kg ha^{-1} less N fertiliser. At non-optimal sowing dates reductions were observed not only in the yield, but also in the efficiency of N fertilisation. The optimum plant density for the maize hybrids rose by 8 000 plants per hectare every 10 years between 1981 and 2010. Significant N fertiliser × hybrid and N fertiliser × plant density × hybrid interactions were detected in the experiments. Determining the growth characteristics of maize hybrids facilitates the more precise identification of agronomic responses. Studies on the year effect are especially important in the light of the expected effects of climate change.

Introduction

Right from the beginning, there has been close cooperation between maize production and maize breeding research in Martonvásár. As early as the 1950s, the results of maize production experiments revealed that yields, or yield increases, were determined by the joint effect of several factors, not just by the variety. It became clear that the spread of hybrids and the exploitation of their latent yield potential would necessitate basic research on modern agronomic technologies. From the 1960s onwards, the production technologies and recommendations elaborated by scientists from Martonvásár, based on several decades of experimental data, became important components of Hungarian maize production (*Győrffy,* 1979). From the very start, investigations on the agronomic responses (fertiliser utilisation, plant density and sowing date responses) of new Martonvásár hybrids, carried out in bi- and multi-factorial experiments, formed an important part of maize production research, the aim of which was to determine or predict the optimum ranges of agronomic responses and the expected yield levels. In crop production research, in addition to measuring the direct effect of various factors, the analysis of interactions between two or more factors in various types of factorial experiments is gaining in importance.

Materials and methods

The N fertiliser responses of maize hybrids were investigated in two diverse environments: a 50-year-old monoculture (stress environment) and a Norfolk crop rotation (optimum environment). In both experiments various N doses were applied with identical supplies of P and K. In the maize monoculture the N fertiliser doses were the following (in kg ha^{-1}): 0, 80, 160 and 240 (designated as N_0, N_{80}, N_{160} and N_{240}), while in all the treatments the P and K doses were 160 kg ha^{-1}. In the crop rotation (maize, spring barley, peas, winter wheat) the N fertiliser responses of the maize hybrids were examined over a range of 0–280 kg N ha^{-1} in 40 kg ha^{-1} steps. The P and K quantities were the same in all cases (120 kg ha^{-1}). The effect of the year on the N fertiliser response was investigated in the maize monoculture between 1970 and 2009, together with the N fertiliser × maize hybrid interaction. In the years 1995–2007 the N fertiliser responses of the hybrids were compared at four N fertiliser levels (N_0, N_{80}, N_{160} and N_{240}) in monoculture and crop rotation, using five hybrids from different maturity groups in both experiments.

The effect of sowing date and N fertiliser on the grain yield of maize hybrids was studied between 1991 and 2009 in a long-term N fertilisation experiment set up in 1980. In this three-factor, split-split plot experiment, N fertilisation formed the main plots, the sowing date the sub-plots and the maize hybrid the sub-subplots. The N treatments were as follows: 0, 60, 120, 180 and 240 kg ha^{-1}. The sowing date treatment involved four sowing dates: 10 days before the optimum date (early), the optimum date (around April 24), 10 days after the optimum date (late) and 20 days after the optimum date (very late). The maize hybrids represented various maturity groups.

The effect of plant density on the maize grain yield was investigated in a strip-plot series of experiments at nine plant densities between 20 000 and 100 000 plants ha^{-1} (20, 30, 40, 50, 60, 70, 80, 90 and 100 thousand plants ha^{-1}). The effect of year and plant density on the yield and yield stability of maize is presented on the average of 20–45 hybrids a year, based on data for the years 1981–2010.

The effect of mineral fertilisation and plant density on the yield of maize hybrids was examined between 1989 and 1999 in a three-factor long-term experiment, set up in a split-split plot design with four replications. The treatments were as follows: three fertiliser levels (A, C, E) with the following NPK doses (kg ha^{-1}): A: $N_0P_0K_0$, C: $N_{100}P_{48}K_{87}$, E: $N_{300}P_{144}K_{261}$; three plant densities: 40, 70 and 100 thousand plants ha^{-1}; and three maize hybrids with diverse vegetation periods: *Mara* (FAO 297), *Norma* (FAO 380) and *Maraton* (FAO 450). In the years 2001–2003 the effects of fertilisation, plant density and hybrid on the growth and yield of maize were examined using growth analysis, using the program (HP model) developed by *Hunt* and *Parsons* (1974) The following growth parameters were calculated: absolute growth rate (dry matter: AGR, leaf area: ALGR), net assimilation rate (NAR), leaf area per plant and total dry matter.

Results and discusson

N fertiliser response of maize hybrids

Among the agronomic factors, nitrogen (N) fertilisation is one of the most important factors for increasing the yield of maize. At the same time, under Hungarian conditions water deficit stress regularly limits plant yields and nutrient utilisation. *The effect of N fertilisation and the year* on maize grain yields is presented on the basis of data for 14 dry and 26 wet years between 1970 and 2009 (*Fig. 1*). It is clear from the figure that the maize yield was smallest in the N_0 treatment in both types of years, sig-

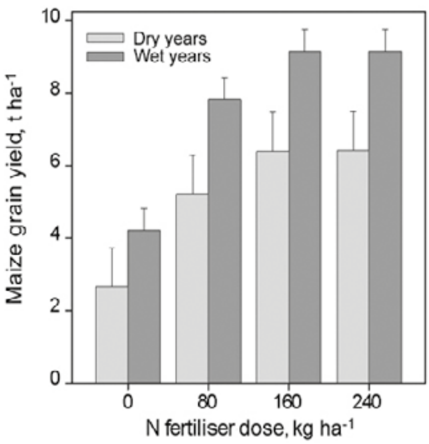


Fig. 1. Effect of N fertilisation on the grain yield of maize in dry (14 years) and wet years (26 years) between 1970 and 2009

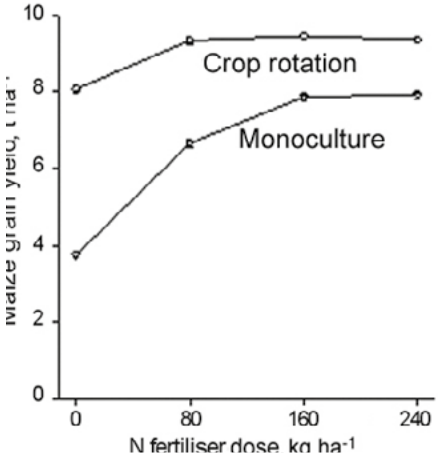


Fig. 2. N fertiliser responses of maize hybrids in monoculture and crop rotation (1995–2007)

nificantly increasing in the N_{80} and N_{160} treatments. The highest yields were obtained at the 160 kg ha⁻¹ N rate. When the dry and wet years were compared, the yield surplus (t ha⁻¹) achieved at each N rate in the wet years amounted to: N_0: 1.567, N_{80}: 2.616, N_{160}: 2.764, N_{240}: 2.740.

The effect of the *N fertiliser × maize hybrid interaction* on the yield was evaluated by means of combined analysis of variance in a maize monoculture between 1985 and 1994 (on the same five hybrids) and between 1995 and 2002 (on the same seven hybrids), taking the years into account *(Berzsenyi* and *Lap*, 2003). The results

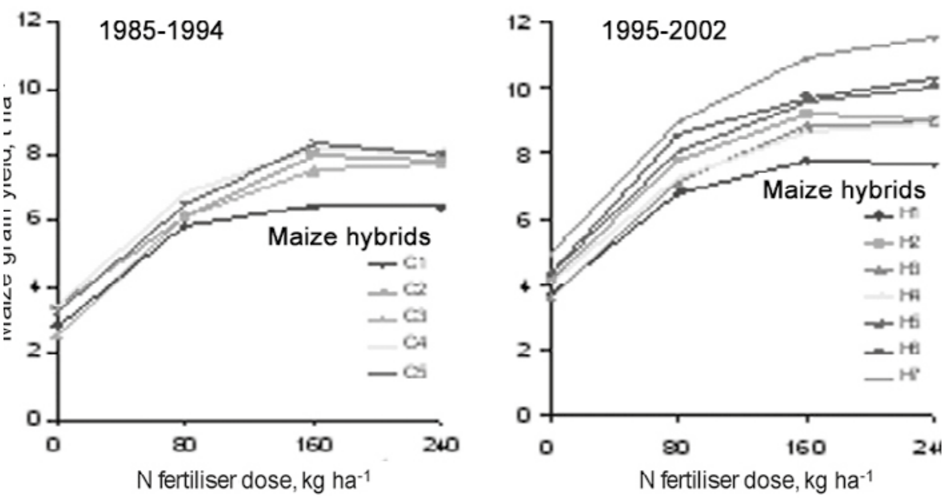


Fig. 3. N fertiliser × maize hybrid interaction during the periods 1985–1994 and 1995–2002 based on measured data (Berzsenyi and Lap, 2003). Vegetation periods increase from C1 and H1 to C5 and H5.

showed that all three factors (year, N fertiliser, hybrid) had a significant effect. Based on the MS values, the main effects were clearly the most important. The significant interaction between N fertiliser and the maize hybrid indicated that the differences between the hybrids were not the same at different N fertiliser levels. Further dissection of the N fertiliser × hybrid interaction SS using the orthogonal polynomials method revealed that the N fertiliser × hybrid interaction could be attributed primarily to differences in the linear section of the yield responses of the hybrids to N fertiliser. However, there may also be significant differences in the second or third degree components (*Fig. 3*).

Sowing date response of maize hybrids

The *sowing date × N fertiliser interaction* in dry (12) and wet (7) years is illustrated in *Figure 4*. It is clear that in both years the efficiency of N fertilisation declined after late sowing, especially in dry years. Averaged over the treatments the yield was 2,553 t ha^{-1} greater in wet years than in dry years.

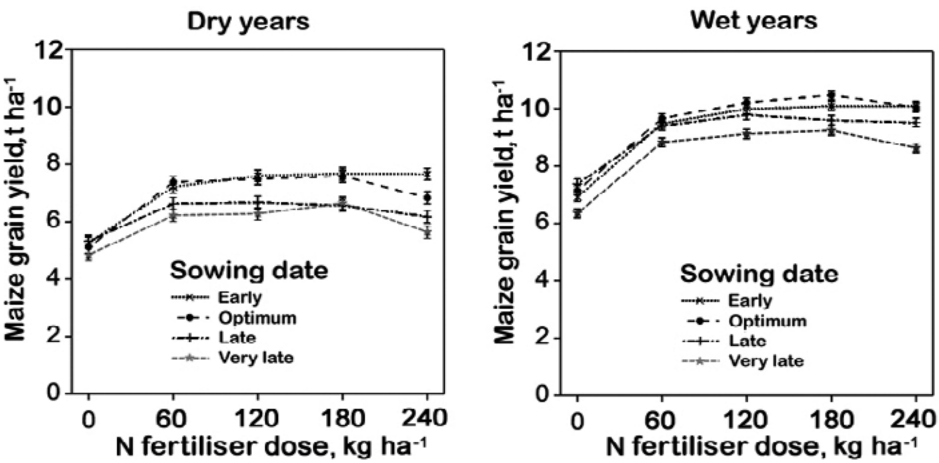


Fig. 4. Effect of the N fertiliser × sowing date interaction on the grain yield of maize in dry (7) and wet (12) years (1991–2009)

The yields were highest after early or optimum sowing in the dry years (7.083 and 6.880 t ha^{-1}, respectively), decreasing significantly when sowing took place 10 or 20 days later (6.273 and 5.925 t ha^{-1}, respectively). In dry years N fertiliser rates higher than 60 kg ha^{-1} did not increase the yield; in fact, a significant yield reduction was observed in the N_{240} treatment. In wet years the highest yields (t ha^{-1}) were obtained in the optimum (9.500) and early (9.312) sowing date treatments, with significant declines after late (9.131) and very late (8.431) sowing. In wet years the optimum N fertiliser dose was 120 kg ha^{-1} and at higher doses there was no significant change in the yield. The greatest yield stability was exhibited after sowing at the optimum date or 10 days later and with N fertiliser rates of 60 and 120 kg ha^{-1}.

Plant density responses of maize hybrids

Of all the agronomic factors, the plant density has changed to the greatest extent in recent decades. The increase in the plant density and the identification of hybrids with high genetic yield potential at high plant density were important components in the increase in maize grain yields during this period.

Trends in the optimum plant density over time were examined using the 1980–2010 data of plant density experiments in Martonvásár. The optimum plant density each year was calculated by fitting quadratic functions and changes over time were recorded. A significant ($P<0.01$) linear correlation characterised the change in the optimum plant density over time (*Fig. 5a*) and revealed an increase of 8100 plants ha^{-1} every 10 years between 1981 and 2010.

Averaged over the hybrids investigated, the optimum plant density per hectare was found to increase from 64 000 in the 1980s to 80 000 in 2010.

Under the given experimental conditions fluctuations in the rainfall had a substantial influence on the *optimum plant density and the yield*. The effect of the year on the optimum plant density and grain yield of maize was examined during the period 1981–2010. A quadratic function gave a good fit to the experimental data. The year was found to have a significant effect on the grain yield and optimum plant density (*Fig. 5b*). In dry years the optimum plant density was 64 630 plants ha^{-1}, with a maximum yield of 6.639 t ha^{-1}, while in wet years these figures were 80 790 plants ha^{-1} and 9.864 t ha^{-1}. It was established that in unfavourable years the yield was approximately 1/3 less than in favourable years, while the optimum plant density was 20% lower. In dry years a plant density greater than the optimum resulted in substantial yield losses (as shown by the increase in the distance between the two curves).

When the diverse weather conditions over 22 years (1981–2002) were taken into consideration, the yield stability was found to be greatest at a plant density of 60 000 t ha^{-1}. A plant density of 40 000 t ha^{-1} was more stable at an environmental mean of less than 4.6 t ha^{-1}, while a plant density of 80 000 t ha^{-1} can be expected to be more stable

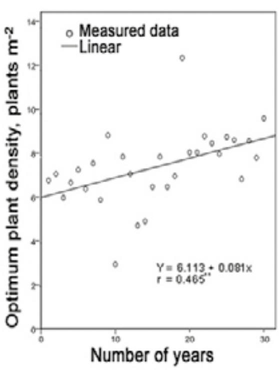


Fig. 5a. Linear trend in the change in optimum plant density each year between 1981 and 2010

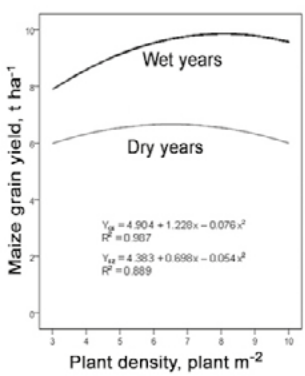


Fig. 5b. Effect of the year on the maize grain yield and the optimum plant density between 1981 and 2010

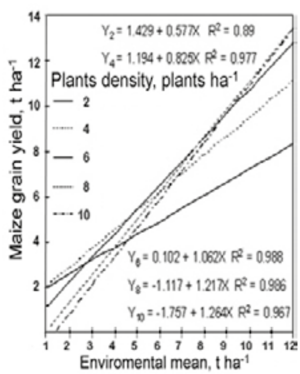


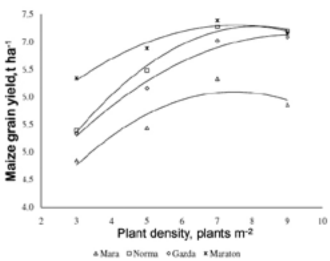
Fig. 5c. Yield stability of maize hybrids at various plant densities between 1981 and 2010

when the environmental mean is over 7.9 t ha^{-1}. If the stability of 100 000 plants ha^{-1} is to exceed that of 80 000 ha^{-1}, an environmental mean of >13.6 t ha^{-1} is required (*Fig. 5c*).

The analysis of data from a series of plant density experiments from 1981–2005 revealed that the stability of the plant density ranges was a function of the hybrid maturity group. The widest range of stable plant densities (50–90 thousand plants ha^{-1}) was found for the FAO 200–299 group. As the vegetation period lengthened the range of stable plant densities narrowed, shifting towards lower plant densities (50–70 thousand ha^{-1} in the FAO 400–499 and FAO 500–599 groups).

Plant density × N fertilisation and plant density × hybrid interactions

Among the various types of interactions, the most important from the crop production point of view are those that characterise the relative performance of the genotypes in diverse environments. *Figure 6* illustrates the plant density responses of the hybrids at 200 kg N ha^{-1}. The quadratic functions demonstrate that the optimum plant density was 7.48, 8.11, 9.22 and 7.53 plants m^{-2} for the hybrids *Mara, Norma, Gazda* and *Maraton,* with maximum grain yields of 6.09, 7.28. 7.13 and 7.30 t ha^{-1}, respectively. These results show that the comparative evaluation of these four hybrids is impossible at a single plant density, due to the strong hybrid × plant density interaction. Although the yield potential of *Norma* and *Gazda* is similar to that of *Maraton*, the latter hybrid produces maximum grain yield at a much lower plant density. The results indicate that *Maraton* is capable of achieving high yields over a relatively wide plant density range of 6–9 plants m^{-2}, while *Norma* and *Gazda* only reached this level at 8.11 and 9.22 plants m^{-2}, respectively. This demonstrates that *Maraton* is substantially less dependent on the plant density. When comparing the hybrids in the same maturity group, it could be seen that the optimum plant density was 1.7 plants m^{-2} lower for *Maraton* than for *Gazda* (*Berzsenyi* and *Tokatlidis*, 2012).



Hybrid	Fitted quadratic function	Optimum plant density (plants m^{-2})	Potential yield (t ha^{-1})
Mara	$y=-0.0661x^2+0.989x+2.3954$ ($R^2=0.88$)	7.48	6.09
Norma	$y=-0.0733x^2+1.189x+2.4593$ ($R^2=0.99$)	8.11	7.28
Gazda	$y=-0.0475x^2+0.8758x+3.0957$ ($R^2=0.98$)	9.22	7.13
Maraton	$y=-0.0485x^2+0.7305x+4.5529$ ($R^2=0.96$)	7.53	7.30

Fig. 6. Grain yield responses of four maize hybrids to changes in the plant density at a N rate of 200 kg ha–1, averaged over 10 years

Effect of mineral fertilisation and plant density on the growth dynamics and yield of maize hybrids.

The effect of mineral fertilisation and plant density on the growth and yield of maize hybrids was investigated in a long-term three-factor experiment in the years 2001–2003. The mean values of the growth parameters and yield are presented in *Table 1* as a function of plant density, fertilisation, hybrid and year.

Treatments and years	NAR	AGR_{max}	AGR_{mean}	$ALGR_{max}$	Leaf $area_{max}$	Grain yield	
	g m^{-2} day^{-1}	g $plant^{-1}$ day^{-1}		cm^2 $plant^{-1}$ day^{-1}	cm^2 $plant^{-1}$	g $plant^{-1}$	t ha^{-1}
Plant density							
40 000	8.16	4.31	1.91	164.7	5740	141.0	5.38
70 000	7.61	3.33	1.49	142.6	5028	110.9	7.08
100 000	6.75	2.83	1.34	135.4	4917	85.4	6.81
Fertilisation							
A	7.57	3.01	1.35	125.5	4468	91.3	5.17
C	782	3.82	1.73	153.1	5432	130.9	7.48
E	712	3.63	1.67	164.2	5784	115.2	6.62
Hybrid							
Mara	7.45	3.23	1.48	140.8	4763	104.8	5.65
Norma	7.71	3.62	1.66	146.7	5317	118.9	6.56
Year							
2001	7.64	3.85	1.58	155.9	5844	140.5	8.63
2002	7.57	4.18	1.86	176.1	6107	119.3	7.93
2003	7.31	2.44	1.31	110.8	3734	76.6	2.71

As the plant density increased there was a reduction in the mean values of all individual plant growth parameters and in the grain yield (g $plant^{-1}$), due to the competition for resources. The grain yield of the plant stand (t ha^{-1}) was greatest at 70 000 plants ha^{-1}, with significant reductions in yield at higher or lower plant densities. In response to fertilisation the absolute growth rate (AGR), the net assimilation rate (NAR) and the grain yield reached their maximum values at the C fertiliser level ($N_{100}P_{48}K_{87}$) and the leaf growth parameters (maximum leaf area, leaf area growth rate) at the E fertiliser level ($N_{300}P_{144}K_{261}$). Hybrids with longer vegetation periods typically had higher values both for the growth parameters and the yield. There was a substantial year effect, with the lowest values of parameter means and yield in the droughty year of 2003, when the yield (t ha^{-1}) was only a third as much as in the favourable years (2001, 2002).

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Analysis of the sulphur content in corn in field experiments

Zoltán Győri[1], Norbert Boros[2]
[1] *Szent István University*
Institute of Regional economics and Rural Development
2100 Gödöllő, Páter Károly u. 1.
gyori.zoltan@gtk.szie.hu
[2] *University of Debrecen*
Department of Environmental and Chemical Engineering
4028 Debrecen, Ótemető u. 2-4.
nboros@eng.unideb.hu

Abstract

The effects of different NPK fertilisation doses on the sulphur contents in the grain yields of two corn hybrids were studied in a fertilisation and irrigation experiment. Furthermore, the dynamics of sulphur uptake of several hybrids was also determined. The determination of the sulphur content of the corn produced on a calcareous chernozem soil was done by using ICP-OES equipment. The readings obtained yielded data as regards the dynamics of sulphur uptake and the modifying effects of irrigation and NPK fertilisation.

Introduction

Sulphur is the fourth macro-element, which occurs in the crops analysed mostly in organic form as constituents of sulphurous amino acids (methionine, cystine, cysteine). In a number of cases these amino acids together with lysine are limiting amino acids.

In the practice of mineral fertilisation its use is primarily recommended for rape and also for winter wheat. In winter wheat the S – S cross binding of the components in gluten is very important for the formation of an adequate crumb of bread. The sulphur content in corn is a less-researched area in Hungary and so we tried to obtain as many data as possible on fertilisation, (one production location one hybrid) hybrids and the differences between growing locations (NLTFT, National Long-Term Fertilization Trials) not illustrated here.

There is a linear and weakly negative correlation between the average yield of corn, while between the average yield and the crude protein yield there is a strongly positive linear correlation, which is also true for the sulphur content *(Bálint 1977; Dudley et al. 1977; Bhatia and Rabson 1987; Sander et al. 1987)*. Also, the importance of sulphur fertilisation and the increasing methionine content are highlighted together with the relationship between the carotene content and the supply of sulphur. In the joint application of nitrogen and sulphur, others attribute the increase in the crude protein content to the effect of nitrogen *(Soliman 1989; O'Leary and Rehm, 1990)*. During dry growing seasons and at an approximately 50% decrease in the yield, the protein content of corn was higher, which also manifested itself in the sulphur content *(Lilburn et al. 1991)*.

In *T*ölgyesi's (1991) experiment the sulphur uptake of corn increased the most under the influence of a phosphorous – calcium – sulphur fertiliser treatment. Sulphur fertilisation increased the sulphur content of corn *(Reneau 1983, Buttrey et al. 1987a,b* and the nitrogen: sulphur ratio proved to be a good indicator of sulphur deficiency. *Allen (1979)* also gives a value of 0.14 g/100g for the sulphur content of corn. *Salunkhe et al. (1985)* also communicate a corresponding data. In contrast, *Loch and Nosticzius (1992)* have provided the following data: 0.17 g/100g. *Fageria et al. (1991)* publish the dynamics of the uptake of several elements of corn including sulphur and according to this publication the sulphur uptake is 40 kg/ha in the case of a yield of 30 t/ha dry matter.

Materials and methods

A field plot experiment was carried out at the Látókép Farm of University of Debrecen Center for Agricultural Sciences (*Ruzsányi, 1992*). It is located 15 km from Debrecen in Hungary. The soil of the experimental farm is a lowland calcareous chernozem soil with a deep layer of humus formed on a loess soil. The phosphorous and nitrogen supplies of the soil is medium, and it has a high sodium content (humus content 2.8-3.0 %, total nitrogen = 0.14-0.18%, AL-P_2O_5 = 130-200 mg/kg, AL-K_2O = 240-280 mg/kg). The depth of the humus layer is 70 – 90 cm. It has a pH value (KCl) of 6.2. There cannot be indicated any deficiency in micro elements. The groundwater level is between 6 and 8 meters.

The main plots in the individual experimental fields at Látókép were divided without repetition according to the irrigation alternatives and cultivation methods while the subplots were formed according to fertilisation levels *(Table 1)* in four repetitions. The size of each plot was 46 m^2. In the poli-factorial experiment there are different crop rotations, such as legumes – wheat – corn in tri-culture, wheat – corn in bi-culture and corn monoculture. Irrigation was conducted at a water norm of 80 mm.

Treatments	N	P_2O_5	K_2O
1	0	0	0
2	60	45	45
3	120	90	90
4	180	135	135
5	240	180	180

Table 1. The fertilization treatments applied in the experiments (kg/ha)

It is a well-known fact that fertilisers contain several accompanying elements, one of which is sulphur. Superphosphate contained 10.9% of it and considering this the sulphur content of 0.13% in nitrogenous fertilisers and that of 0.23% in potassium-chloride is almost negligible. According to the above the fertilisation levels applied supply the following amounts of sulphur to the soil 24.9; 49.8; 74.6; 99.5 kg/ha. In comparison to the 2.1 mg/kg of sulphate-sulphur obtained in the control treatment the medium level treatment resulted in 23.3 mg/kg while the highest level of NPK yielded 38.9 mg/kg sulphate-sulphur.

The existing analytical capacity and performance allowed us to take samples for sulphur content measurements regularly from both the soil and plant parts.

After purifying and drying the samples when necessary, they were ground with Retsch Sk-1, and Sk-3 type grinders to a particle size of 1-2 mm. In order to determine the element contents of the plant samples the $HNO_3 + H_2O_2$ digestion method was used while the analytical determination was conducted by using an inductively coupled plasma-emission spectrometer (ICP-OES) *Kovács (1998)*. The adequate control of the measurements was ensured by the use a certified reference material (CRM) of wholemeal wheat flour marked BCR CRM 189.

Results and discussion

According to the results of our measurements the increasing fertilizer doses increased the sulphur contents of the corn grains but this influence is significant only depending on the cropping year and the previous crop and only manifests a trend. In the case of the hybrid Pannónia the sulphur-content increasing effect of fertilisation is statistically proven only in di-culture, while in the hybrid Furio it is proven in tri-culture only. The influence of irrigation brought about decreases in the sulphur contents in the hybrids Pannónia and Furio in monoculture and in all the three crop rotations, respectively. In this latter case, however, the yields became several times higher under the influence of irrigation *(Table 2)*.

			Treatment 1	**Treatment 2**	**Treatment 3**	**Treatment 4**	**Treatment 5**	**Average**
Pannónia	monoculture	Non-irrigated	798	838	853	893	900	**856**
		Irrigated	810	780	798	800	758	**789**
		Average	**804**	**809**	**825**	**846**	**829**	
		$SzD_{5\%}$ between irrigations: 64***						
		$SzD_{5\%}$ between treatments: 55						
Pannónia	bi-culture	Non-irrigated	748	683	718	868	898	**783**
		Irrigated	748	775	748	840	810	**784**
		Average	**748**	**729**	**733**	**854**	**854**	
		$SzD_{5\%}$ between irrigations: 40						
		$SzD_{5\%}$ between treatments: 42***						
Pannónia	tri-culture	Non-irrigated	790	820	818	838	840	**821**
		Irrigated	853	830	845	803	870	**840**
		Average	**821**	**825**	**831**	**820**	**855**	
		$SzD_{5\%}$ between irrigations: 21						
		$SzD_{5\%}$ between treatments: 35						

			Treat-ment 1	Treat-ment 2	Treat-ment 3	Treat-ment 4	Treat-ment 5	Aver-age
		Non-irrigated	1151	1184	1190	1101	1163	**1158**
		irrigated	975	914	927	955	934	**941**
Furió	monoculture	**Average**	**1063**	**1049**	**1059**	**1028**	**1048**	
		$SzD_{5\%}$ between irrigations: 77***						
		$SzD_{5\%}$ between treatments: 54						
		Non-irrigated	1134	1097	1224	1254	1187	**1179**
		Irrigated	897	939	1039	1004	988	**974**
Furió	bi-culture	**Average**	**1015**	**1018**	**1132**	**1129**	**1087**	
		$SzD_{5\%}$ between irrigations: 71***						
		$SzD_{5\%}$ between treatments: 112						
		Non-irrigated	1014	1311	1334	1351	1252	**1253**
		Irrigated	861	924	971	969	933	**932**
Furió	tri-culture	**Average**	**938**	**1117**	**1153**	**1160**	**1093**	
		$LSD_{5\%}$ between irrigations: 37***						
		$LSD_{5\%}$ between treatments: 58***						

Table 2. The influence of mineral fertilization and irrigation on the sulphur contents of corn grains (mg/kg)

In the case of an element that is analyzed so infrequently and if and when there is an adequate capacity for measurement at our disposal, it is justified to ask the question what is the dynamics of the uptake of this element by the plant like. To answer the question there is a need for a series of data that is to throw enough light on the concentration of the element and about its amount taken up until the given phenophase.

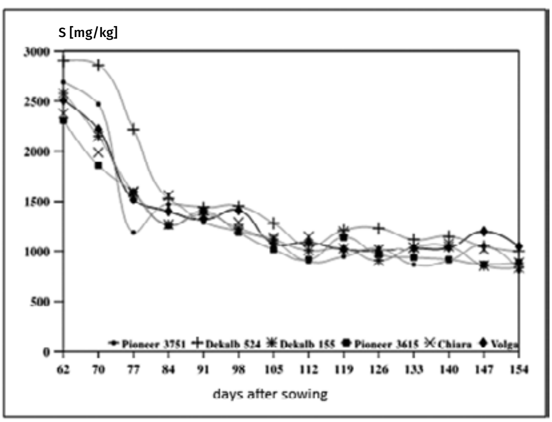

Figure 1. Changes in the sulphur contents of different corn hybrids during the growing season

Figure 1 shows a series of data on the samples taken frequently or less frequently (weekly, at critical phenophases) where the changes in the sulphur contents of the hybrid can be followed. This series of data comes from the analyses of corn hybrids where even the supply of water was not a limiting factor. According to the analytical data of the samples the sulphur concentrations from 2500 mg/kg in the dry matter in the early phenophases (ten leaves) decrease to 1000 mg/kg in the mature plants and remain at this value in the stages of grain formation and ripening as well. According to our results the supply of sulphur was secure continuously even in the periods of the intensive formation of dry matter.

We obtained very important data for establishing the sulphur balance of corn since we did an analysis of the whole plant. The above the ground masses of the different hybrids vary and so the different hybrids remove different amounts of sulphur from the soil. The amounts of sulphur removed by the plants with PI3615 15 kg/ha, Volga 20 kg/ha, Chiara 25 kg/ha) different dry matter productions vary between 15 and 25 kg/ha.

According to our experimental data the calculated amount of sulphur removed together with the grain yield was between 8 and 13 kg/ha (in the case of grain yields of 7-10 t/ha) *(Table 3).* Considering the total amount of 15-25 kg/ha of sulphur removed by the whole plant these data are to be included in the balance together with the way the stalk is utilized.

At the same time, attention has to be paid to the fact that in the case of a grain yield of 5 t/ha the amount of sulphur removed from the soil is also around 15 kg/ha, which can be made up for by the equivalent of 30-35 kg/ha P_2O_5 superphosphate.

			Average yield (kg/ha)	**Average S (g/kg)**	**Amount of sulphur removed with the yield (kg/ha)**
Pannónia	monoculture	non-irrigate	10968	0.856	9.39
		irrigated	11849	0.789	9.35
	bi-culture	non-irrigated	12257	0.783	9.59
		irrigated	11638	0.784	9.12
	tri-culture	non-irrigated	12210	0.821	10.02
		irrigated	12524	0.840	10.52
Furió	monoculture	non-irrigated	1666	1.158	1.93
		irrigated	10668	0.941	10.04
	bi-culture	non-irrigated	7565	1.179	8.92
		irrigated	13027	0.974	12.68
	tri-culture	non-irrigated	3824	1.253	4.79
		irrigated	12936	0.932	12.05

Table 3. The amount of sulphur removed by grain yields of corn

The average value of the sulphur contents of the hybrids grown is also important in the case of corn. *Figure 2* gives an overall picture of these data, according to which the average sulphur content was 1.1 g/kg ± 0.1 on the average of the corn hybrids analyzed. Thus it is recommended to calculate with these data in giving technical advice on both nutrient replacement and animal feeding.

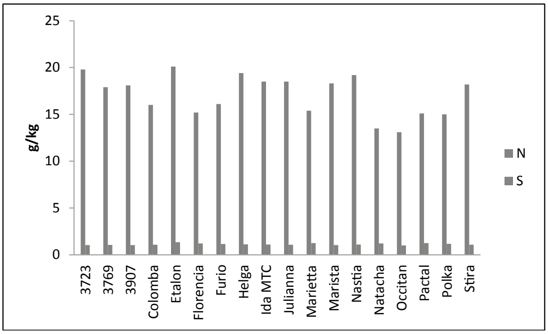

Figure 2. Nitrogen and sulphur contents in different hybrids (g/kg)

With the help of the measurement data for nitrogen and sulphur we can obtain information as regards the ratios of these two elements. The international technical literature provides critical data for this ratio and according to our investigations the nitrogen-sulphur ratio in corn is mostly 15-16 *(Table 4)*, which suggests an adequate or good supply of sulphur.

			Average N (g/kg)	Average S (g/kg)	N:S
Pannónia	monoculture	on-irrigated	13.06	0.856	15.25
		irrigated	12.21	0.789	15.48
	bi-culture	non-irrigated	12.66	0.783	16.18
		irrigated	12.24	0.784	15.61
	tri-culture	non-irrigated	13.32	0.821	16.22
		irrigated	12.77	0.840	15.20
Furió	monoculture	non-irrigated	18.28	1.158	15.79
		irrigated	14.04	0.941	14.92
	bi-culture	non-irrigated	15.68	1.179	13.30
		irrigated	14.07	0.974	14.45
	tri-culture	on-irrigated	14.20	1.253	11.34
		irrigated	13.66	0.932	14.66

Table 4. The N:S ratio in corn grains

The differences between the hybrids are shown by the fact that this ratio is below 15 in the case of Furio since in the year analyzed the fluctuation in the yields was very high *(table 3)*. At the same time, the amounts removed in the cases of irrigated treatments were the highest, over 12 kg/ha.

The correlations between nitrogen-sulphur contents were also carried out. The findings show that the sulphur contents increase linearly in the case of all the three crop rotations (y=0.21x+0.71; y=0.62x+0.16; y=0.51x+0.38).

The sulphur getting into the soil through NPK fertilizer application increased not only the sulphur content of the soil but that of the corn as well in proportion with the protein content. On the average of the hybrids analyzed the sulphur content of the grain was 1.1 kg/ha. According to the data we obtained the amount of the sulphur removed was less than the figure published in the paper by *Fageria et al. (1991)*. The nitrogen-sulphur ratio varied between 15 and 16, which corresponded to the data found in the technical literature and suggested a good supply of sulphur. The effect of irrigation can be judged by considering the yields and the changes in the nitrogen contents.

Acknowledgement

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Special agrotechnical crop models for Martonvásár maize genotypes

Péter Pepó, Mihály Sárvári
University of Debrecen CAAES
Institute of Crop Sciences
4032 Debrecen, Böszörményi út 138.
pepopeter@agr.unideb.hu, sarvari@agr.unideb.hu

Abstract

We have investigation the herbicide tolerances (post-emergent, optimal and late treatments) of various maize hybrids of different genotypes on Hajdúság chernozem soil. Our studies confirmed that each herbicide treatment is a stress for the growing and development of maize, which is reflected by the yield result as lesser-greater yield decrease. The herbicide tolerances of the maize hybrids were influenced by the genotype, the active substance and the application time of the herbicide, which confirms the importance of the hybrid specific herbicide use.

One can conclude that there are very tight interactions between the agrotechnical factors that influence yield. Hybrid specific production technology has to be applied; the fertilizer dose, the sowing time and plant number have to be optimized for the given hybrid.

Introduction

Cereals play crucial role in Hungarian crop production. Among them, the most important ones are winter wheat and maize. During the past decades, Hungarian maize production went through a considerable change. At the beginning of the 1960s, the extreme rapidly introduced inbred hybrids provided adequate base for the wide spreading and use of inputs of industrial origin. From the 60s, chemical fertilization use began to grow rapidly, chemical weed control widely spread and the uses of modern machines and tools were introduced. In the production technology of maize, *Győrffy (1976)* evaluated the roles of the different agrotechnical factors as follows: fertilization 27%, variety 26%, care 24%, plant number 20%, and deep cultivation 3%. Besides the interactions between the production factors, it is important to guarantee the positive interactions between the agrotechnical factors due to the climate change (*Pepó and Sárvári 2013*, *Sárvári et al. 2008*).

Weed control is one of the most important elements of the production technology of maize. Modern weed control has to be based on the integrated principles (*Berzsenyi 2000*, *Széll et al. 1985*, *Chui et al. 1997*). The importance of early and normal post-emergent herbicide using increased recently in Hungarian maize production, in case of which the extent of weediness and weed composition is known. The effect and efficiency of post-emergent weed control depend on the environmental conditions (*Tapia et al. 1997*, *Fayolle 1996*) and the sensitivity of the hybrids (*Bónis et al. 2000*, *Hart and Wax 1999).* In recent years, the number of state-certified grain and silage maize hybrids increased to a considerable extent, altogether their number is close to 400. These different genotypes can significantly vary in terms of their agrotechnical reactions, thus, they can perform different herbicide reactions.

The Martonvásár Agricultural Institute has been dealing with the selection of field crop species for six decades. Their winter wheat varieties and maize hybrids are very popular both in Hungary and abroad. The varieties of the Agricultural Institute are tested by universities and research institutes for adaptation to different ecological conditions. Our PhD students also investigate the agrotechnical reactions of the Martonvásár maize hybrids (e.g. NPK fertilizer reaction, connection between sowing time and crop safety, the plant density increase of maize hybrids, etc.).

As an effect of climate change, weather extremes increase. The interactions between the agrotechnical factors enhance, and it is reasonable to optimize them.

Materials and methods

Weed control experiment:

The small plot field experiments were carried out at the Látókép Experimental Farm of the University of Debrecen, CAAES, Institute of Crop Sciences, on calcareous chernozem soil.

The forecrop of the experiment was winter wheat. The agrotechnical elements fulfilled the requirements of modern production.

In the experiment, the following Martonvásár hybrids were studied for herbicide sensitivity: Gazda, Maraton, Norma.

The sowing of the experiment was conducted on 22 April 2002, with 68.000 ha^{-1} uniform germ number.

In the experiment, the following herbicide treatments were set:

1. Weedy control
2. Hoed control
3. Escort 4.0 L ha^{-1} (early post)
4. Merlin SC 0.22 L ha^{-1}+Dezormon 1.0 L ha^{-1} (early post)
5. Escort 4.0 L ha^{-1} (normal post)
6. Merlin SC 0.22 L ha^{-1}+Dezormon 1.0 L ha^{-1} (normal post)
7. Motivel 1.0 L ha^{-1}+Cambio 3.0 L ha^{-1} (normal post)
8. Titus 25 DF 40 g ha^{-1}+Callisto 0.25 L ha^{-1}+Trend 0.1% (normal post)
9. Motivel 1.0 L ha^{-1}+Cambio 0.25 L ha^{-1} (late post)
10. Titus 25 DF 40 g ha^{-1}+Callisto 0.25 L ha^{-1}+Trend 0.1% (late post)

The experimental treatments were carried out at the following dates and developmental stages:

Manual hoeing:	4 May 2002	
	2 June 2002	
Early post-emergent:	7 May 2002	2-3-leaf stage
Normal post-emergent:	14 May 2002	5-leaf stage
Late post-emergent:	20 May 2002	7-8-leaf stage
The harvest was carried out on 9 October 2002 by Sampo plot harvester.		

Within the research project, we have conducted population dynamics, agronomical, plant health, weed dynamics, phytotoxicity and crop forming element studies; we have measured the yield results and the grain moisture content values at harvest.

Agrotechnical experiments:

Between 2004 and 2006, we investigated the NPK fertilizer reaction, productivity and crop stability of Mv Maraton (FAO 450) and Mv Vilma (FAO 510) on calcareous chernozem soil. In addition, we tested the plant density increase of Mv Maraton and the effect of sowing time on yield and grain moisture content at harvest, between 2005 and 2007.

During the NPK fertilization experiment, besides the control (without fertilization), we applied five different fertilizer doses. The basic treatment was N 40 P_2O_5 25, K_2O 30 kg ha^{-1} active substance, while the maximum dose was fivefold.

In the experiment on sowing time, we set the experiment on 8 April, 20 April and 15 May. The nutrient treatment in the sowing time and plant density increasing experiments were equally N 120, P_2O_5 90, K_2O 110 kg ha^{-1} active substance.

In the plant density increasing experiment, stock densities were 45, 60, 75, and 90 thousand plant ha^{-1}. The plot sizes were a gross of 25.2 m^2, net 16.8 m^2. Soil preparation and plant care were carried out in accordance with the agronomic conditions.

The difference of the annual precipitation amount during the past 23 years was -525 mm, compared to the 30-year average of 565.3 mm. Sixty-one percent of the years was droughty *(Figure 1.)*.

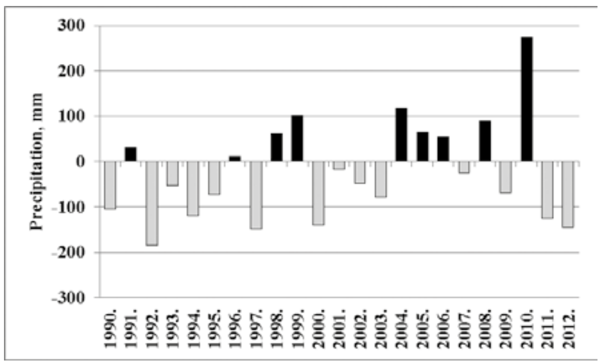


Figure 1. Deviation of yearly precipitation from the 30 years average (Debrecen, 1990-2012)

Results and discussion

Weed control experiment:

We have investigated the sensitivity of Martonvásár maize hybrids of different genotypes to herbicides of various active substances besides mixed weed composition (*Echinocloa ssp.*, *Setaria ssp.*, *Hibiscus trionum*, *Xanthium ssp.*, *Amaranthus ssp.*, other) on Hajdúság chernozem soil. We have also studied the complex effects (on weeds and hybrids) of herbicides applied at optimal and later-than-optimal times.

Our experimental results confirmed that by the progression of the vegetation period, the weed coverage values – depending on herbicide treatment – gradually increased *(Tables 1, 2, 3)*. To the end of the vegetation period, the weed coverage values of the weedy control (Treatment No. 1) varied between 7.8 and 8.6%, while those of the hoed control performed 4.9-5.1%. Following herbicide treatments, the weed coverage significantly decreased (measurements at 14/05, 20/05, 02/06). At the end of the vegetation period, the extent of weed coverage was determined partially by the active substance of the herbicide, partially the time of its application, and partially the presence and development of the cultivated genotypes (in treatment No. 5, the dying of all hybrids). The herbicide treatments were characterized as of adequate efficiency, which was confirmed by the weed surveys performed before harvest (measurement time of 29/09):

Treatment No. 3	4.9-5.1% weed coverage
Treatment No. 4	1.5-1.7% weed coverage
Treatment No. 6	1.5-1.7% weed coverage
Treatment No. 7	1.1-1.4% weed coverage
Treatment No. 8	1.0-1.1% weed coverage
Treatment No. 9	2.2-2.3% weed coverage
Treatment No. 10	2.1-2.2% weed coverage

During treatment No. 5, which was a late one of extreme active substance (Escort 4.0 L ha^{-1}, normal post-emergent), all maize hybrids died 2-3 weeks after the spraying. Due to the lack of the coverage by the cultivated crop, a weed coverage exceeding the control treatments was formed (at the measurement time of 29/09, the weed coverage was 12.9-14.7%). One week after the herbicide treatments, we have determined the extents of phytotoxicity of the treatments of different application times (early, normal, late post-emergent) in the maize hybrids of different genotype *(Tables 1, 2, 3)*. Our experimental results confirmed that every herbicide caused phytotoxicity in every hybrid to lesser-greater extents. The extent of phytotoxicity significantly differed depending on the

- active substance of the herbicide
- the time of application
- the herbicide tolerance of the hybrid.

Herbicide treatment	Weed covering (%)*					Phytotoxicity (%)**		
	07/05	14/05	20/05	02/06	24/09	14/05	20/05	02/06
1.	0.4	1.0	2.7	3.9	7.8	0	0	0
2.	0.4	0.7	1.4	3.1	5.1	0	0	0
3.	0.5	0.6	0.5	1.8	5.0	24	34	56
4.	0.5	0.5	0.5	0.7	1.5	0	2	4
5.	0.4	0.9	2.5	0.3	14.7	-	49	-
6.	0.4	1.1	2.4	0.4	1.6	-	2	9
7.	0.4	1.2	2.5	0.1	1.4	-	5	15
8.	0.4	1.1	2.6	0.1	1.1	-	10	21
9.	0.5	0.9	2.8	2.1	2.3	-	-	21
10.	0.5	1.1	2.9	2.0	2.2	-	-	29

** Weed covering % m^{-2} soil surface*
*** Phytotoxicity on leaf area %*

Table 1. Effect of herbicide using on the weed covering and phytotoxicity. Gazda (Debrecen, 2002)

Herbicide treatment	Weed covering (%)*					Phytotoxicity (%)**		
	07/05	14/05	20/05	02/06	24/09	14/05	20/05	02/06
1.	0.4	0.9	2.6	4.1	8.4	0	0	0
2.	0.4	0.6	1.4	2.9	4.9	0	0	0
3.	0.5	0.4	0.4	1.7	4.9	6	8	10
4.	0.4	0.4	0.4	0.7	1.7	0	6	8
5.	0.5	1.1	2.5	0.4	13.1	-	42	-
6.	0.5	1.2	2.6	0.3	1.7	-	4	5
7.	0.6	1.1	2.5	0.1	1.2	-	8	17
8.	0.6	1.0	2.4	0.2	1.0	-	2	14
9.	0.5	0.9	2.9	2.1	2.3	-	-	22
10.	0.5	1.1	2.8	1.9	2.1	-	-	20

** Weed covering % m^{-2} soil surface*
*** Phytotoxicity on leaf area %*

Table 2. Effect of herbicide using on the weed covering and phytotoxicity. Maraton (Debrecen, 2002)

Herbicide treatment	Weed covering (%)*					Phytotoxicity (%)**		
	07/05	14/05	20/05	02/06	24/09	14/05	20/05	02/06
1.	0.5	1.0	2.6	4.1	8.4	0	0	0
2.	0.6	0.6	1.4	2.9	5.0	0	0	0
3.	0.6	0.5	0.4	1.9	5.1	26	42	68
4.	0.5	0.4	0.4	0.8	1.5	0	9	21
5.	0.5	1.1	2.6	0.3	12.9	-	47	-
6.	0.5	1.2	2.5	0.3	1.5	-	4	7
7.	0.6	1.0	2.6	0.2	1.1	-	8	14
8.	0.5	1.2	2.4	0.2	1.4	-	5	11
9.	0.6	1.1	2.8	2.0	2.2	-	-	12
10.	0.5	1.0	2.7	1.9	2.2	-	-	25

** Weed covering % m^{-2} soil surface*
*** Phytotoxicity on leaf area %*

Table 3. Effect of herbicide using on the weed covering and phytotoxicity. Norma (Debrecen, 2002)

The highest phytotoxicity was caused by the late (normal post) application of the herbicide of special active substance (Escort = imazamox + pendimethalin). In the fifth treatment, the total lethality of all hybrids was experienced 2-3 weeks after the treatment. This herbicide caused considerable phytotoxicity applied even at optimal time (treatment No. 3, 10-68% phytotoxicity), which was observed in the yield too. Among the hybrids, Norma and Gazda were extremely sensitive to Escort, while Maraton showed a relatively favourable tolerance. The other herbicide treatments caused lower phytotoxicities to the hybrids (phytotoxicity below 20%, except late application of rimsulfuron active substance combination [treatment No. 10], which resulted in phytotoxicity above 20% in the case of certain hybrids).

In our experiment, we have achieved favourable yield results. Except the extreme herbicide treatments, yield varied between 7 and 13 t ha^{-1}, depending on the genotype, the application time and active ingredients of herbicides. The most favourable yield results were performed by the hybrid Maraton (yield results of 10-13 t ha^{-1}) (*Table 4*).

Herbicides	Yield (14 % grain moisture) kg ha^{-1}		
	Gazda	Maraton	Norma
1.	9 447	12 531	9 445
2.	10 514	13 022	10 178
3.	7 213	11 924	3 997
4.	10 263	10 152	8 670
5.	-	-	-
6.	9 812	12 166	10 097
7.	10 062	10 690	10 136
8.	9 513	12 698	10 049
9.	9 281	10 874	10 047
10.	8 802	10 835	9 812

Table 4. Effect of herbicide on the yield of maize genotypes. (Debrecen, 2002)

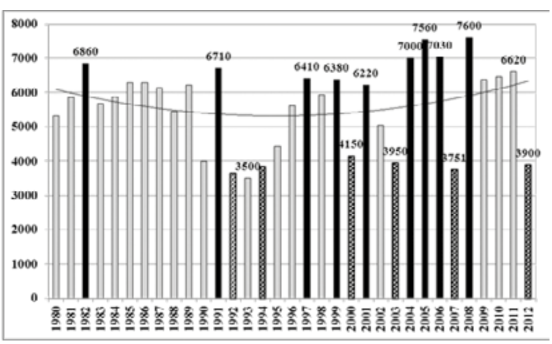


Figure 2. Country-average yield (kg ha^{-1}) of maize in Hungary (1980-2012, by Erzsébet Kiss)

According to our experimental results, the hybrid tolerances (against active substance, application time) of the maize hybrids of different genotypes significantly differed. In the case of the studied hybrids, Norma possessed favourable hybrid tolerance, while Gazda and Maraton were considered as sensitive.

Agrotechnical experiments:

In Hungary, during the recent decades, the country yield average reached 6.5-7.5 t ha^{-1} only in rainy years, while in droughty ones, it fall below 4 t ha^{-1} *(Figure 2)*. This means that yield fluctuation reaches 50-60%.

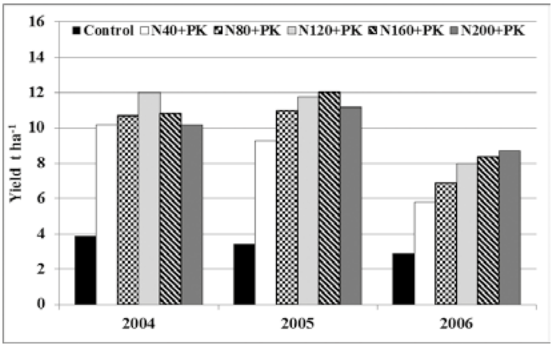
The yield averages of the Martonvásár hybrids – in long-term experiment – were above 10 t ha^{-1}, even in the droughty 2007. They can be considered as good genetic bases, alongside with other good traits.

The connection between chemical fertilization and the yields of hybrids is depicted in *Figure 3*. Without fertilization, Mv Maraton performed a yield of 3-4 t ha^{-1}. As a result of fertilization (N40+PK), its yield increased by 2-3 t ha^{-1}. Its yield maximum was 12 t ha^{-1} in 2004-2005, while its agro-ecological fertilizer optimum was N 120, P_2O_5 75, K_2O 90 kg ha^{-1} active substance.

The yield of Mv Vilma reached 14-15 t ha^{-1} in 2004-2005, it is a hybrid of excellent productivity and of good fertilizer reaction. The lower yield results of 8-9 t ha^{-1} in 2006 was caused by the ice storm on 22 July *(Figure 4)*.

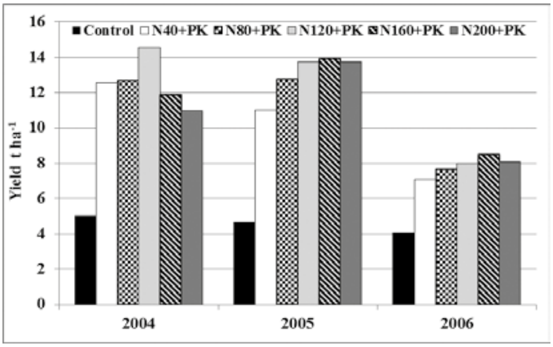
Mv Maraton reached its yield maximum of 11-12 t ha^{-1} in the first sowing time (8 April) in 2005 and 2007, while in 2006 at the 20 April (second) sowing *(Figure 5)*.

Mv Maraton was a hybrid of good individual production, susceptible to be multi-eared. In the above-than-average moist year of 2005, it reached it maximum yield of 13 t ha^{-1} at stock density of 90 thousand plant ha^{-1}, while in the unfavourable cropyears of 2006 and 2007, its yield at 75 thousand plants was 9-11 t ha^{-1} *(Figure 6)*.



LSD5% (2004 year) 0.68 t ha^{-1}, LSD5% (2005 year) 0.48 t ha^{-1}, LSD5% (2006 year) 0.33 t ha^{-1}

Figure 3. Effect of cropyear and fertilization on the yields of Mv Maraton hybrid (Debrecen, 2004-2006, by Nóra El Hallof)



LSD5% (2004 year) 0.68 t ha^{-1}, LSD5% (2005 year) 0.48 t ha^{-1}, LSD5% (2006 year) 0.33 t ha^{-1}

Figure 4. Effect of cropyear and fertilization on the yields of Mv Vilma hybrid (Debrecen, 2004-2006, by Nóra El Hallof)

The results confirm that the Martonvásár hybrids provide outstanding biological bases.

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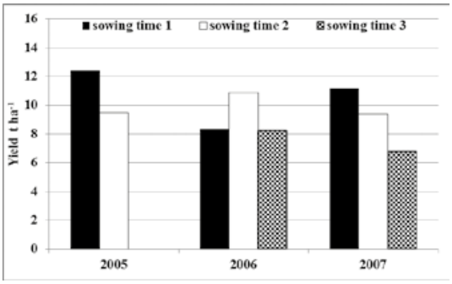
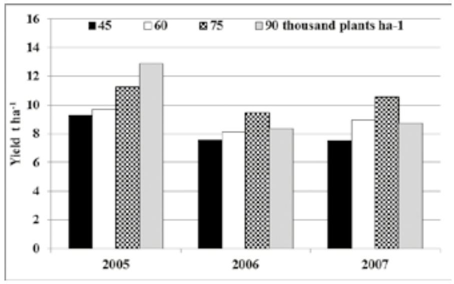


LSD5% (2005 year) 5.37 t ha^{-1}, LSD5% (2006 year) 1.84 t ha^{-1}, LSD5% (2007 year) 1.43 t ha^{-1}

Figure 5. Effects of cropyear and sowing time on the yields of Mv Maraton hybrid (Debrecen, 2005-2007, by Zsuzsa Molnár)



LSD5% (2005 year) 0.85 t ha^{-1}, LSD5% (2006 year) 0.62 t ha^{-1}, LSD5% (2007 year) 0.54 t ha^{-1}

Figure 6. Effects of cropyear and plant density on the yields of Mv Maraton hybrid (Debrecen, 2005-2007, by Zsuzsa Molnár)

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The impact of drought as one of the strongest stress factors on the yield of maize (Zea mays L.)

Adrienn Széles, Péter Ragán, János Nagy
University of Debrecen, Centre for Agricultural and Applied Economic Sciences
Institute for Land Utilisation, Regional Development and Technology
H-4032 Debrecen, Böszörményi út 138.
szelesa@agr.unideb.hu

Abstract

Six different N doses (0-150 kg ha^{-1}) were applied in a field experiment in Debrecen on calcareous chernozem soil with the aim to compare the chlorophyll (Chl) content of maize (*Zea mays* L.) as measured by the SPAD-502 meter with the nitrate N content of the soil and to conclude to the level of N supply of the soil, as well as the grain yield of maize in dry and wet crop years.

In dry crop years, we detected a moisture profile with increasing moisture saturation from the surface (8.5–9.5 v/v%) towards the deeper layers (15–20 v/v%). In wet crop years, the moisture profile showed opposite characteristics.

A significant part of the nitrate N applied before sowing accumulated in the 0-0.2 m layer of the soil profile and its quantity did not significantly reduce during the growing season. No mobility towards deeper layers was detected. On the contrary, the accumulation zone was located in the deeper, 0.4-0.6 m deep layers in wet years. In parallel, the easily available mineral N stock of the soil greatly decreased until silking.

The CMR value determined the easily available N stock of the soil and yield at a 0.1% significance level in the R1 development phase in dry crop years. In wet crop years, yield can be reliably predicted ($P<0.001$) and the N stock of the soil can also be concluded =$P<0.01$) even at the V12 development phase.

Introduction

The change in climatic factors (e.g. temperature, precipitation) and the more frequent weather extremities (drought, floods, storms) greatly affect crop yields (*Láng 2005, Nováky 2007*). The amount of precipitation in the summer period decreases by 10-30 mm between 2011 and 2020, while this reduction could even be 40–50 mm between 2031–2040 (*Gaál* 2007).
Averaged over 2000-2012, the quantity of harvested maize was 7.049 in a year in Hungary and the most extreme years were 2005 (9.050 million tons) and 2007 (4.027 million tons). In 2007, maize was only 3.7 t ha^{-1}, one of the weakest results in the recent years. As a result of the favourable weather, the average yield in 2008 (7.4 t ha^{-1}) was double as much as that of the preceding year. In 2011, 7.992 million tons of maize was harvested from the 1.230 million hectares of maize cropland. Due to the unfavourable

weather in 2012, yield was 41% lower (4.741 million tons) than that of 2011 and the average yield (3.9 t ha^{-1}) also decreased by 39%.

In order to avoid yield fluctuations which depend on crop years, preserving and utilising precipitation will become inevitably necessary. Also, cultivation, rational nutrient replenishment adapting to crop needs and their nutrient uptake dynamism, as well as the production of drought-resistant varieties are equally as important and related factors (*Rátonyi 1998, Gyuricza and Birkás 2000, Marton and colleagues 2003, Csete 2005, Jolánkai and Birkás 2010, Nagy 2011, Birkás and colleagues 2012, Ványiné Széles and colleagues 2012*).

Warming greatly affects the incorporation of nutrients. In permanent drought, the incorporation of fertiliser decreases and a yield reduction impact will occur. In the case of drought, overabundant nutrient replenishment results in unfavourable nutrient concentration (*Széll and colleagues 2005, Víg and colleagues 2010, Nagy 2012*).

The aim of this paper is to evaluate the impact of dry and wet crop years on N fertilisation, the water and nitrate N content of the soil, the chlorophyll content of maize and maize yield.

Materials and methods

The examinations were carried out at the Látókép Experiment Site of the University of Debrecen, Centre for Agricultural and Applied Economic Sciences on loess-based, moderately heavy calcareous Great Plain chernozem with deep humus layer in a multifactoral small plot experiment with strip plot design and four replications during the growing seasons of the period between 2004-2011. Plot sizes were 15 m^2.

Weather was evaluated on the basis of the data measured and logged by the automatic weather station set up at the experiment site. The measured data include air and soil temperature (°C), relative humidity (%), wind speed (m s^{-1}), incoming radiation (W m^{-2}) and precipitation.

The average pH_{KCl} of the soil is 6.6 (slightly acidic) which is optimal from the aspect of the nutrient uptake of plants. The Arany plasticity number of the upper (20 cm) layer of soil is 39 and the amount of water-soluble salts (anions and cations) was 0.04% altogether. The carbonic chalk content was around 0% in the upper 80 cm layer and 12% from 100 cm down. The organic matter content in the upper 20 cm of the soil was 2.3% and it does not exceed 1.00% at the 120 cm depth. The potassium supply of the soil is favourable, while its P supply is average.

In accordance with the bibliographical references (*Yadava* 1986, *Berzsenyi* and *Lap* 2005, the relative chlorophyll concentration of the maize leaf was measured using a SPAD-502 (Minolta, Japan) portable chlorophyll meter. The measurements were started every year even at the vegetative 6-leaf-stage (V6) of maize (*Ritchie and colleagues* 1997). Further CMR (Chlorophyll Meter Readings) data were obtained at the 12-leaf-stage (V12) and in the reproductive period (R1).

In order to evaluate the level of easily available mineral N supply of the soil, disturbed soil samples were taken three times (at the V6, V12 and R1 phenophases) from the 0-120 cm soil profile, i.e., the rooting depth of maize. The analysis of the NO_3-N content of the samples was performed with a Spectroquant® Nova 60 A photometer, using a Spectroquant® nitrate test. The quantity of N available for plants was determined on the basis of the nitrate N content (mg kg^{-1}), bulk density (g cm^{-3}) and layer thickness of the soil.

The changes in the soil moisture cycle were monitored with soil moisture probes (CS 615 Water Content Reflectometer, Campbell Scientific, Inc.) which are based on the TDR principle. The change in soil moisture (v/v%) was monitored on a daily basis in the 0–120 cm soil profile from sowing to harvesting.

A general linear model (GLM) was used to show the effect of treatments on the moisture stock and nitrate-N content of the soil, the chlorophyll content of maize and maize yield (*Huzsvai,* 2008). The 5% significant difference ($LSD_{5\%}$) was determined in order to compare the means of treatment. During the multiple comparisons, the confidence intervals were corrected with Duncan's method in order to avoid alpha error. The quality parameters and yield within the homogeneous group did not differ from each other at the 5% level of significance. A regression analysis was performed in order to determine the correlations between the nitrate-N of the soil, the chlorophyll (Chl) content of maize and maize yield. Statistical evaluation was performed using SPSS for Windows 13.0.

Results and discussion

The lowest *soil moisture* results were measured in the 1.2 m soil profile in dry crop year in the 0–0.15m layer of the soil (8.5–9.5 v/v%). In deeper layers, the moisture content was increasing with depth. The significant ($p<0.05$) moisture difference (10–12 v/v%) measured at the 11–12-leaf-stage of maize was constantly decreasing by the end of the growing season as the moisture content of the soil was being exhausted (*Figure 1*). In wet crop years, the moisture content of the soil profile was the highest in the 0.12–0.30 m layer (36–38 v/v%) and it was constantly decreasing with depth (12–15 v/v%) (*Figure 2*). The typical "bulge" in the moisture curve of the 0.15–0.30 m soil layer is the result of the soil compaction and bulk density increase caused by autumn primary tillage and spring seedbed preparation.

In dry crop years, the *nitrate N concentration* of control plots was between 0.6–2.7 mg kg^{-1}. As a result of the N fertilisation performed before sowing, the nitrate N found in the soil was accumulated in the 0.4 m profile near the surface. The maximum value of accumulation was detected in the 0–0.05 m layer and the nitrate N concentration of the soil strongly decreased with the increase of depth. The

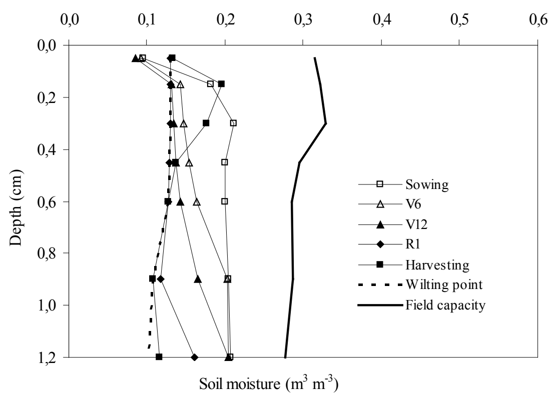

Figure 1. Soil moisture in a dry crop year

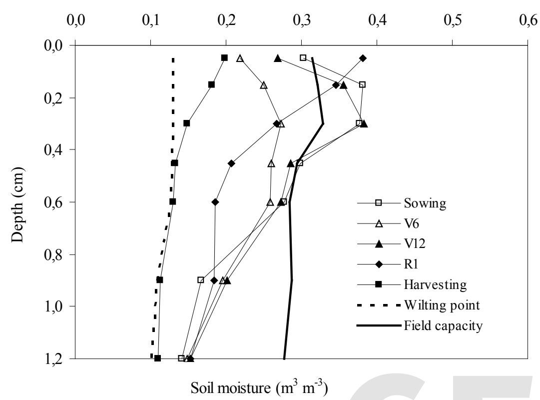

Figure 2. Soil moisture in a wet crop year

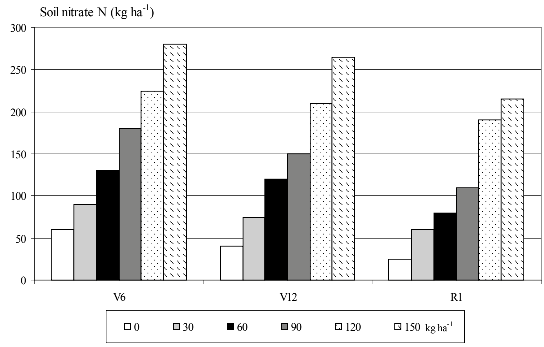

Figure 3. Soil nitrate N content available to crops in a dry crop year

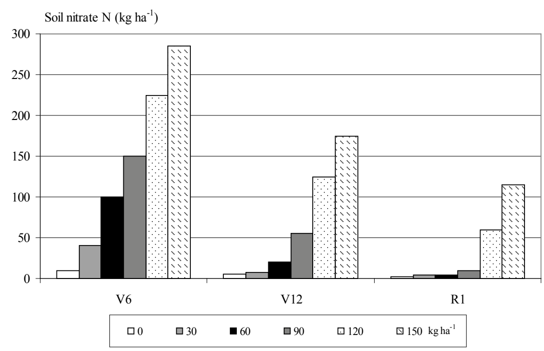

Figure 4. Soil nitrate N content available to crops in a wet crop year

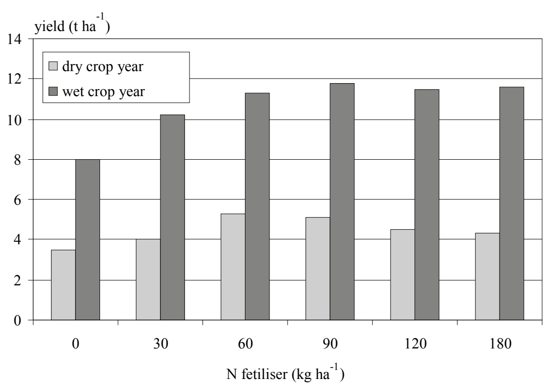

Figure 5. Maize yield as a result of fertiliser treatments

nitrate N quantity measured in the accumulation level was between 11.7–23.0 mg kg^{-1} even in the case of moderate fertiliser doses (30–60 kg ha^{-1} N) and between 33.9–56.6 mg kg^{-1} (90–150 kg ha^{-1} N) in the case of large doses. The soil nitrate N content was especially low (1–7 mg kg^{-1}) in the 0.4–1.2 m soil profile below the accumulation zone at all three measurement dates. As a result of the lack of precipitation and the moderate nutrient uptake of crops, the nitrate N content of the 0–1.2 m soil profile decreased only by 20–35% depending on the extent of fertilisation during the period between the 6/7-leaf-stage and silking. The nitrate N content did not change with soil depth (*Figure 3*).

In wet crop years, low nitrate N concentration (0.3–0.9 mg kg^{-1}) was observed also on non-fertilised plots. The fertiliser applied in the spring reached its accumulation peak in the 0.4–0.6 m layer at all three measurement date, which is the result of the early spring weather that was rainier than average. The mineral N concentration of the soil below the accumulation zone did not exceed 8 mg kg^{-1}. The nitrate N content reduction measured in the accumulation zone could be observed in all fertiliser treatments during the growing season. The nitrate N content stored in the 1.2 m soil profile was only 10–20 kg ha^{-1} in the case of 30–90 kg ha^{-1} N treatments. Results above 50 and 100 kg ha^{-1} were only obtained in the case of 120–150 kg ha^{-1} N doses (*Figure 4*).

The Chl content of the leaf did not change significantly as a result of various N doses at the V6 development stage either in dry or wet crop years. The N doses did not increase the Chl content even at the V12 phase in dry crop years, but there was a significant effect ($p<0.001$) in wet crop years and the highest Chl content was the result of the 120 kg ha^{-1} N dose. At the R1 phase, the N dose significantly increased the Chl content in both crop years $p<0.001$; $p<0.001$).

Yield was linearly increasing in dry (5.3 t ha^{-1}) and in wet crop years (11.2 t ha^{-1}) until the application of 60 kg ha^{-1} N. Beyond this point, the further increase of the fertiliser dose did not result in higher yield (*Figure 5*). The average yield increasing effect of fertilisation was 1.34 t ha^{-1} in dry crop years ($P<0.01$). This impact was more significant in wet crop years (3,27 t ha^{-1}, $P<0.001$).

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Pathological studies on maize (Zea mays L.) in Martonvásár

Csaba Szőke[1], Györgyi Micskei[2], Zoltán Nagy[1], Tamás Spitkó[1], L.Csaba Marton[1]
Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences [1]Maize Breeding Department,
[2]Crop Production Department
2462 Martonvásár, Brunszvik u. 2.
e-mail: szokecs@mail.mgki.hu

Abstract

Nowadays the greatest challenge for maize yield security is global warming and the resulting increase in the number of droughty years. Conditions that are unfavourable for the crop are in many cases ideal for various pathogens. The *Aspergillus* ssp. previously known in Hungary as warehouse diseases have now reared their heads in the field, and of course the various *Fusarium* species attacking maize must not be forgotten either. Both of these fungal species produce mycotoxins that are dangerous to both humans and animals. At present the best defence against these pathogens is maize resistance breeding, as no efficient, economical plant protection method has yet been elaborated. In the course of investigations on Fusarium stalk and ear rot and Aspergillus ear rot a stalk rot test based on cellulase enzyme activity has been elaborated, which could be of assistance in breeding for stalk rot resistance. Lines resistant to ear rot were also identified, the hybrids of which exhibited a high degree of resistance. The results achieved so far suggest that the genetic background of resistance to *F. verticillioides* and *A. flavus* is similar, while resistance to *F. graminearum* appears to have a different mechanism.

Introduction

Under Hungarian climatic conditions, rainfall and temperature are the two factors that play a fundamental role in determining the success of maize production. If these factors are unfavourable they not only lead to undesirable physiological effects, but also promote attack by numerous pathogens, the most important of which are toxin-producing fungal diseases: maize *Fusarium* and the *Aspergillus* ssp. attacking maize ears.

Maize Fusarium diseases

In a Continental climate maize may be attacked by several *Fusarium* species, the most frequent of which are *F. graminearum, F. culmorum, F. verticillioides* and *F. subglutinans* (*Békési and Hinfner*, 1970; *Mesterházy and Vojtovics*, 1977; *Szécsi*, 1994; *Dorn et al.*, 2009; *Scauflaire et al.*, 2011). Depending on the year, the pathogen may cause yield losses of 6–35% (*Bottalico*, 1998; *Logrieco et al.*, 2002), but the qualitative damage caused by the mycotoxins produced by these pathogens is a much bigger problem.

These secondary metabolic products not only result in a reduction in the nutritional value of the feed, but are also absorbed by the digestive system, accumulating in the tissues and causing severe abnormalities in the digestive and sexual organs (*Marasas*, 1995; *Raffai*, 1999; *Guerre et al.*, 2000). They also have an indirect effect, since the consumption of animal products contaminated with mycotoxins represents a potential risk for humans (*Kovács*, 2001). The most effective way of combating these pathogens is through maize resistance breeding. The mechanism of resistance to individual *Fusarium* species has not yet been completely clarified. *Presello et al.* (2006) identified resistance sources that provided very good resistance to the species *F. graminearum* (GER) and *F. verticillioides* (FER). *Löffler et al.* (2010) found a close correlation between the species *F. graminearum* and *F. verticillioides* for the phenotypic and genotypic resistance of flint and dent maize types, but the correlation between the two species was negative for silking date and ear rot severity. In the case of *F. graminearum* a very close correlation (r=0.94) was obtained between the mycotoxin content and the level of ear infection, suggesting that visual evaluation could be an efficient way of improving the level of resistance to GER *(Bolduan et al.*, 2009). Resistance to the *F. verticillioides* species and the accumulation of fumonisin exhibited greater variance in inbred lines, so the best way of increasing the FER resistance of hybrids is to test the initial lines for FER resistance (*Hung and Holland*, 2012).

Aspergillus ssp. ear rot (AER)

The incidence of these pathogens in the field was rare until a few years ago, and they were primarily known as warehouse diseases. At present they do not represent a serious danger, but as a consequence of global warming, they may well spread. AER also produce mycotoxins (various aflatoxins), which, like the *Fusarium* toxins, may cause severe physiological problems both in humans and animals. When examining several lots of maize kernel samples for *Aspergillus flavus* contamination, *Dobolyi et al.* (2011) found a level of infection of over 50% in lots from all the regions investigated. They also noted that the values in 42% of the samples exceeded the limit (5 μg kg^{-1}) laid down in EU fodder and food quality guidelines, and the pathogen was found to produce aflatoxin B_1 within 10 days under laboratory conditions at 26°C. *Henry et al.* (2009) identified maize genotypes with good resistance to *F. verticillioides* and *A. flavus*, and reported correlations of r=0.72 for ear infection and r=0.61 for mycotoxin concentration between the two pathogens, suggesting that the resistance of maize genotypes to both pathogens can be enhanced simultaneously.

Materials and methods

The experiments were set up over several years in the pathological nursery of the Maize Breeding Department, where the ears and stalks of inbred maize lines and hybrids were artificially inoculated according to the methods of *Young* (1943) and *Szőke et al.* (2009). Isolates of a number of *Fusarium* ssp. and of *Aspergillus flavus* were used for the inoculations. The percentage of the ear covered with mould (severity) and the proportion of infected ears (frequency) were scored in the nursery, while infected stalks were cut in half and evaluated in the laboratory after harvest. Natural ear and stalk infection were evaluated in performance trials at several locations in Hungary. The cellulase enzyme activity was determined with a modified cup-plate method (*Szőke*, 2011). The statistical evaluation was performed using the modules incorporated in the MS® Excel program and the Agronomix Inc. Agrobase program.

Results and discussion

Fusarium stalk rot of maize genotypes

Stalk samples from the artificially inoculated treatments, involving two *F. graminearum* isolates, and from the two control treatments (inoculated with *Fusarium*-free wheat grains, and without inoculation) were cut in half lengthwise to evaluate the health status of the pith. Significant differences were observed between the genotypes and, as expected, the lowest level of infection was found in the control treatments and the highest for the artificially inoculated stalks. Stalk tissue extracts were then prepared for each genotype and the cellulase enzyme activity was measured. No cellulase enzyme activity could be detected in tissue extracts prepared from healthy pith. The extract prepared from stalks inoculated with sterile grains exhibited detectable enzyme activity, depending on the degree of infection, but the greatest cellulase activity was recorded for the two artificially inoculated treatments. Correlations between the two factors were analysed, and a very close, positive correlation was found between the two traits for all the treatments (*Fig. 1*).

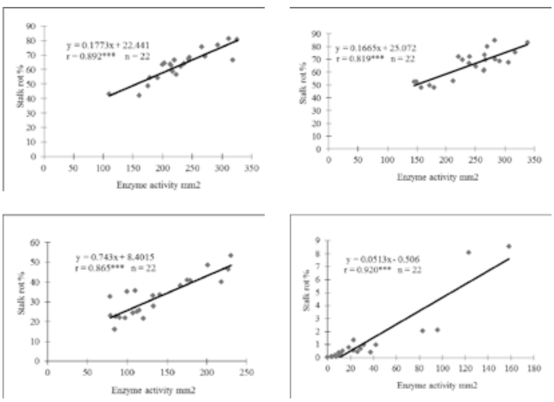

Fig. 1. Correlations between stalk rot and cellulase enzyme activity after inoculation with the isolates FG36 (upper left) and FGH4 (upper right), with sterile wheat grains (lower left) and without inoculation (control; lower right) (Martonvásár, 2006–2008)

The results showed lower fungal cellulase enzyme activity in more resistant maize genotypes and greater activity in more susceptible genotypes, while no enzyme activity could be detected in healthy tissues. As there was a very close correlation between the cellulase enzyme activity of the stalk tissue and the extent of stalk rot, the determination of cellulase enzyme activity could be an efficient method for use in breeding for resistance to stalk rot.

Fusarium and Aspergillus ear rot of maize genotypes

The correlation between the relative frequency of natural ear rot on 96 genotypes and the percentage of mould cover on the ear was determined over the average of five locations and three years (2004–2006). A close, positive correlation was obtained between the two parameters (*Fig. 2*), i.e. if the environmental factors are favourable for the development of *Fusarium* ear rot, the ears are likely to be severely affected. In the majority of cases this also means that there will be greater accumulation of mycotoxins, though for some species that produce mycotoxins the correlation between mould cover and toxin production is less close (*F. verticillioides, A. flavus*).

Artificial ear infection was the most serious in 2006 in terms of both severity (50.75%) and frequency (97.45%). When the *Fusarium* ear rot resistance of the 96 genotypes was determined at this high rate of infection, only four (4.16%) were found to be susceptible, while 51% were moderately resistant and seven (7.29%) exhibited very good resistance to ear *Fusarium* (*Fig. 3*).

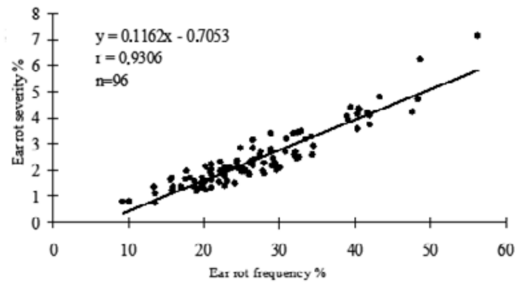

Fig. 2. Correlation between FER severity and frequency, averaged over the locations (2004–2006)

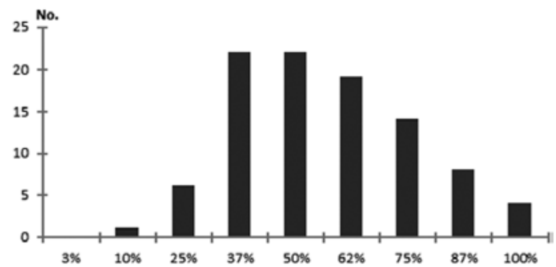

Fig. 3. Fusarium ear rot resistance of the genotypes tested (2006)

The correlation between natural infection and artificial inoculation was also analysed and was found to be medium (r=0.49) for frequency and very loose (r=0.14) for severity. This indicates that breeding for resistance to ear rot is impossible without artificial inoculation.

The parental components of the hybrids and new inbred lines were also examined in the pathological nursery. The data were then used for parent–progeny regression analysis to determine the heritability of *Fusarium* severity and frequency traits. The heritability of *Fusarium* severity (h^2=0.52) proved to be lower than that of the relative frequency of rotted ears (h^2=0.69). The h^2 values indicated, however, that additive gene effects played an important role in the heritability of both traits, providing a firm basis for selection in the course of inbreeding. Some of the hybrids created using new lines exhibiting resistance to ear rot were found to have a high level of resistance. These lines could thus be used as resistance sources. Some of the maize hybrids and lines exhibited satisfactory ear *Fusarium* resistance on the basis of field data.

The resistance of ten Martonvásár genotypes to ear rot caused by *F. graminearum, F. verticillioides* and *A. flavus* was studied in 2013, a year that was unfavourable for the development of ear rot. Even after artificial inoculation the highest mean value of severity (a symptom of susceptibility that can be visually evaluated) was only 16.5% for *F. graminearum*, the most infective of the species tested. Data in the literature suggest that the infectiveness of the other two species is much lower, and this was reflected in the present data. *F. verticillioides* caused the greater infection of the two (severity: 2.5%), while this figure was little more than 1% for the *A. flavus* isolate. Due to the low level of infection, significant differences between the genotypes were only found in the case of *F. graminearum* (*Fig. 4*).

The correlation between the resistance of the maize hybrids and the fungus species was also examined. Little similarity was observed between the level of resistance to *F. graminearum* and *F. verticillioides* (r=0.17). A similar correlation was detected be-

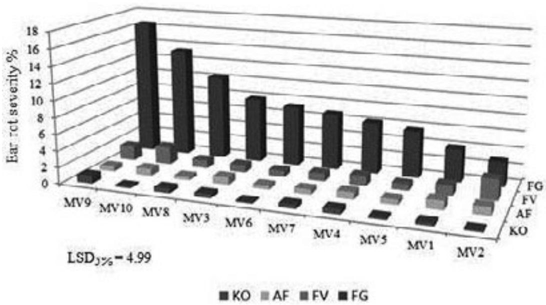

Fig. 4. Resistance of the hybrids to mould fungi (Martonvásár, 2013)

tween *F. graminearum* and *A. flavus*, but a medium correlation (r=0.50) was found between *F. verticillioides* and *A. flavus*. It can thus be concluded that although the genetic background of resistance to *F. graminearum* and *F. verticillioides* is different to that of *A. flavus*, plant resistance to *F. verticillioides* and *A. flavus* may be similar. In this year a medium correlation (r=0.59) was found between the level of natural infection and infection after inoculation with *F. graminearum*, while no correlation was found for the other two pathogens. This indicates that natural ear rot in Martonvásár in 2013 was caused chiefly by *F. graminearum*. To confirm this, infected ears have been collected, from which the pathogens responsible for infection will be determined.

The data achieved so far suggest that the genetic background may differ for resistance to individual pathogens causing ear rot. It is therefore advisable to use a number of pathogen species when breeding maize for resistance to ear rot.

Acknowledgement

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Section lectures
Section 2

Maize production under extreme conditions

Gábor Barla-Szabó
United Seeds CC
Executive Director
101 Argyle Str. Pretoria, 0081, South Africa
gabor@croppro.co.za

When I was finally admitted to study agriculture, my proud grandfather hugged me; I was his only grandchild who had chosen agriculture as a career. "I've farmed for more than sixty years", he said, "but remember my son, for a farmer there's no such a thing as a good year". Having farmed since then (the last 22 years in Africa), I can say he was right. There is no year which couldn't have been better for a farmer.

The "Rule of Thumb" predicts a Hectare of maize can produce approximately 15 Kg grain crop out of every millimetre of water. With 100mm that gives us 1.5 tons, with 500mm 7.5 tons, and so on. This "Thumb" however only works when it is connected to a strong hand, which is then connected to an arm over the shoulder, finally ending in a real Boer (farmer). Without that rain can fall, the sun can shine, but not much crop will grow. In addition, the variety and agronomy can significantly affect the applicability of the above rule.

White maize is the staple food in Africa. Over a billion African people eat maize porridge, or as they call it "pap" every day. Without white maize, the African people cannot be fed. It is therefore the most important food of the continent.

South Africa is 13 times larger than Hungary but the arable land of the country is only approx: 6 million Ha, just slightly more than in Hungary. This land should produce enough food for the 53 million populations.

The cultivated area of the country is divided into two major parts.

1. The summer rainfall zone. That is located in the middle and north section of the country, mostly high land areas. The so-called Eastern Highland falls east of the Pretoria- Johannesburg line. That region is 1400 – 2000 m above sea level. The average annual rainfall of 600-800 mm. The Western Highland 1300-1500 m above sea level and the average annual rainfall 400-600 mm.
2. The winter rainfall zone. Southern and south – western parts of the country where the rain comes mostly in winter. The summer is usually hot and dry.

30 % of the country is arid, semi-desert on the North-West side, while the Indian Ocean on moderately wet similar to Mediterranean climate.

The 3-3.5 million Hectares of the Corn Belt is located on the "summer rainfall" zone. In spring before the planting time in October- November, the soil is "stone" dry, as the result of the 6 or more Rainless months (Dry season). When the rain arrives the farmers will start immediately to prepare the soil and plant to save the time. Frequently they plant after the first 30-40 mm of precipitation, expecting more falls.

However, the extreme and unpredictable climate, in many cases the root of the 4-6 leaf stage juvenile plant growing against the "concrete" hard deeper ground layer and die off if rain stays away too long.

The clean and dry air of the high altitude results very strong Sun radiation, which accelerates the harmful processes as well. Sometimes the opposite happens, like two years ago in the North – Eastern regions of 620 mm of rain poured down in 36 hours.

Still, the food should be produced, the public must be provided with basic products. So what could be the basis for success in this country where the best corn fields are not even matches with the average European or U.S. production conditions.

Firstly: The most important law is that: we do not use American hybrids and do not follow the American farming methods!

A. We must use and produce hybrids that are extremely tolerant against wide range of environmental stresses. They can be planted in low populations, but in good conditions able to respond with larger and multiple cobs development. In other words, under good condition these hybrids are able to produce 2-3 large cobs and the size of the cobs are significantly bigger than under stress conditions (cob flex). *Figure 1.*

Figure 1. A Stand-alone plant with 8 cobs, 1.2 kg yield

A. They choose an appropriate, low-cost but effective agronomy. For example; broad row distance. Figure *2. Low- Density planting (7 feet row distance) is cheaper in every aspect.*

Secondly: While in the US, Europe and other higher potential countries like in Hungary 6-6.5 tonnes of crop yield offsets the input per hectare, whereas South Africa lower potential areas (about 2 million Ha), the input parity is only 3-3.5 tons/ ha. This makes it possible that under unfavourable weather conditions the 3-4 ton/Ha yield even can be economic and production remains sustainable. Then in a good years 6-9 ton/Ha yield resulting in a serious income.

Thirdly: In the U.S. and European Corn Belt the most cases have good soils supported by favourable climate and outstanding agronomy. During the last about 80 years the "American" hybrids have been selected to adapt for high potential environment, to bear dense population and make the yield on a single cob. Therefore, the farmers

Figure 2. 212 cm (7 feet) spacing

are always advised to plant higher population by the American Seed companies. In good year that strategy works well benefiting both farmer and the seed companies. Of course! More seed is used! More seed is sold! Better income for the seed company. The risk thus stays with the farmer. In a stressful season he takes the knock, his income will be hurt.

Fourthly: In Africa the safest maize production is based on the "African" hybrids. Some people are probably a little incredulous query, what could the African hybrid be? The correct answer is that in Southern Africa from the late 1940s farmers have been planting hybrid maize seeds already. The hybrids had been developed by the Government Research Institutes and the seed was distributed by private seed companies. As a fact the first commercial SC hybrid in the world was put on market in Southern Africa.

The hybrid registration eventually resolved in 1952, the best performer was registered under the name of SR52. The SR stands for Southern Rhodesia, the 52 year of registration. Interesting and strange SR52 was yield world record holder in the nineteen sixties, and 62 years after its registration this hybrid is still on the market.

Figure 3. United Seeds US9610 Hybrid and Gábor Barla-Szabó breeder

Today's "African" hybrids are a bit different, because the most important characteristic of these are the extreme stress tolerance, outstanding disease resistance, and capability for high yield with low population (18-40 thousand/ha). In good (rainy) years these hybrids are able to make high yield (even 9-13 t/Ha) although they were planted with low population.

The United Seeds Company (which I am also a founding member of) yearly brings new hybrids to the market. In the last production season our best hybrid "US9610" became the winner of the National Farmers trials. This is genetically half Hungarian half African hybrid. *Figure 3-4.*

Figure 4. Low Density good harvest

In South Africa annually approximately 3 million hectare commercial maize land is planted. In the early nineties, the U.S. multinationals have tried to adapt their, hybrids but due to single cob nature, the lower disease resistance, the frequent sun burn and poor drought tolerance, they were changed to locally developed hybrids.

Today American hybrids are planted on the irrigated areas where the earliness and fast dry down is more important than top yield. After maize immediately wheat comes, so time is very crucial. These are FAO 600 hybrids here they are called “Super Early” group).

The African hybrids, in lower plant density tend to develop 2-3 cobs/plant and they are also flexible with cob size. Good, rainy weather is not only developing and fully nursing multi cobs but their sizes are also significantly increased.

In the 2012-13 maize production season the US9610 became the overall best performer of the National Farmer Trials. This hybrid has one of the best drought tolerance but very highly capable to produce multi-cob plants under favourable conditions.

Photo 4 is a picture about US9610 in the Eastern Highland Davel area. Plant population was 35000/Ha but due to good rain 60 000 cobs developed and resulted 9.3 t/Ha average yield on more than 120Ha.

	Davel Trial	Yellow Maize Hybird Trial 2013			
	Cultivar	**TON/HA**	**Moist%**	**Stand**	**% v Trial**
1	Phb 32W72W	7.53	11.00	40000	121.80
2	**US 9610**	**7.38**	**11.60**	**40000**	**119.20**
3	LS 8526	7.27	11.50	40000	118.63
4	PAN 6Q-408CB	7.22	12.50	40000	118.42
6	DKC80-12B	7.17	11.70	40000	115.88
7	LS8536B	7.04	11.20	40000	113.84
8	PAN 6Q-308B	6.79	12.20	40000	109.90
9	PAN 4P-228	6.76	11.90	48000	109.21
10	PAN 6P-110	6.74	12.20	40000	108.98
11	US 9620	6.63	11.5	40000	107.17
12	KKS 8408R	6.54	11.6	48000	105.68
13	DKC73-72	6.48	11.8	48000	104.8
14	DKC 73-76R	6.26	11.9	48000	101.18
15	Phb 33H56	6.13	10.9	48000	99.01
16	KK 4410	6.06	11.7	48000	97.96
17	DKC 73-70B	6.05	11.7	48000	97.71
18	LS 8524R	6.03	11.6	40000	97.48
19	Phb 1615BR	5.93	11.2	48000	95.9

Table 1, National Farmer Trail at Davel area, South Africa

Several trials data support the outstanding performance of the US9610. I can show here one which does make sense to study in more detail. Table 1.

The trial management allowed for the contributor companies to choose between 2 plant populations. The result speaks for itself, the lower plant density plots namely 40000/ha became significantly better than higher 48000 thousands/ha.

Itamba Highland		White Maize Hybrid Trial 2013		
No.	**Hybrid Name**	**Grain Yield 15% mc. (ton/ha.)**	**Relative Phb3253 %**	**Relative mean %**
1	**US9747**	**8.11**	**150**	**135**
2	**US9755**	**7.59**	**140**	**126**
3	HCSY1201	6.87	127	115
4	SC 627	6.58	122	110
5	EXP3	6.20	115	103
6	DK8073	5.89	109	98
7	PAN67	5.87	108	98
8	UH6303	5.84	108	97
9	Phb 3253	5.41	100	90
10	DK8053	5.12	95	85
11	H614D	5.05	93	84
12	HCSY1201	4.97	92	83
13	PAN691	4.54	84	76
	Mean	6.00	-	-

Table 2. Itumba, Tanzania 2013th

The “American” corn cultivation method deeply embedded in the Hungarian maze production and plant breeding. If you would ask 100 Hungarian agro-professionals to explain in detail of the maize planting and production, we would receive 100 same answers without any consideration or doubt “, despite of the last 2 years severe conditions and yield losses.

They would say: the maize must be planted to 65-70 thousand plant/Ha with a 30 inch (76cm) planter machine. The Hungarian breeders in this area continue to follow and try to compete with the breeders of multinational companies. They try to do the same development with a very limited budget hoping to beat the dollar billions spending oppositions.

In few isolated cases thus the maize breeders from Central Europe could make some remarkable trend braking developments based on their outstanding knowledge. These hybrids thus could never been utilised on the market level what they deserve, due to the less developed seed production, QA, processing-warehousing, marketing and sales systems.

The Hungarian variety registration is equally inflexible. All types of experiments to be carried out using the method, what the last 40 years recommended by the multi-nationals and would be very difficult to change that.

I am sure it would be struggle to make accepted a new “low population multi cob” type, drought -tolerant hybrid.

In Hungary there is about 400 hectares of land, which is not utilised for maize production due to the high input but moderate yield expectations. With a lower density, early "African" type hybrid the maize production could be made profitable in these areas as well.

The corn seed production in Hungary in the last 60 years

Zoltán Benke
National Food Chain Safety Office
Directorate of Plant Production and Horticulture
Department for Seed Inspection and Certification
1024 Budapest, Keleti K. u.24.
benkez@nebih.gov.hu

Hybrid maize seed production has traditionally dual purpose as to fulfill domestic seed demand, and to provide a powerful export market presence.
Domestic ecological conditions are suitable for the hybrid maize production, and the technical and professional background provides basis for successful seed production

The 50-60% of the hybrid maize seeds produced traditionally has been sold on export markets for long.

The hybrid maize seed production was economically always competitive in relation to the other crops.

This profitability was in line wit the work and effort invested. Fertile and homogeneous field, the irrigation, the professional expertise, good labour and management a high level of machinery are the main parameters are of high quality seed production.

Seed production area is strongly dependent on the otput of preceding year as well as marketing plan prospects. The hybrid corn seed production has 50 year history. The annual publications of the inspection authorities (OVEF, MMI, OMMI, MgSzH NÉBIH) provide information on how seed production area and the amount of sealed lots were evolved.

The begining
The spread of hybrid corn varieties was stimulated greatly by the hybrid program approved in 1954 in Hungary. In the summer of 1956 the Martonvásári Experimental Institute received the first diesel-fired, automatic Campell type grain dryer with thermostat. In the same year they built the first corn grain dryer with six chambers in the country. In1957 the Research Institute of Martonvasar established a special hybrid corn working group hence the production of hybrid maize seed production was put into their own hands.

The Headquater of State Farms (ÁGK) worked together with the Research Institute to solve the hybrid corn seed production in Hungary. In 1958 they built the first six hybrid seed corn plants owned by state farms in Baja, Bóly, Mezohegyes, Mezőnagymihály, Debrecen and Murony. Subsequently, some more seed processing plants were also built in Mezőfalva Dalmand, Mosonmagyarovar, Szenttamás, Hódmezõvásárhely and in Cegléd between 1959-1964. Thus was created the hybrid maize seed industry in Hungary with capacity of 36 thousand tons per year production taking into account the seasonal period.

Meanwhile, the professional and scientific background of seed production had been built as well. The staff of Hungarian Academy of Sciences (HAS) developed and introduced to the practice of modern types of maintenance of varieties. The experts of the research institutes had developed an overall production contracts including quality of conditions of hybrid corn seed production.

Until 1957 the National Seed Inspectorate (OVEF) carried out only field control and certification of improved varieties of maize in accordance with the system of seed renewal. From the mid-year of 1957 the staff of the National Seed Inspectorate jointly with the staff of Martonásárhely started to carry out joints assessments experimentally, that started the state control of hybrid maize seed propagation in Hubgary.

Hungary achieved close to 100% the use of hybrid maize seed production between 1957 and 1964. In 1958 the total seed production area was 18 088 ha in Hungary and on more than half (53.1%) of the total field of seed production the inbred lines, base materials, single, double or multiple-cross hybrid maize varieties (eg. Mv 1, Mv 5, Mv 26, Mv 39, Mv 40) were propagated. On the 8.4 percent of the total growth area the cross polinated varieties (Golden Flood, Fleischmann early, Szeged yellow, yellow Mindszentpusztai etc.) were produced and on the 38.5% of the field hybrids (eg, Óvári 5) seeds were produced.

45.6% of 37 710 t of sealed maize was hybrid production and large amounts of commercial maize were still at sealing (42.1%), most for export purpose.

In 1964, the base materials seeds were produced on 1 percent of total propagation area (25 717 ha), 90.3 % of the total area was HF1 and 8.7 percent was cross polinated varieties produced by. At that time, the amount of sealed maize was close to 50.000 tonnes. Year of 1968 was also a turning point in hybrid maize seed production.

Many of the farms also reached that level of agronomy where the highly demanding, sensitive, but intensive single cross hybrids gave better production and became more efficient compare to the multiple hybrids.

In 1968, following the positive experience at large-scale experiments the volume of a single cross hybrid maize producing areas has almost tripled. This year the single cross (SC) hybrid seeds was propagated on 4912 ha and had only been used a minor part for basic material propagation. Unlike the previous period, the majority of the crop had become commodity. In SC hybrid maize seed varieties was propagated on 58% of the total planting area (20.992 ha)

The future

The hybrid maize seed production area is expected to grow due to in growth is expected to be, however, the successful implementation of seed production is not easy task. The conditions for further expansion are in the first place the irrigable and nutritious lands with crop rotation and ensured processing capacities in the background.

Further seed production can be achieved by increasing sales in Central and Eastern Europe and in the EU due to constant level of domestic production.

Production safety gains greater emphasis by production increase year by year.

Varieties with stable production and season tolerance, the crop rotation preventing pest and disease problems and balanced soil nutrition input are the main parameters that should be taken into account primarily in seed production.

Year	Area in-spected (ha)	Number of varieties (pieces)	Number of fields (pieces)	Average yield (kgs/ha)	Yield (mto))	Quantity of certified seed (metric tons)			
						Total	Inland	Export	Export (%)*
1985.	48.984	Na*	na	na	na	na	na	na	na
1986.	56.373	315	na	na	na	125.467	41.840	83.627	67
1987.	61.453	295	na	na	na	148.418	56.126	92.292	62
1988.	65.488	313	na	na	na	135.162	42.097	93.065	69
1989.	58.072	351	na	1.715	99.626	135.459	42.472	92.987	69
1990.	46.986	342	1.034	1.113	52.306	82.813	32.100	50.710	61
1991.	47.388	363	1.065	2.386	113.100	79.300	43.215	36.092	45
1992.	41.104	304	898	1.691	69.534	89.571	40.472	49.099	55
1993.	39.153	327	929	1.980	77.515	95.463	30.189	65.274	68
1994.	42.030	324	1.002	2.160	90.735	81.085	24.800	56.284	69
1995.	32.699	289	849	2.516	82.283	88.553	29.525	59.028	67
1996.	16.568	245	514	2.713	44.954	74.518	29.958	44.560	60
1997.	19.262	238	511	2.903	54.392	84.379	32.804	51.545	61
1998.	23.904	288	626	3.480	81.929	68.075	27.485	40.590	59
1999.	25.912	309	731	3.952	96.007	79.262	29.171	50.090	63
2000.	24.836	306	718	2.223	54.394	79.503	35.118	44.384	55
2001.	29.017	369	842	3.793	108.741	54.820	22.348	32.471	59
2002.	30.420	418	947	2.826	83.645	66.547	36.362	30.184	45
2003.	27.126	384	915	2.613	70.296	74.822	31.353	43.469	58
2004.	28.287	373	1.021	3.607	101.284	69.872	24.212	45.660	65
2005.	25.597	390	1.023	3.907	99.257	78.434	37.554	40.880	52
2006.	19.491	277	837	2.857	55.311	84.847	38.065	46.781	55
2007.	22.851	330	786	2.142	47.075	77.996	37.224	40.613	52
2008.	26.137	364	860	3.525	91.819	78.049	36.187	41.749	53
2009.	33.807	430	1.022	3.274	109.848	85.323	31.761	53.450	63
2010.	16.227	308	663	2.848	45.315	94.265	30.089	64.176	68
2011.	25.377	340	891	3.120	78.928	84.162	43.853	39.470	47
2012.	34.203	420	1089	1.952	63.036	76.160	32.722	43.438	57
2013**	39.597								

**Not dates*
*** Date of 15.10.2013*

Table 1. Maize seed multiplication 1985-2013

Correlation between phenotype and quality in leafy (Lfy) silage maize hybrids

János Pintér[1], Francis Glenn[2], István Pók[1], Géza Hadi[1], Zsuzsanna Tóth Zsubori[1], Zoltán Nagy[1], Csaba Szőke[1], Tamás Spitkó[1], Tamás Berzy[1], L. Csaba Marton[1]
[1]*Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences*
2462 Martonvásár, Brunszvik u. 2.
[2]*Glenn Maize France BV, 81 Rte de la chapelle de Rousse, 64290 Gan, France*
e-mail: pinter.janos@agrar.mta.hu

Abstract

Maize is currently grown on around 170 million hectares on a world scale. The USA and China make up 60% of the total production, each with a growing area of 35 million ha. The third largest growing region consists of the 28 member states of the EU, with 13 million ha. Data on the size of the growing area (which mainly refer to grain maize) provide little information on the size of the silage maize area either within or over and above this. In the USA, China and Canada the ratio of silage maize is 10–15%, while in European countries with large stocks of cattle (e.g. France, Germany, the Netherlands, Denmark) this ratio is closer to 50% or more (*FAO,* 2013*; Eurostat,* 2013*;USDA,* 2013*; IndexMundi,* 2013). In Hungary, in addition to an average annual growing area of 1.1 million ha grain maize, the area sown to silage maize is increasing in order to satisfy the growing feed requirements of the cattle stocks, which are on the rise particularly on intensive farms. It has thus become essential for silage maize hybrids with excellent quality and capable of producing larger fresh and dry matter mass to be introduced into commercial production in order to achieve higher milk and beef yields per hectare. Changes in livestock keeping technologies require a new, up-to-date generation of silage maize hybrids. Under Hungarian conditions the silage and dual-purpose hybrids previously grown belonged to the FAO 300–500 groups, but farmers are increasingly demanding hybrids with an FAO number of over 600.

Martonvásár not only developed the first European maize hybrid (Mv 5, 1953), but has always led the way in silage maize breeding. In earlier years large volumes of silage hybrid seed from early maturity groups were exported to Germany, Russia and Poland.

As the result of the use of new genetic material, the first leafy (*Lfy*) hybrid registered in Europe was granted state registration in Hungary in 2002 under the name Kámasil.

The present work sought an answer to the question of what quantitative and qualitative changes were caused by the presence of the *Lfy-1* gene and the resulting increase in the leaf number per plant in the seven leafy hybrids registered to date in Martonvásár. The results confirmed that, thanks to the greater photosynthesising leaf area, these *Lfy* hybrids produce greater fresh and dry matter yield per unit area compared with conventional silage hybrids in the same maturity group. A further advantage, also induced by the *Lfy-1* gene, is the quality gain, manifested as an improvement in the digestibility of the silage.

Introduction

Although the leaves do not have the highest value in terms of starch index, they are one of the most important components of silage maize, as they provide the roughage essential for digestion in ruminants. In addition to the choice of variety and the production technology, the state of the whole plant at silage maturity, including its freshness at harvesting, the digestible dry matter content, the fibre content (ADF, NDF) and the lignin content, is also of fundamental importance, as together these determine the value, quality and level of utilisation of the silage after fermentation.

The main aim in the experiments was to discover what changes the *Lfy-1* gene causes in the canopy structure of the hybrids and how this change influences the quantity, quality and, most importantly, the digestibility of the yield.

The majority of the "quality enhancing" genes used up to now in maize breeding (e.g. opaque, waxy, floury, bmr), while improving one of the quality parameters, have also caused undesirable changes (lower yield, slower drying down, greater disease susceptibility, stalk lodging, etc.) from the agronomic point of view, thus explaining why, after a short period of popularity, they all fell into disuse. In earlier years, while conventional silage maize breeding was successful in improving yield levels, very little progress was made in terms of quality, especially the digestibility of the silage. A breakthrough in this field was achieved when *Lfy* hybrids were introduced into commercial production (*Lauer et al.*, 2001; *Sanavy et al.*, 2009; *Hegyi*, 2011).

The experience gained so far indicates that the use of the *Lfy-1* gene in breeding programmes is likely to lead to improvements in both quantity and quality.

The *Lfy-1* mutant gene was discovered more than 30 years ago by Robert C. Muirhead and Donald L. Shaver. *Shaver* (1983) was the first to report the increase in the leaf number and leaf area, the reduction in the ear attachment height and the greater yield recorded for hybrids carrying the *Lfy-1* gene. In most of the experiments carried out by Begna et al. (1997, 2001), Modarres et al. (1997a), Dwyer et al. (1998), Andrews et al. (2000) and Sanavy et al. (2009) it was found that the increase in the plant leaf area and the whole biomass could be attributed chiefly to an increase in the leaf number above the ear. However, in most *Lfy* hybrids the leaf number below the ear was also greater than in non-*Lfy* hybrids. These authors observed a shortening of the vegetative period and a lengthening of the grain-filling period compared with normal analogues. Other authors (*Tollenar and Dwyer*, 1990; *Dwyer*, 1995; *Modarres et al.*, 1997b; *Dijak et al.*, 1999; *Subedi and Ma*, 2005; *Hammer et al.*, 2009) also reported on the positive consequences and benefits of the increased photosynthesising area induced by the *Lfy-1* gene. These authors made special mention of the fact that the greater photosynthesising area played a very important role in biomass accumulation during the flowering and grain-filling stages. *Dwyer et al.* (1995) and *Stewart et al.* (1997) compared the canopy structure of non-*Lfy* and *Lfy* hybrids above the ear and analysed the soluble and insoluble carbohydrate contents in each leaf zone. The quantity of soluble carbohydrates in the leaf canopy above the ear was found to be twice as high in *Lfy* hybrids as in that of conventional hybrids. In an earlier paper *Perry and Caldwell* (1969) reported that the portion of the nutrients present in the leaves (soluble carbohydrates, proteins) could be utilised just as well as those already accumulated in the kernels (mainly as starch). The kernel quality parameters of *Lfy* hybrids have been analysed by many authors, who in most cases found a positive change in the *Lfy* hybrids, especially for parameters promoting digestibility. However, few papers have dealt with how these high-yielding, high-quality *Lfy* hybrids should be grown if their yield and quality potential is to be exploited to the full. Dr Francis Glenn, the breeder of the *Lfy* inbred lines used in Martonvásár, reported (oral communication) that the special, large canopy structure of these hybrids has an important role in the more efficient fixation of light energy (due to the high LAI value). For this reason, caution is required in adjusting the plant density, in order to minimise shading. As these plants are extremely tall, due to their late maturing date, they have considerably more leaves and greater leaf area than conventional hybrids in the same maturity group, so the recommended plant density is 62–65 thousand plants/ha.

Materials and methods

The experiments were set up in 2012 and 2013 in the breeding nursery in Martonvásár (47°18' N, 18°46' E) in 3-row plots in a random block design with 4 replications and a plant density of 62,000/ha. The soil was pseudomyceliar chernozom on a mother rock of loess-containing sand. During the vegetation period of silage maize (Apr.–Aug.) the mean temperature was 18.83°C in 2012 and 18.33°C in 2013, compared with the 30-year mean of 17.93°C, while the rainfall sum was 34.2 mm in 2012 and 34.4 mm in 2013, compared with the 30-year mean of 54.2 mm. Irrigation was carried out on 4 occasions in both years, with a quantity of 20 mm on each occasion. There were 43 extremely hot days in 2012 and 38 in 2013. The heat sum was 1269°C in 2012 and 1192°C in 2013.

Four plants were taken randomly from the middle row of each plot for the measurement of leaf area and for the qualitative analysis of silage samples. In 2012 two non-*Lfy* standards were investigated together with three Martonvásár *Lfy* hybrids, while in 2013 three non-*Lfy* standards and six Martonvásár *Lfy* hybrids were tested, together with their parental components.

Hybrids		Leaf No.		Total leaf area (cm^2)	Whole fresh mass (t/ha)	Whole dry matter yield (t/ha)	Ear ratio (%)	Whole digestible dry matter yield (t/ha)
		Above the ear	Total					
non-*Lfy*	St. FAO 480	6.90	14.00	6206.75	61.85	21.48	**58.42**	14.49
	St. Maros	6.80	13.40	5978.00	64.54	20.90	48.28	14.33
	St. Maxima	6.60	14.50	6876.16	77.84	25.83	48.44	17.92
Lfy	**Kámasil**	10.80	17.10	7608.61	68.60	22.56	49.46	15.01
	Limasil	10.30	16.60	7032.45	63.28	21.99	49.84	14.62
	Massil	10.30	**18.70**	**8329.78**	79.27	25.23	43.47	16.73
	Megasil	**11.10**	17.90	7823.47	75.44	26.48	49.16	18.01
	Nutrisil	8.10	15.10	7908.13	78.53	24.83	52.44	16.73
	Siloking	**11.10**	18.30	8250.59	**84.62**	**28.67**	49.52	**19.26**
	Lfy mean	10.28	17.28	7825.51	74.96	24.96	48.98	16.73
	Check mean	6.77	13.97	6353.63	68.08	22.74	51.71	15.58
	CV	5.17	3.39	5.67	16.90	14.82	11.25	14.55
	$SD_{5\%}$	0.57	0.66	503.03	14.86	4.34	6.79	2.88
St.=Standard								

Table 1. Trends in leaf number, leaf area and various quality traits. Martonvásár, 2013

	Hybrids	Plant height (cm)	Ear attachment height (cm)	Leaf No. above the ear	Leaf area (cm^2)	
					above the ear	Total
Non-*Lfy*	St. Maxima	**311.22**	**159.03**	6.75	2977.38	7067.25
	St. FAO 480	284.28	129.41	6.88	2796.75	6234.25
Lfy	**Massil**	307.22	141.72	9.84	4256.75	8017.00
	Nutrisil	293.38	124.78	8.22	4087.13	7879.25
	Siloking	303.91	127.25	**10.72**	**4459.75**	**8259.75**
	Grand mean	300.00	136.44	8.48	3715.55	7491.50
	Check mean	297.78	144.22	6.81	2887.06	6650.75
	$SD_{5\%}$	5.70	7.44	0.37	332.31	460.91

Table 2. Trends in the phenotype of Lfy and non-Lfy hybrids. Martonvásár, 2013

A LI-COR LI-3100C instrument was used to measure the leaf area. In both years silaging was performed mechanically and the size was adjusted to 1 cm. The samples were harvested at silage maturity, i.e. at a dry matter content of approx. 35%. The quality parameters were determined using a BRUKNER MPA NIR spectrometer with the help of INGOT calibration software. The samples were dried in a MEMMERT/800 drying cabinet at 105°C for 72 hours. The data were statistically evaluated with analysis of variance using AGROBASE software.

Results and discussion

Phenotypic traits of Lfy and non-Lfy silage hybrids and changes in major quality traits

Three *Lfy* hybrids were compared in the first year and six in the second year. The results of phenological measurements are summarised in *Tables 1 and 2*.

There was little difference between the two types in terms of plant height, but the ears of the *Lfy* hybrids were attached significantly lower than those of the non-*Lfy* hybrids It was clear from the data that the vast majority of the leafy hybrids had significantly more leaves than the non-leafy hybrids, with a surplus leaf number of +3.5 above the ear (*Fig. 1*). In 2013 Silóking had the greatest leaf number (11), and the average leaf number over the two years was also close to this value (10.72).

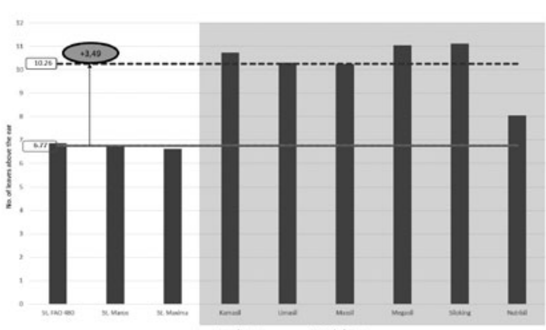

Fig. 1. Number of leaves above the ear for non-leafy and leafy hybrids. Martonvásár, 2013

The most pronounced difference was found in the canopy structure, i.e. in the leaf area of the canopy. In both years Massil and Silóking had outstanding values of 8200–8300 cm², compared with that of the hybrid used as standard in official variety trials (6200 cm^2). The main morphological parameters of the *Lfy* hybrid Kámasil, the non-*Lfy* hybrid Maxima and their parental components are illustrated in *Figure 2*. It is clear that the leaf area above the ear was considerably greater (+56%) for Kámasil, although the plants were shorter than those of Maxima.

Analysis of the major quality parameters of Lfy hybrids

Figure 3 illustrates the leaf area per hectare at silage maturity for a conventional and a leafy (*Lfy*) hybrid sown at the same plant density. It is clear that it was the size of the canopy, not the plant height that was chiefly responsible for the advantage of *Lfy* hybrids over non-*Lfy* hybrids in terms of total fresh and dry mass and total digestible dry matter yield. The leafy hybrid Silóking had better values of all three yield components influencing the yield and quality of the silage (total fresh mass, dry matter yield and digestible dry matter yield) in 2012, compared with the mean for the standards, and significantly better values when averaged over the two years (*Tables 1 and 3, Fig. 4*).

The values of the most important quality traits in terms of digestibility, based on NIR measurements, are presented in *Table 4*. The starch, water-soluble carbohydrate, protein and crude fat contents (in terms of yield/ha) were higher than those of the standards for the Martonvásár *Lfy* hybrids registered in recent years (Massil, Megasil, Nutrisil, Silóking). For two of these (Megasil, Silóking) the differences were significant.

It should be noted that the Martonvásár silage hybrid Maxima, which also performed very well in the present experiments, was included among the standards in both years. As a member of the standard group it was a worthy rival to the new leafy hybrids registered in Martonvásár in recent years.

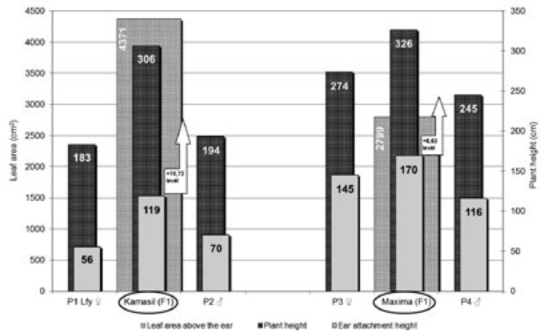

Fig. 2. Plant height, ear attachment height and number of leaves above the ear for Lfy and non-Lfy hybrids. Martonvásár, 2013

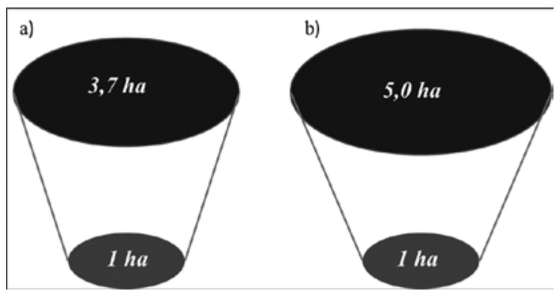

Fig. 3. Total leaf area per hectare for a conventional standard (FAO 480) (a) and for a Lfy silage hybrid (Mv Silóking) (b). Martonvásár, 2013

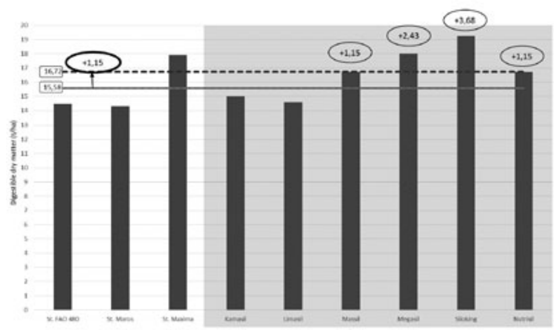

Fig. 4. Digestible dry matter yield of non-Lfy and Lfy hybrids (t/ha). Martonvásár, 2013

	Hybrids	Total fresh matter yield (t/ha)	Total dry matter yield (t/ha)	Starch yield (t/ha)	Total digestible dry matter yield (t/ha)
non-Lfy	St. Maxima	74.73	26.43	**12.59**	18.08
	St. FAO 480	60.74	21.51	9.31	14.67
Lfy	**Massil**	75.80	25.06	10.56	16.83
	Nutrisil	71.11	23.99	10.85	16.28
	Siloking	**80.41**	**28.00**	11.55	**18.38**
	Grand mean	72.56	25.00	10.97	16.85
	Check mean	67.73	23.97	10.95	16.37
	CV	14.90	18.37	31.98	19.83
	$SD_{5\%}$	9.21	3.91	2.99	2.84

Table 3. Trends in major parameters determining silage value. Martonvásár, 2013

	Hybrids	Starch		Water-soluble carbohydrates		Protein		Fat	
		%	t/ha	%	t/ha	%	t/ha	%	t/ha
Non-*Lfy*	St. FAO 480	44.96	9.67	7.11	1.54	8.48	1.81	3.85	0.83
	St. Maros	45.41	9.47	7.26	1.50	8.18	1.71	3.88	0.81
	St. Maxima	48.51	12.50	8.54	1.88	8.54	2.20	3.90	1.01
Lfy	**Kámasil**	45.6	10.31	7.11	1.61	8.23	1.86	3.85	0.87
	Limasil	42.76	9.46	6.96	1.54	8.30	1.83	3.82	0.84
	Massil	44.47	11.19	7.16	1.80	8.17	**2.06**	3.69	0.93
	Megasil	51.44	**13.57**	8.93	**2.36**	7.92	**2.10**	3.73	**0.99**
	Nutrisil	46.33	11.54	8.23	1.90	8.23	**2.04**	3.90	0.97
	Siloking	46.29	**13.15**	8.28	**2.02**	8.28	**2.38**	3.80	**1.09**
	Lfy mean	46.15	11.54	7.78	1.87	8.19	2.05	3.80	0.95
	Check mean	46.29	10.56	7.22	1.64	8.40	1.91	3.88	0.88
	CV	9.46	16.08	9.81	16.53	4.12	14.94	4.06	14.94
	$SD_{5\%}$	5.28	2.18	0.88	0.36	0.41	0.36	0.19	0.17

Table 4. Quality traits of importance for digestibility. Martonvásár, 2013

Heterosis analysis

In the 2013 experiments the six Martonvásár *Lfy* hybrids were sown together with their parental components.

As an above-average increase in the leaf number and leaf area can be expected when developing leafy-type hybrids, due to the use of a parental partner bearing the dominant *Lfy-1* gene, this provided an excellent opportunity to examine the heterosis effects between the hybrids and their parental components. The results are summarised in *Table 5*.

The greatest heterosis was detected for the plant height, ear attachment height, leaf number above the ear and total leaf area for the Limasil hybrid, with a value of around 180% averaged over these parameters. This high value of heterosis can be attributed to the fact that there were very substantial differences between the two parental components of the hybrids with respect to size, morphology and vegetation period. Among the morphological traits, the greatest heterosis and heterobeltiosis, averaged over the *Lfy* hybrids, were observed for the total leaf area (170 and 150%, respectively).

	Plant height (cm)	Ear attachment height (cm)	Leaf number and leaf area					
			Total	Below the ear		Above the ear		Total
				db	(m^2)	db	(m^2)	(m^2)
Nutrisil	305	130	15.1	7.00	0.36	**8.10**	0.43	**0.79**
H	149	158		120	155	101	170	163
HB	140	142		107_{Lfy}	153_{Lfy}	85_{Lfy}	146_{Lfy}	149_{Lfy}
Kámasil	306	119	17	6.30	0.32	**10.70**	0.44	**0.76**
H	163	189		130	215	130	173	189
HB	158	169		115_{Lfy}	223	115	135_{Lfy}	162_{Lfy}
Limasil	308	117	16.6	6.30	0.27	**10.30**	0.43	**0.70**
H	174	201		141	253	136	175	199
HB	168_{Lfy}	193		126_{Lfy}	189	111_{Lfy}	132_{Lfy}	150_{Lfy}
Siloking	322	135	18.2	7.10	0.37	**11.10**	0.45	**0.82**
H	140	139		120	171	135	155	162
HB	116	97		104	129	119_{Lfy}	141_{Lfy}	150
Massil	323	160	18.7	8.40	0.39	**10.30**	0.45	**0.83**
H	141	159		124	138	127	167	152
HB	118	110		98	94	110_{Lfy}	138_{Lfy}	133
Megasil	327	130	18	6.90	0.33	**11.10**	0.45	**0.78**
H	155	158		121	177	141	160	167
HB	138	119		108	144	119_{Lfy}	138_{Lfy}	166
H=heterosis (%), HB=heterobeltiosis (%)								

Table 5. Values of heterosis and heterobeltiosis for the morphological traits of six Martonvásár Lfy hybrids. Martonvásár, 2013.

When calculating heterobeltiosis for the six hybrids, the *Lfy* parental component was found to have the greatest influence on the leaf number above the ear for 5 hybrids and on the leaf area above the ear for all 6 hybrids. The dominance of the *Lfy* parent was significant for the total leaf area in the case of SC hybrids, but not for the three TC hybrids, since the SC components of the non-*Lfy* female parent themselves had large leaf area, which obviously made a considerable contribution to the total leaf area of the F_1 progeny plants.

The development of leafy (*Lfy*) hybrids has been part of the Martonvásár silage maize breeding programme for more than 10 years. Based on the one- and two-year data presented here it can be stated that the use of the *Lfy-1* gene has numerous benefits, thanks to which a total of 11 leafy hybrids have been registered so far.

As a consequence of the greater number of leaves above the ear and for the whole plant these high-quality silage hybrids have a substantially greater foliage area than conventional hybrids, especially above the ear, representing an increase in the assimilating surface.

As a result, the fresh and dry matter weight per hectare, and numerous other digestibility and quality parameters are considerably greater for these leafy hybrids than for the standards. Their greatest advantage, however, is that they also have an excellent yield of total digestible dry matter. Thanks to their slow drying down and to their phenology at silage maturity (stay-green, larger number of fresh, nutrient-rich leaves above the ear, smaller fraction of indigestible fibre) the length of the harvest period can be extended with practically no deterioration in quality. Especially good results were achieved with the hybrids Massil and Silóking.

Acknowledgement

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Posters

In vitro selection of microspores to enhance the oxidative stress tolerance of single cross maize hybrids with breeding value

Helga Ambrus [1*], Éva Darkó [2], Tamás Spitkó [3], János Pintér [3], Beáta Barnabás [1]
Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
[1] Plant Cell Biology Department,
[2] Department of Plant Physiology,
[3] Maize Breeding Department
H-2462, Martonvásár, Brunszvik Street 2
e-mail:[*] ambrus.helga@agrar.mta.hu

Abstract

In order to improve the abiotic and biotic stress tolerance of maize, 29 doubled-haploid (DH) maize (*Zea mays* L.) plants were selected and regenerated from microspores exposed to oxidative stress stimulators. A single cross maize hybrid A-18 (originating from a paraquat resistant DH line as a female parent and from another DH line with high androgenic capacity as the male parent) were used for *in vitro* selection. After the model experiment, a couple of single cross hybrids of breeding value were also tested in selection experiments. Physiological investigations were performed on the first generation fertile microspore-selected lines (DH_1) (originating from H1, H2, H3 hybrids) in order to verify that they have higher oxidative stress tolerance (caused by paraquat and cold) than the original hybrid. From 29 paraquat selected lines, eleven of them presented higher oxidative stress tolerance against Pq, 23 of them had a higher cold tolerance during the germination. A total of ten selected genotypes, six from hybrid H1, two from H2 and two from H3, proved to have better tolerance index in terms of both Pq-induced oxidative stress and chilling stress at germination than the non-selected DH lines or the initial hybrids.

Introduction

In addition to the challenge represented by the extreme weather conditions experienced nowadays (sudden warming up early in the year, heat waves in May, dry or excessively wet summers, the late arrival of spring, long periods of hot weather) and by the increasing cost of raw materials, plant breeders must also be prepared to develop new breeding material or hybrids with great ecological plasticity and resistance to abiotic (e.g. cold, drought, etc.) and biotic (pathogens) stress factors within a short space of time.

Numerous methods are now available to plant breeders to optimise the gene composition and gene functioning of the breeding materials, including both conventional methods (selection based on phenotype or molecular markers) and up-to-date gene technological and biotechnological breeding methods. It is known from the scientific literature that most of the abiotic and biotic stresses damage the plants by the production of toxic reactive oxygen species (ROS) (*Apel and Hirt 2004*). It has been proven in a number of experiments that the enhanced cold, drought, and pathogen tolerance of plants was caused by an improved activity of various antioxidant enzymes (*Pastori and Tippi, 1992; Király 2002*). The genetic overlap in the expression of the genes responsible for stress tolerance of plants during the sporophytic and gametophytic life cycles (*Frova, 1990*) enables to apply *in vitro* microspore selection to improve oxidative stress tolerance of regenerated DH plants.

In recent years an anther culture system suitable for routine application has been elaborated in the Plant Cell Biology Department of the Agricultural Institute (Centre for Agricultural Research, Hungarian Academy of Sciences). Moreover a new *in vitro* selection methodology was elaborated, which is based on the selection of haploid maize microspores in the presence of reactive oxygen species (ROS) generating agents, such as paraquat (Pq), t-butylhydroperoxide (*t-BHP*), menadion and methionine combined with riboflavine (MR) (*Ambrus et al., 2006*).

The aim of the present experiment was to elaborate a new *in vitro* selection methodology, which is based on the selection of haploid maize microspores in the presence of reactive oxygen species (ROS) generating agents. A single cross maize hybrid A18 (originating from a paraquat resistant DH line as a female parent and from another DH line with high androgenic capacity as the male parent) were used for *in vitro* selection. After the model experiment, a couple of single cross hybrids of breeding value were also tested in selection experiments. In order to determine whether the DH_1 progeny of Pq-selected lines really possessed greater oxidative stress tolerance (Pq and chilling tolerance) than the control genotype, various physiological tests were performed.

Materials and Methods

In vitro selection: Single cross hybrids of breeding value (H1, H2, H3) were used for *in vitro* selection and a single cross maize hybrid A18 (as a control genotype) was also involved in the experiments. The ROS progenitors (Pq, MR, and *t-BHP*) were applied at various concentrations and were added to the induction and regeneration media (*Ambrus et al., 2006*). The fertile DH regenerants have been self-pollinated. Various physiological parameters (chlorophyll *a* fluorescence, electrolyte leakage) of the DH1 plants were investigated.

Plant treatment and the determination of oxidative stress tolerance: The oxidative stress tolerance of the plants was examined in flotation experiments. Leaf discs 1.3 cm in diameter were cut using a cork borer and floated in Tris-HCl solution (50 mM, pH 7.6) containing 50 µM Pq, using 1 ml/leaf disc. The control solution did not contain Pq. After infiltration with the control or Pq-containing solution, the leaf discs were floated for 4 or 24 h with 400 µE m^{-2} s^{-1} PAR illumination (Darkó et al., 2009). The leaf discs were then used for physiological and biochemical analyses and measurements.

The oxidative stress tolerance of the plants was tested using the following techniques:

Measurement of chlorophyll *a* fluorescence induction: after floating the leaf discs for 4 h, measurements were done with a PAM 2000 fluorimeter (Walz, Effeltrich, Germany). The optimum quantum efficiency of photochemical system II (PS II), F_v/F_m, was calculated from the measured parameters using the equation $F_v/F_m = (F_m–F_0)/F_m$ (*van Kooten and Snel, 1990*).

Determination of ion leakage: after floating the leaf discs for 24 h, the electrical conductance of the solution was recorded using an Automatic Seed Analyser (ASA610, Agro Sciences, USA) (*Darkó et al., 2009*).

Calculation of tolerance index (TI): Tolerance index were calculated from the results of the physiological measurements in order to make it easier to evaluate the data and to compare the DH lines. The resistance factor of the chlorophyll fluorescence induction parameter was determined using the following equation: $TI_{Fv/Fm} = P_x[(Fv/Fm)_{Pq}/(Fv/Fm)_B]/DH[(Fv/Fm)_{Pq}/(Fv/Fm)_B]$ where Fv/Fm = the optimum quantum efficiency of PS II, DH = non-selected DH lines, P_x = genotypes selected using,

B = values measured in buffer in the flotation experiment, Pq = values measured in solution containing Pq.

The tolerance index calculated from the results of ion leakage measurements was determined using the equation: $TI_{Cond} = DH(Cond_{Pq}–Cond_B)/P_x(Cond_{Pq}–Cond_B)$, where Cond = the electrical conductivity of the solution in which the leaf discs were placed. For other legends, see above.

Chilling tolerance test at emergence: The 'cold test' of chilling tolerance at emergence was performed as described by *Herczegh* (1978), *Marton* (1992) and *Marton and Kőszegi* (1997). The kernels were germinated at two temperatures: at 22°C and in the cold, at 8°C. The time required for germination and the germination percentage were recorded at both temperatures and the germination index (GI) of each line, i.e. the ratio of the maximum emergence percentage to the number of days to germination, and the chilling tolerance index, calculated as $Px(GI_{T8°C}/GI_{T22°C})/DH(GI_{T8°C}/GI_{T22°C})$, were calculated from the results.

Results and Discussion

In vitro selection: First of all, we had to determine the embryogenic response of the F_1 hybrids. Seven F_1 hybrids of embryogenic response were examined (*Table. 1.)*. The model genotype (A18) had good haploid induction ability, while half of the tested F_1 lines had a low androgenic response. From three of the seven F_1 hybrids could produce fertile DH plants.

Hybrids	Nr. of the anthers plated	Anther responding %	MDS/ 100 plated anthers	Plant regeneration (%)	Nr. of fertile DH plants
A18	8000	50.0±2.5	124.0±6.2	14.0±0.4	28
H4	2000	5.0±0.25	8.0±0.04	1.5±0.07	0±0
H5	2000	4.2±0.21	9.2±0.46	2.3±0.11	0±0
H6	2000	4.8±0.24	6.0±0.3	0.5±0.02	0±0
H7	2000	10.0±0.5	12.0±0.6	2.5±0.12	0±0
H1	2000	42.1±2.1	99.0±4.95	10.0±0.5	15±0,75
H2	2000	48.7±2.4	98.3±4.91	13.0±0.65	7±0,35
H3	2000	45.2±2.26	99.2±4.96	12.9±0.64	10±0,5

Table 1.: Anther induction, embryo induction, regeneration potential and the number of the DH plants of the F1 hybrids and A18 (without ROS progenitors)

The *in vitro* selection was carried out only on H1, H2 and H3 hybrids with high androgenic capacity. Those ROS progenitors and those concentrations were used for present *in vitro* selection, which were successful in pervious experiment (Pq, MR, *t*-BHP) (*Ambrus et al., 2006*). The results of *in vitro* selection are demonstrated in *Table 2*.

Hybrids	Parameters	Pq		MR	*t*-BHP	
		0.5 μM	1.0 μM	10 μM	100 μM	1000 μM
A18	Anther response %	20.8±1.04	13.0±0.65	30.0±1.5	52.6±2.63	28.0±1.4
	MDS/100 plated anthers %	40.2±2.01	22.3±1.11	73.0±3.8	102.0±5.1	49.0±2.45
	Plant regeneration (%)	10.1±0.5	3.4±0.17	3.8±0.19	5.4±0.27	4.4±0.22
	Nr. of fertile DH plants	**10±0.5**	**5±0.25**	**10±0.5**	**8±0.4**	**2±0.1**
H1	Anther response %	21.0±1.05	19.7±0.98	34.0±1.7	21.3±1.06	0±0
	MDS/100 plated anthers %	45.0±2.25	34.0±1.7	57.3±1.86	46.0±2.3	0±0
	Plant regeneration (%)	6.6±0.33	5.8±0.29	0,3±0	7.3±0.36	0±0
	Nr. of fertile DH plants	**9±0.45**	**7±0.35**	**1±0.05**	**2±0.1**	**0±0**
H2	Anther response %	26.2±1.31	19.8±0.99	33.0±1.65	18.4±0.92	0.7±0.03
	MDS/100 plated anthers %	42.3±2.11	25.3±1.26	68.0±3.4	59.8±2.99	0.8±0.04
	Plant regeneration (%)	7.5±0.37	4.4±0.22	8.2±0.45	11.0±0.55	0±0
	Nr. of fertile DH plants	**4±0.2**	**3±0.15**	**4±0.2**	**6±0.3**	**0±0**
H3	Anther response %	33.0±0.16	28.3±1.41	20.8±1.04	31.0±1.55	9.4±0.2
	MDS/100 plated anthers %	64.0±3.25	62.3±3.11	50.6±2.53	64.2±3.21	19.9±0.99
	Plant regeneration (%)	11.2±0.56	6.8±0.34	9.0±0.45	8.9±0.44	5.4±0.27
	Nr. of fertile DH plants	**4±0.2**	**2±0.1**	**2±0.1**	**2±0.1**	**1±0.05**

Table 2.: The effects of the ROS progenitors on the anther response, embryo induction, frequency of plant regeneration and the number of fertile DH plants.

The experiment of data showed that the androgenic induction (anther response %), the frequency of microspore-derived embryo and calli (MDS/100 plated anthers), the frequency of the plant regeneration and a number of fertile DH regenerants were decreased in various degrees by all of the applied ROS generating compounds (*Table 2.).*

Physiological investigation of the Pq selected lines: In the present experiment sixteen from H1, seven from H2, six from H3 Pq selected genotype were used. Non-selected DH plants were used as control, moreover the original hybrids were also involved in the investigation.

Paraquat is a redoxiactive herbicide, which is produced by PSI in the leaves. In green leaves, ROS are highly reactive and can cause protein breakdown, chlorophyll bleaching finally membrane integrity and cell death. The optimum quantum efficiency of photochemical system II (PS II), F_v/F_m, was calculated from the measured parameters using the equation $F_v/F_m = (F_m – F_0)/F_m$. Tolerance index *(Table 3.)* were calculated from these data.

The electrolyte leakage was also measured. From these data tolerance index of the lines were also calculated (*Table 3.*).

Genotype	H1	H1D	**H1P1**	**H1P2**	H1P3	H1P4	**H1P5**	H1P6	H1P7
$TI_{Fv/Fm}$	0.8	1.0	**3.8****	**1.5***	1.1	1.6*	**3.2***	1.2	0.9
TI_{cond}	1.1	1.0	**1.2***	**1.3***	0.8	1.1	**6.9****	1.5*	0.9
Genotype	**H1P8**	**H1P9**	**H1P10**	**H1P11**	H1P12	H1P13	H1P14	H1P15	H1P16
$TI_{Fv/Fm}$	**12.6****	**2.8****	**2.1***	**15.9****	1.1	0.9	2.7**	1.4	0.9
TI_{cond}	**1.4***	**1.2**	**1.2***	**1.4***	1.0	1.1	1.1	1.2	1.1
Genotype	H2	H2D	H2P1	H2P2	**H2P3**	H2P4	H2P5	H2P6	**H2P7**
$TI_{Fv/Fm}$	1.2	1.0	1.3	1.3	**1.5***	1.0	1.4	1.1	**3.9****
TI_{cond}	1.1	1.0	1.2*	1.3*	**1.3***	0.8	1.3*	0.9	**1.3***
Genotype	H3	H3D	H3P1	H3P2	**H3P3**	H3P4	**H3P5**	H3P6	
$TI_{Fv/Fm}$	0.7	1.0	1.0	1.0	**3.1****	1.2	**1.8***	1.0	
TI_{cond}	0.9	1.0	1.1	1.1	**1.3***	1.2*	**1.7***	1.2*	

Tolerance index (TI) of DH_1 lines derived from Pq-selected microspores (H1P1-16, H2P7, H3P6) and from the original hybrids (H1, H2, H3)
compared to the non-selected DH (H1D, H2D, H3D) lines.
** P<0.05 ** P< 0.01 significantly different from non-selected control (H1D, H2D, H3D).*

Table 3.: Pq tolerance indicated by tolerance index

The determination of the chlorophyll *a* fluorescence induction parameter revealed nine (H1P1-2, H1P4-5, H1P8-11 and H1P14), (*) genotypes from H1, two (H2P3, H2P7) (*) from H2 and two (H3P3, H3P5) (*) from H3 which exhibited a significantly smaller reduction in photosynthetic activity compared to the relevant non-selected genotype in response to Pq as it

The determination of ion leakage in response to Pq treatment revealed seven lines (H1P1-2, H1P5-6, H1P8, H1P10-11) for H1, five (H2P1-3, H2P5, H2P7) for H2 and four (H3P3-6) for H3 where the electrical conductivity decreased to a significantly smaller extent than for the non-selected DH lines and the initial hybrids.

As regards the tolerance index, the results indicated that significantly higher tolerance index were recorded for seven lines (H1P1-2, H1P5, H1P8-11) from hybrid H1, two (H2P3, H2P7) from H2 and two H3P3, H3P5) from H3 than for the non-selected DH lines or the initial hybrids for both traits examined.

Chilling tolerance test at emergence:

The ’cold test’ of chilling tolerance at emergence was also tested. The time required for germination and the germination percentage were recorded at both temperatures and the germination index of each line, i.e. the ratio of the maximum emergence percentage to the number of days to germination (data not shown), and the chilling tolerance index, were calculated from the results (*Table 4).*

Genotype	H1	H1D	H1P1	**H1P2**	H1P3	**H1P4**	**H1P5**	**H1P6**	**H1P7**
TI	0.3	1,0	0.5	**3.6****	0.9	**2.5****	**2.2****	**2.9****	**3.5****
Genotype	**H1P8**	**H1P9**	**H1P10**	**H1P11**	H1P12	**H1P13**	**H1P14**	**H1P15**	**H1P16**
TI	**3.2****	**3,8****	**6.1****	**2.9***	0	**3.8****	**2.6****	**2.0***	**1.7***
Genotype	H2	H2D	**H2P1**	**H2P2**	H2P3	H2P4	H2P5	**H2P6**	**H2P7**
TI	0.1	1.0	**1.5***	**2.0***	1.5*	0.3	1.3	**1.6***	**1.7***
Genotype	H3	H3D	**H3P1**	H3P2	**H3P3**	**H3P4**	**H3P5**	**H3P6**	
TI	1.3	1.0	**1.5***	1.1	**1.6***	**1.5***	**2.0***	**1.9***	

Chilling tolerance indicated by tolerance index (TI) of DH_1 lines derived from Pq-selected microspores (H1P1-16, H2P7, H3P6) and from the original hybrids (H1, H2, H3) compared to the non-selected DH (H1D, H2D, H3D) lines.
** P<0.05 ** P<0.01 significantly different from non-selected control (H1D, H2D, H3D).*

Table 4.: Chilling tolerance index of the Pq selected DH1 lines.

With respect to the chilling tolerance of the selected lines at germination, it was found that the germination percentage decreased and the time required for germination increased in response to cold treatment. When the tolerance index were calculated (see Material and Methods) for chilling tolerance, higher values than those of non-selected DH lines or the initial hybrids were recorded for 13 lines (H1P2, H1P4-11 és H1P13-16) from H1, five (H2P1-3, H2P6-7) from H2 and five (H3P1, H3P3-6) from H3.

In summary, a total of 10 selected genotypes, six from hybrid H1, two from H2 and two from H3, proved to have better tolerance index in terms of both Pq-induced oxidative stress and chilling stress at germination than the non-selected DH lines or the initial hybrids.

The results of all the experiments indicate that the *in vitro* selection of microspores can be successfully performed using compounds inducing the formation of reactive oxygen species, and that fertile DH plants resistant to oxidative stress can be regenerated. The technique elaborated works not only on model genotypes, but also for the *in vitro* selection of genotypes suitable for breeding purposes. It can thus be applied to develop valuable breeding material possessing good adaptability.

Acknowledgement

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Tests on the herbicide tolerance of maize in Martonvásár

Péter Bónis, Tamás Árendás, Zoltán Berzsenyi, L.Csaba Marton
Crop Production Department, Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences,
2462 Martonvásár, Brunszvik u. 2.
e-mail: bonis.peter@agrar.mta.hu

Abstract

Research on weed control and herbicide sensitivity began in Martonvásár immediately after the appearance of herbicides. The experiments set up in the institute later served as a model for herbicide application in large-scale maize production in Hungary. The work done in the 1950s and 1960s by *István I'só, Zoltán Barabás, Béla Győrffy* and *J. László Szabó* proved that selective chemical herbicides caused no damage to maize, or only under certain conditions, and did not reduce the yield. The results achieved by these pioneers and the methods they elaborated formed the starting point for today's research on herbicide sensitivity. Experiments are regularly performed by *Zoltán Berzsenyi* and his colleagues to test the new, up-to-date herbicides constantly being developed by manufacturers.

The results currently achieved by scientists in Martonvásár will be presented using a herbicide tolerance experiment set up in 2013 as an example. When the responses of the parental components of 48 Martonvásár maize hybrids to 5 pre- and 11 post-emergence herbicides were tested, it was found that no damage was caused by the pre-emergence treatments, while relatively strong symptoms were observed for three of the post-emergence herbicides, averaged over the genotypes.

Introduction

Research carried out in Martonvásár made a considerable contribution to the elaboration of a suitable production technology for maize hybrids in Hungary. Compilations of the results of maize production experiments set up in various maize-growing areas of Hungary were published by *I'só* (1958, 1962, 1966, 1969) and *Bajai* (1979). In addition to experiments on sowing date, plant density, mineral fertilisation and hoeing, it became clear over the years that the chemical herbicides that began to appear in the 1950s included many that were very selective and caused no damage to maize, or only under certain conditions, while being extremely effective against weeds. In Martonvásár *I'só (*1958a)*, Barabás* (1955)*, Csongrádyné* (1958) and *Győrffy* (1962) carried out pioneering work on herbicides and reported the symptoms of damage observed on maize. *Kükedi* (1970) also set up experiments on grain and fodder sorghum with the herbicides used in maize.

Experiments on herbicides of the atrazine and thiocarbamate type and on compounds in the carbamide and chloracetanilide groups were carried out in Martonvásár by *Győrffy* (1969)*, Győrffy* and *Szabó* (1969) and *Berzsenyi* and *Győrffy* (1989). The re-

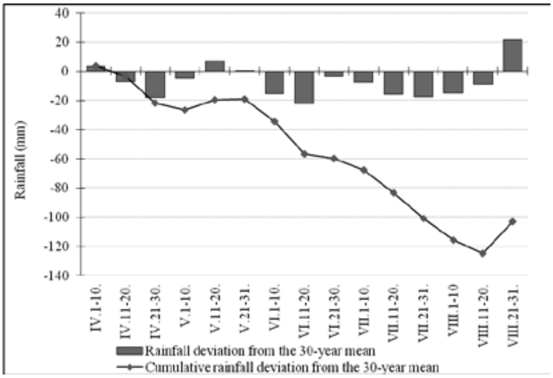

Fig.1. Trend in rainfall deficiency during the vegetation period of maize. Martonvásár, 2013

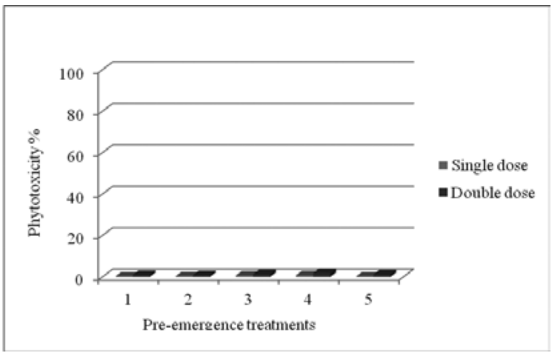

Fig. 2. Extent of phytotoxic damage caused by pre-emergence herbicide treatments, averaged over the genotypes, in a herbicide tolerance experiment. Martonvásár, 2013.

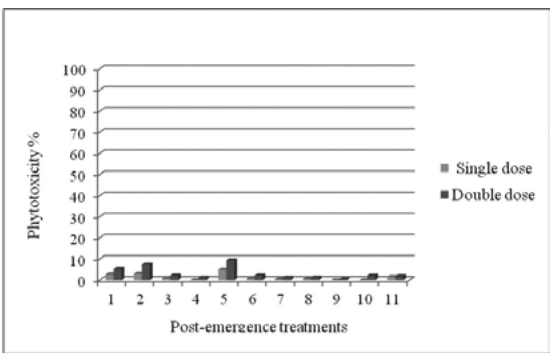

Fig. 3. Extent of phytotoxic damage caused by post-emergence herbicide treatments, averaged over the genotypes, in a herbicide tolerance experiment. Martonvásár, 2013.

sults achieved with new, wide-spectrum herbicides were published by *Berzsenyi et al.* (1994) and *Bónis et al.* (2000, 2011, 2013).

Herbicides form an important part of integrated weed control. The continuous testing of new maize genotypes and herbicides is essential for the elaboration of reliable production technologies.

Materials and methods

The methodology of field studies on herbicide tolerance is presented on the basis of a split-plot experiment set up in 2013. The weather in this year was dry, with low rainfall supplies (*Fig. 1*). Compared with the 30-year mean, only 32% of the normal rainfall was recorded during the maize growing season, while the number of very hot days was 9 more than the long-term mean for this period. In the small-plot field experiment 5 pre-emergence and 11 post-emergence herbicides or herbicide combinations were tested by applying them with a plot sprayer at the maximum permitted dose and at twice this rate. The experiment was carried out in two replications in such a way that there was an untreated control plot for each treated plot. The pre-emergence herbicides were applied before the maize emerged, and the post-emergence herbicides when the maize plants had reached the 8-leaf stage of development. The active ingredients in the pre-emergence herbicides were as follows: *1) mesotrione + S-metolachlor + terbutylazine; 2) isoxaflutole + cyprosulfamide; 3) isoxaflutole + thiencarbazone-methyl + cyprosulfamide; 4) dimethenamide-p + terbutylazine; 5) pendimethalin*. The post-emergence herbicides had the following active ingredients: *1) nicosulfuron; 2) mesotrione + nicosulfuron; 3) tembotrione + isoxadifen-ethyl; (4) topramezone + di-*

camba; 5) foramsulfuron + isoxadifen-ethyl; 6) prosulfuron; 7) dimethenamide-p + terbutylazine + topramezone; 8) pendimethalin + topramezone; 9) bentazone + dicamba; 10) mesotrione + terbutylazine; 11) pethoxamide + terbutylazine. Visible phytotoxic symptoms were scored as a percentage on a 0–100 scale 14 days after treatment.

Results and discussion

The pre-emergence herbicides caused few if any symptoms on the maize genotypes (*Fig. 2*). Spraying was carried out on 11 May, after which the 15–20 mm rain required for the manifestation of the herbicide effect fell within two weeks.

The post-emergence herbicides caused more severe damage to the maize genotypes than the pre-emergence herbicides (*Fig. 3*).

The most severe symptoms were observed on maize plants treated with herbicides 1, 2 and 5 (*Fig. 3*). In these three treatments, doubling the dose of herbicide led to almost double the extent of visible damage. In general, the inbred lines tolerated the herbicide treatments well.

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Importance of the Mindenszentpusztai Yellow Dent heterosis source for Hungarian and European maize breeding

Géza Hadi, János Pintér, L.Csaba Marton
Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
2462 Martonvásár, Brunszvik u. 2.
hadi.geza@agrar.mta.hu

Abstract

Mindszentpusztai Yellow Dent arose from the Leaming variety and played an important role during the first decades of maize breeding in Hungary and in Europe as a whole. The hybrids developed using this source were sown on a total area of over 25 million hectares. It is hoped that lines originating from the Mindszentpusztai Yellow Dent heterosis source will continue to contribute to the breeding of hybrid maize in the future.

Introduction

There is no methodological alternative to hybridisation in maize breeding. Sources of heterosis are needed, however, for the development of commercial hybrids. Although the maize species has great morphological and genetic richness, there are nevertheless only a limited number of heterosis sources.

In the United States of America two fundamental sources were used (Reid Yellow Dent and Lancaster), together with three supplementary sources (Min. 13, Northwestern Dent and Leaming). The contribution of the supplementary sources of heterosis was low, being no more than 10%.

In Hungary Mindszentpusztai Yellow Dent, which was derived from Leaming, is of special importance. The Leaming variety probably arose from genetic mixing between Mexican highland races (Cónico, Chalqueño) and Corn Belt Dent races.

Results and discussion

The Leaming family grew the Leaming variety in the southern part of Ohio from the 1830s onwards, but the final development and stabilisation of the variety can be attributed to the work of Jacob and Chester Leaming, who bred a variety with outstanding importance even on a national scale between 1855 and 1890. The Leaming variety was exhibited at the Chicago Maize Show in 1896, where it won a silver medal. It was probably brought to Hungary by the breeding and seed trading company of Ödön Mauthner in the 1890s, who then multiplied and listed it. The grandfather of Endre Pap obtained a sample of this variety in the 1890s and grew it for years on his estate in the neighbourhood of Baja. After his death it was transferred to his son's estate in Mindszentpuszta, where it came to the attention of Endre Pap.

Breeding was begun in Mindszentpuszta in 1917, and the new variety was registered as Mindszentpusztai Yellow Dent (MPS) in 1928. Its spread and its contribution to Hungarian maize production are illustrated in Table 1.

Period	Open-pollinated varieties	Estimated share of the sowing area (%)
1919–1937	Bánkúti Dent	30
	American Yellow Dent	25
	Old Hungarian Yellow Flint	15
	„F" Golden Dent	7
	Cinquantino (Putyi,Székely)	5
	Pignoletto	5
	Gyérei-Dudás White Dent	3
	Paduan (Pál Kárász type, MPF)	2
	Mindszentpusztai Yellow Dent	1
	Other varieties	7
1938–1962	„F" varietes	50
	Golden Flood Yellow Dent	11
	Mindszentpusztai Yellow Dent	9
	Mindszentpusztai White	6
	Putyi	5
	Bánkúti Dent	4
	Szegedi Yellow Dent	3
	Other varieties	12

Table 1. Contribution of Mindszentpusztai Yellow Dent to Hungarian maize production (1919–1963)

Between 1931 and 1933 Endre Pap evaluated the level of breeding gain and found that a yield increase of around 20% had been achieved in the first 10 years and around 2% in the last 5 years. This did not satisfy Endre Pap, so he looked for new breeding methods. He hoped to fix the excellent yield potential of the variety by means of inbreeding, and thus developed over 200 inbred lines from 1933 onwards. These lines included 01, 014, 0118B, 156 and probably 0118a, which later made an extremely contribution to the success of Hungarian maize breeding. Up till 1983 a total of 22 hybrids developed from MPS lines were cultivated, the seed of which was sufficient for a sowing area of 18.7 million ha (*Table 2*), more than 15 times the maize growing area in Hungary. The most important of these was Mv 5, the first double-cross hybrid registered in Europe, developed using 3 MPS lines and an American line. From 1953 onwards seed sufficient for nearly 3 million ha was sold in Hungary over the course of 16 years. Similar success was achieved with Mv 1, the second hybrid registered in Europe in 1955, and its close relation Mv DC 602, for which seed sales were sufficient for areas of around 4.5 million ha and 2.5 million ha, respectively.

Hybrid	Pedigré	Year of registration	Cultivation period	Total seed sales (ha)
Mv DC5	(0118b*156)*(C5*014)	1953	1965-71	2 796 479
Mv DC1	(WF9*M14)*(C5*014)	1955	1960-71	4 497 800
Mv MC39	((WF9*M14)*(C5*014)*(0118b*156)	1957	1960-63	157 058
Mv MC 40	((A96*A34)*(0118b*156))*(Min6*01)	1959	1961-73	1 497 012
Mv DC42	(Iregi*L17)*(Min6*01)	1960	1962-67	43 261
Mv DC57	(0118b*156)*(A96*A34)	-	1962-64	22 388
Mv DC58	(0118b*156*(Min6*01)	-	1963	25 776
Mv DC48	(C5*WF9)*(0118b*156)	1961	1962-69	758 616
Mv26	(C5*014)*Mindszentpusztai white	1961	1965-83	660 804
Mv DC59	(C5Ms*N6)*(0118b*156)	1962	1965-75	1 854 216
Mv DC 602	(WF9*N6)*(C5*014)	1964	1967-77	2 430 825
Mv DC 502	(156*OH43)*(C5*014)	1966	1968-70	26 184
Mv DC520	(156*N6)*(C5*014)	1968	1969-81	509 700
Mv SC 530	156*N6	1968	1969-78	604 667
Mv TC431	(156*N6)*C5	1970	1968-78	620 234
Mv TC 596	(156*N6)*HMv850	1970	1971-82	953 230
Mv SC 380	156*W153R	1972	1971-78	410 623
Mv SC 580	156*B14	1972	1973-81	477 813
Mv DV405	156*B18/4	1974	1973-80	173 425
Mv SC 429	15Rf*HMv401	1976	1975-81	124 584
Mv SC424	156Rf*F564	1976	1978-79	3 918
Mv DC350	(0118a*A90*(156*W153R)	-	1975	11 300
Total				**18 659 913**

Table 2. Martonvásár maize hybrids developed using parental lines of MPS origin (1953–1983)

Lines of MPS origin were of outstanding importance in Hungarian maize breeding. Between 1959 and 1978 over 60% of the maize growing area was sown to hybrids developed from MPS lines (Table 3).

Year	Maize growing area in Hungary (ha)	Hybrids developed using MPS lines	
		Ha	%
1956	1 162 925	11 629	1.0
1957	1 346 575	40 398	3.0
1958	1 304 000	39 120	3.0
1959	1 358 000	380 240	28.0
1960	1 401 000	868 620	62.0
1961	1 304 000	873 680	67.0
1962	1 288 000	1 030 384	79.9
1963	1 288 847	1 220 360	94.5
1964	1 208 000	1 206 475	99.9
1965	1 217 980	1 192 424	97.9
1966	1 237 000	1 199 892	97.0
1967	1 237 000	1 159 069	93.7
1968	1 258 441	1 097 325	87.2
1969	1 255 140	1 065 552	84.9
1970	1 188 831	901 110	75.8
1971	1 321 000	915 453	69.3
1972	1 392 000	1 087 368	78.1
1973	1 460 764	1 025 312	70.2
1974	1 461 493	925 066	63.3
1975	1 412 540	713 310	50.5
1976	1 339 000	522 210	39.0
1977	1 281 000	457 317	35.7
1978	1 183 000	315 618	24.6
1979	1 352 000	209 560	15.5
1980	1 229 000	98 320	8.0
1981	1 163 000	52 338	4.5
1982	1 130 000	31 640	2.8
1983	1 107 000	4 408	0.4

Table 3. Contribution of hybrids developed with lines of MPS origin to Hungarian maize production (1956–1983)

Lines derived from Mindszentpusztai Yellow Dent, or improved variants of these lines, were still used in Martonvásár to develop registered hybrids after 1983, and these were grown not only in Hungary, but also in Germany, Poland, Russia and Ukraine (*Table 4*). In Ukraine, for example, an annual area of a million hectares was sown with seed of ODMA 310 over a period of nearly 10 years.

Hybrid	Pedigree	Year of registration	FAO number	Utilisation
Bermador	(Co158*HMv651)*CM7/Mv	1983	240	Silage
Bermasil		1987	290	Silage
Bermarit		1988	260	Silage
Mv To 289 TC		1989	286	Grain
Mv TC 287		1999	187	Grain
Mv NK 333		1993	333	Silage
ODMA 310		1994	310	Silage
Mv TC 272		1997	270	Grain
Mv 273		1999	290	Grain
Marusya		2013	180	Silage

Table 4. Hybrids developed from MPS lines and cultivated in Hungary (1983–1994)

Lines originating from MPS or their improved variants were used to develop hybrids in many countries. At least six hybrids involving lines related to MPS were registered in Moldavia and four or five in Slovakia.

The genetic material of MPS was also successfully used by INRA and Mais Angevin in France.

Hybrids developed using lines of Mindszentpusztai Yellow Dent produced especially high yields when grown at moderate plant density (35–55 thousand plants ha^{-1}), and had excellent adaptability and good drought tolerance. High rates of fertiliser were not required for their cultivation.

The first 30 years of hybrid maize in Hungary

Géza Hadi, János Pintér, L.Csaba Marton
Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
2462 Martonvásár, Brunszvik u. 2.
hadi.geza@agrar.mta.hu

Abstract

The first maize hybrids to be registered in Hungary, and in the whole of Europe, were bred in Martonvásár. The seed industry and seed trade quickly became established, so by 1962–1963 hybrid maize was sown on the major part of the maize-growing area in Hungary.

Between 1953 and 1983 seed sufficient for 20 million hectares was sold in Hungary and for nearly 25 million hectares in Europe as a whole.

Hybrids developed using lines C5, 156, O14, N6 and WF9 were popular in Hungary, as they not only had excellent yield potential, but were also well adapted to the natural fertility of the soil and had good resistance to head smut infection.

Introduction

As the result of industrialisation, many country people ceased to work in agriculture. Due to the lack of labour, it became necessary to elaborate mechanical techniques for cultivating crops.

The spread of hybrid maize provided an excellent opportunity for industrialising maize production.

Hybrid maize breeding is a process in which homozygous lines are developed by self-pollination, after which they are tested for their general usefulness. SC, TC and DC hybrids are then developed with levels of yield potential and biotic and abiotic adaptability that make them suitable for commercial production.

The parental lines are multiplied in isolated fields. The seed produced is then multiplied in the requisite manner on an industrial scale, which can be repeated every year, as required. European law only permits the sale of officially certified seed from registered hybrids. Registered varieties are required if hybrids are to be grown.

Results and discussion

The first maize hybrids registered in Europe (Mv 5 in 1953 and Mv 1 in 1955) were bred by Endre Pap in Martonvásár (*Table 1*).

Hybrid	Pedigree	Year of registration	Total sales of seed (ha
Mv DC 5	(0118B*156)*(C5*014)	1953	2 769 479
Mv DC 1	(WF9*M14)*(C5*014)	1955	4 497 800
Mv MC 39	[(WF9*M14)*(C5*014)*(0118b*156)]	1957	157 058
Mv MC 40	[(A96*A34)*(0117b*156)*(Min6*01)]	1959	1 497 012
Mv DC 42	(Iregi*L17)*(Min6*01)	1960	43 262
Mv DC 57	(0118b*156)*(A96*A34)	-	22 388
Mv DC 58	(0118b*156)*Min6*01)	-	25 776
Mv DC 48	(C5*WF9)*(0118b*156)	1961	758 616
Mv 26	(C5*014)*Mindszentpusztai White	1961	660 804
Mv DC 59	C5CmsC*N6)*(0118b*156)	1962	1 854 216
Mv DC 602	(WF9*N6)*(C5*014)	1964	2 430 825
Mv DC 502	(156*OH43)*(C5*014)	1966	26 184
Mv SC 520	(156*N6)*(C5*014)	1968	509 700
Mv TC 521	(C5*N6)*B125	1968	50 896
Mv SC 530	156*N6	1968	604 667
Mv SC 620	WF9*N6	1968	254 569
Mv TC 651	(WF9*N6)*c103	1969	34 016
Mv TC 290	(0118a*W153R)EP1	1970	68 429
Mv SC 370	156*A90	1970	105 898
Mv TC 431	(156*N6)*C5	1970	620 234
Mv TC 540	(Be03b*N6)*B125	1970	190 880
Mv SC 570	C5*N6	1970	42 368
Mv TC 596	(156*N6)*HMv850	1970	953 230
Mv TC 610	(C5*N6)*HMv850	1970	64 555
Mv TC 281	(0118a*A90)*EP1	1971	55 911
Mv DC 460	(B125*B18/4)*(Be03b*N6)	1971	406 366
Mv SC 660	N6*C103	1971	30 931
Mv MSC 262	(0118a*0118aR2)*EP1	1972	17 121
Mv SC 380	156*w153R	1972	410 623
Mv SC 580	156*B14	1972	477 813
Mv SC 587	A374h*CE187	1972	27 717

Hybrid	Pedigree	Year of registration	Total sales of seed (ha
BEMA 250	(0118aR2*W153R)*(Dbe19*DBe42)	1974	179 549
Mv SC 405	156*B18/4	1974	173 425
Mv DC 350	(0118a*A90)*(156*W153R	-	11 300
Mv MSC 342	(HMv480*W153R)*A90	1976	36 299
Mv SC 424	156*F564	1976	3 918
Mv SC 429	156*HMv404	1976	124 584
Mv TC 296	(0118aR2*W153R)*HMv404-C	1978	40 888
Mv SC 484	F564*A632	1978	19 865
BEMA TC 210	(F7CmsC*F2)*CM7/Mv	1980	19 151
Mv SC 434	HMv403*MA61A47D	1981	15 623
Total:			**20 304 230**

Table 1. Martonvásár hybrids and their sowing area (1953–1983)

In later years, Martonvásár continued to play a leading role in hybrid maize breeding. Up till 1983 Endre Pap and his colleagues, István Kovács, András Csetneki, Károly Kovács, Márton Herczegh, Bertalan Dolinka and István Manninger, developed 42 registered hybrids, the seed of which was produced and marketed initially by the Martonvásár seed plant, and later by other seed plants that were gradually built throughout the country.

It can be seen from Table 1 that the most successful hybrid in Hungary was Mv 1, from which sufficient seed was produced to sow 4.5 million hectares, while seed for a further 2.5 million hectares was produced from a closely related hybrid, Mv DC 602.

The first maize hybrid registered in Europe in 1953 was Mv 5. As this was an earlier maturing hybrid (FAO 350) it was not as popular as Mv 1 at a time when maize was ear harvested. Nevertheless, sufficient seed to sow almost 3 million hectares was produced during the 16 years when it was registered.

It is clear from the table that hybrid maize spread throughout Hungary within 7 years, and sufficient seed to sow more than 20.3 million hectares was sold up to 1983. Many of the hybrids listed in the table were also registered abroad, so the total quantity of seed sales was able to satisfy the seed requirements of nearly 25 million ha.

It is worth considering the genotype composition of the hybrids used between 1953 and 1983 (*Table 2*).

The most popular line was C5, which made a contribution of over 18%, but the lines 156, 014 and 0118b, bred by Endre Pap from Mindszentpusztai Yellow Dent, were also very popular. The largest quantity of seed in the world was developed on line WF9, which was also extremely popular in Hungary. The popularity of N6 could be attributed to the fact that it was resistant to head smut, which often caused epidemics on unfertilised areas, and that it passed this trait on to its hybrids. Among the lines developed later, only HMv 850 became popular (2.51%), as it produced 2–3 ears and had good adaptability, a trait that was inherited in its progeny. It is interesting to note that the Lancaster lines (OH43, A619, C103, Mo17) were popular in the USA but not in Hungary. Instead, the Mindszentpusztai Yellow Dent lines belonging to the Chester Leaming group played a decisive role.

Hybrids developed on these lines dominated maize production in Hungary between 1953 and 1983.

Line	Origin	Sowing area calculated on the basis	Frequency of the genetic contribution of the line (ha) (%)
C5	W23 =Golden Glow	3 791 562	18, 67
156	MPS C_0	3 035 966	14,95
014	MPS C_0	2 750 080	13,54
N6	Hayes Golden	2 276 511	11,21
WF	Wilson Farm Reid	2 077 230	10,23
0118b	MPS C_0	1 590 761	7,83
M 14	BR 10*R 8	1 144 082	5,63
HMV850	U.W.W.30(HY-2 rel.)	508 893	2,51
01	MPS C_0	391 512	1,93
Min.6	Min. No. 13	385 068	1,90
M.p.f.	Mindszentpusztai White Flint	330 402	1,63
W153R	(I.a.153*W8)*I.a 153	289 428	1,43
B 14	BSSS C_0	238 907	1,18
B 125	Bánkuti Early Dent	222 229	1,09
A 96	64*H	192 724	0,95
A 34	Rustler (C5)	192 724	0,95
B 18/4	A374h*A118,A374h rec.	188 054	0,93
Be03b	Béllyei Yellow Dent	149 062	0,73
EP 1	Lizzargarote	70 730	0,35
HMV 401	N6*M5226, N6 rec	62 292	0,31
0118Ar2	0118 mutant	59 389	0,29
A 90	64*15-28	56 137	0,28
Dbe 19	Pommermais	44 887	0,22
Dbe 42	Sammerlaktion Lübears	44 887	0,22
0118a	MPS C_0	38 190	0,19
C 103	Lancaster Sure Crop	32 474	0,16
HMV404-C	(156*C131A)*156BC_3	20 444	0,10
A 374h	A 374 rec.	13 858	0,07

Line	Origin	Sowing area calculated on the basis	Frequency of the genetic contribution of the line (ha) (%)
CE 187	C.I. 187-2 rec	13 859	0,07
Iregi	Iregi 12-week flint	10 815	0,05
L 17	Poland O.P.V. (Wielkopolanka?)	10 815	0,05
F 564	F 564 rec.	9 932	0,05
A 632	(Mt42*B14)*B14[3]	9 932	0,05
CM7/MV	CM 7 rec.	9 575	0,05
HMV 480	W153R rec.	9 075	0,04
HMV 403	A632*M5226, A 632 rec	7 811	0,04
MA61A47D	B37*W 79A	7 811	0,04
OH 43	OH40B*W8	6 546	0,03
F2	Lacaune	4 787	0,02
F7	Lacaune	4 788	0,02
Total:		**20 304 230**	**100,00**

Table 2. Parental lines used in Martonvásár hybrids (1953–1983)

Comparison of the activity of the glutamine synthetase enzyme in wheat (Triticum aestivum L.) and maize (Zea mays L.)

Zoltán Nagy[1], Tomas Zakar[2], Edit Németh[2], Attila Pécsváradi[2], L.Csaba Marton[1]
[1]*Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences*
2462 Martonvásár, Brunszvik u. 2.
[2]*Department of Plant Biology, Faculty of Science and Informatics, University of Szeged*
6726 Szeged, Közép fasor 52.
e-mail: nagy.zoltan.mgi@agrar.mta.hu

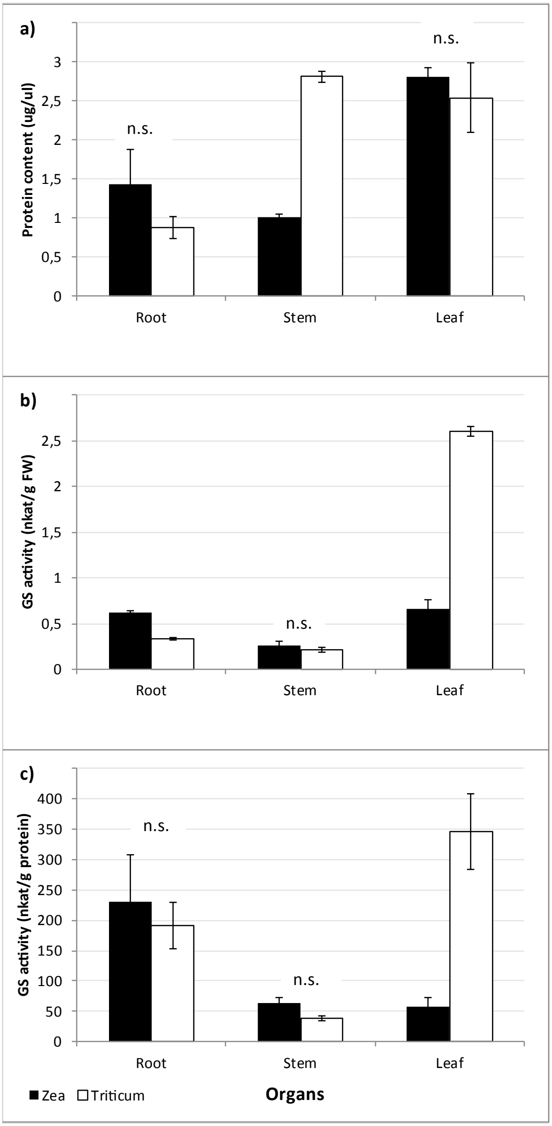
Abstract

In summary it can be said that glutamine synthetase is an indicator of the state of the nitrogen metabolism in plants. In wheat the highest enzyme activity was recorded in the leaves. This enhanced nitrogen metabolism could be attributed to the photorespiration characteristic of C3 plants. To ensure satisfactory plant development, the surplus ammonia liberated by this process needs to be fixed as efficiently as possible. In the case of maize, similar activities were measured in the roots and leaves. Enhanced activity is required in the roots for nitrogen uptake, while in the leaves the presence of GS is important due to its role in protein biosynthesis. This, however, requires a lower activity level than that needed to fix the ammonia generated by photorespiration in wheat.

Introduction

Glutamine synthetase (E.C. 6.3.1.2, GS) is one of the key enzymes of the nitrogen metabolism, and is found in all living beings, from prokaryotes to eukaryotes. It plays a decisive role in plants in the incorporation of inorganic nitrogen, taken up from the soil or fixed by symbiotic bacteria, into organic molecules, and in the protein-amino acid metabolism. The substrates of GS are ammonia and glutamate, while the reaction product is glutamine. ATP is required for the reaction and the presence of two-valence metal ions is essential (*Miflin and Habas, 2002*).

Investigations on the fixation of carbon dioxide were carried out on two diverse plants. In the case of bread wheat (*Triticum aestivum* L.), which has the C3 type of photosynthesis, the concentration of atmospheric carbon dioxide determines the processes taking part in the dark phase of photosynthesis, since the concentration of the CO_2 serving as the substrate for ribulose-1,5-biphosphate carboxylase/oxygenase (Rubisco) within the cells and chloroplasts is proportionate to the concentration outside the plant. In the C4 plant maize (*Zea mays* L.), on the other hand, the CO_2 fixation in the plant is spatially separated from the enzymes belonging to the Calvin cycle. CO_2 fixation takes place in the mesophyll cells, where it is bound to pyruvate by phosphoenolpyruvate carboxylase (PEP-carboxylase), resulting in the formation of malate. The latter then acts as the substrate of malate



n.s.: differences between the data were non-significant at the p≤0.1% level.

Fig. 1. Protein content in the tissues of 1-week-old wheat (☐) and maize (■) plants (a), and the glutamine synthetase activity of the plants in terms of fresh weight (b) and protein content (c).

dehydrogenase in the bundle sheath parenchyma cells, where CO_2 is released and pyruvate is formed. In this case the CO_2 concentration is higher in cells involved in the carboxylation of ribulose (*Edwards and Voznesenskaya, 2011*).

Rubisco has two different functions, being capable of both oxidation and carboxylation, depending on the quantity of substrates available. At high CO_2 concentration there is a greater proportion of carboxylation, while low CO_2 concentration induces oxidation, and phosphoglycolate is formed as well as phosphoglycerate. The connection between phosphoglycolate and the Calvin cycle is know as the photorespiration cycle, a process in which ammonia is formed (*Ogren, 1984*). The presence of NH_3 within the cell is unfavourable for various reasons, primarily due to its toxic nature, but also because it is able to diffuse through the membranes, leading to the loss of the nitrogen taken up and fixed by the plant. In C3 plants photorespiration is much more intense than in C4 plants due to the lower CO_2 concentration. GS plays an important role in fixing the ammonia arising in the course of photorespiration. In the present work the total protein content and GS activity were compared in various tissues of wheat and maize plants, in order to discover whether differences in the way carbon dioxide is fixed were reflected in the measurable activity of GS.

Materials and methods

The plant material consisted of wheat (*Triticum aestivum* L.) and maize (*Zea mays* L.). After germination the plants were grown for a week in modified Hoagland solution in a plant growth room. Samples were taken on the seventh day from the roots, stems and leaves (0.5 g). The water-soluble protein content of the samples was determined using the method of Bradford, and the GS activity by means of the modified synthetase reaction, using a spectrophotometer. The results were statistically analysed using Microsoft Office Excel software. For each measurement the data were compared using a two-sample *t*-test. Significance was tested at the p≤0.1% level, and non-significant differences were indicated as n.s. (*Pécsváradi et al., 2009*).

Results and discussion

The results of the measurements are illustrated in *Figure 1*. With the exception of the stems, the protein contents (*a*) in the organs of wheat and maize did not differ significantly, and the protein contents of the stem and leaves of wheat were also similar. Although in both species the stem is covered by the leaf sheath, stalk differentiation commenced earlier in maize, while in wheat the high protein content was indicative of active cell division. The quantity of water-soluble proteins is correlated with the assimilation ability and developmental stage of the given organ. Young, metabolically active organs have high protein content, which is why the highest values were recorded in the leaves, where the process of photosynthesis takes place.

The GS activity values were expressed in terms of fresh plant weight (*b*) and the protein content of the tissues (*c*), as in some cases the values given in terms of protein content are not really characteristic of the given tissue. In the present case, however, there was no great difference between the two data series, except that slightly higher mean values were obtained in the stems for maize than for wheat due to the different protein concentrations. Nevertheless, the difference between the two species was not significant. The greatest difference in the levels of GS activity in wheat and maize were detected in the leaves. Differences were also observed in the roots when enzyme activity was given in terms of fresh weight, but these differences were not as great as in the leaves. It can thus be concluded that the type of photosynthesis probably plays a role in the difference in GS activity, as the GS activity in the leaves of the C3 species wheat was almost five times as great as that in the C4 species maize, compared with a difference of slightly less than two times in the roots. As mentioned in the introduction, the level of photorespiration is connected with the functioning of glutamine synthetase, as it is this process that is involved in the nitrogen metabolism and differs in the two species. The surplus ammonia arising due to the Rubisco oxidase activity in wheat is fixed by glutamine synthetase. This is unnecessary in maize, as the level of photorespiration is low in C4 plants.

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Effect of drought on yield components of maize

Tamás Spitkó, Zoltán Nagy, Gábor Halmos, Csaba L. Marton
Agricultural Institute, Centre for Agricultural Research, HAS
Maize Breeding Department
H-2462 Martonvásár
e-mail: spitko.tamas@agrar.mta.hu

Abstract

When investigating drought tolerance, it must not be forgotten that drought stress is a complex phenomenon exhibiting quite different characters in different years and locations. For this reason, the plant response to drought is also a complex process. In both experimental years (2011 and 2012) a long period of drought combined with very hot weather was recorded in June and July, during flowering, and even in August, during the grain-filling period, resulting in severe water deficit in the plants. The plants in the control plots were maintained in a satisfactory condition by irrigating them to simulate optimum rainfall supplies. The drought tolerance of plants in the non-irrigated plots was analysed in terms of flowering synchrony and yield components. It could be concluded from the results that in response to long-term water deficit the period between tasselling and silking became longer, while the analysis of yield components revealed the greatest reductions in the number of kernels per ear and in the proportion of seed set. At the same time, the thousand-kernel weight did not increase on the irrigated plots, which could be attributed to the fact that irrigation was only carried out before and during flowering, and immediately after it, while no additional water was supplied during grain filling. As the degree of proterandry increased, there was a decline in the grain yield, confirming that the analysis of this trait could be a way of predicting drought tolerance. Considerable differences in drought tolerance were observed between the genetic materials included in the analysis, suggesting the presence among these parental lines and hybrids of genotypes resistant to long-term water deficit, suitable for cultivation under dry conditions.

Introduction

Plants may employ various strategies to overcome the consequences of drought: (1) improvements in water use efficiency, (2) escape, i.e. completing reproductive processes, the development phase most sensitive to water deficit, before the onset of dry weather, (3) an improvement in the ‘per se’ drought tolerance of the plant (Ribaut et al., 2009).

Maize is most sensitive to drought during the flowering period. The most important symptoms of drought damage during flowering are the following: delay in inflorescence development, flowering asynchrony, tassel blasting, reduction in pollen fertility and viability (possibly complete sterility), reduction in pistil receptivity (in some cases complete sterility), abortion of embryos (Westgate and Boyer, 1985).

It has frequently been observed that during drought the interval between male and female flowering (proterandry) increases (DuPlessis and Dijkhuis, 1967). This is generally caused by the relative delay in the appearance of the pistils compared with that of the tassels (which is less affected by drought). Proterandry is thus a more important trait in determining the drought tolerance of a hybrid than the flowering date itself, and is independent of differences between varieties in different maturity groups (Edmeades et al., 1989). A delay in the appearance of the pistils may be caused by their slower growth, which is greatly influenced by the water supplies to the plant (Herrero and Johnson, 1981). Proterandry, which is a useful index of the degree of flowering asynchrony, increases in response to water deficit, primarily due to the later appearance of the ear primordium and the pistils and to their slower growth (Bolaños and Edmeades, 1993). Many authors also attributed an increase in proterandry to a lower quantity of assimilates in the plant, which could be caused, among other things, by greater plant density (Buren et al., 1974).

Ribaut et al. (2009) summarised the opportunities available in breeding for drought tolerance as follows: selection for genotypes with a smaller leaf area in the upper region of the plant, and selection for short, thick stalks, smaller tassels, erect leaves and later senescence (stay green). These authors consider smaller root biomass to be a relatively easily influenced trait, and suggest that the aim should be to produce deep-rooted genotypes with fewer lateral roots.

The aim in drought stress studies should be to find genotypes with short proterandry or complete flowering synchrony, as simultaneous selection is possible in these genotypes for drought tolerance and higher yields.

Materials and methods

A panel of 83 hybrids has been evaluated over two growing seasons (2011, 2012) under well watered (WW) and water deficit (WD) conditions in the nursery of the Agricultural Institute at Martonvásár (Hungary).

The WW area was irrigated on 4–5 occasions each year (in summer, in the months of June and July), based on the data provided by the soil sensors. At each occasion, 40–50 mm water was supplied. The WD area was irrigated on a single occasion with 15 mm water.

Male and female flowering were recorded daily. The plants in each plot were considered to be flowering when at least 50% of the plants had started shedding pollen, or when the silks were clearly visible on at least half the plants. Proterandry was expressed as the number of days between male and female flowering, while the flowering dates were given as the number of days from sowing.

The plots were harvested with a small-plot combine and the yield data (yield/plot, g, and grain moisture, %) were recorded simultaneously with a single instrument. Ear length and percentage fertilisation were recorded for sample ears (the yield of which was added to the plot yield during data processing), and the number of kernels per ear was counted. The thousand-kernel weight was calculated from the kernel number per ear and the kernel weight per ear. The statistical evaluation was carried out using *Agronomix Inc. Agrobase* software.

Results and discussion

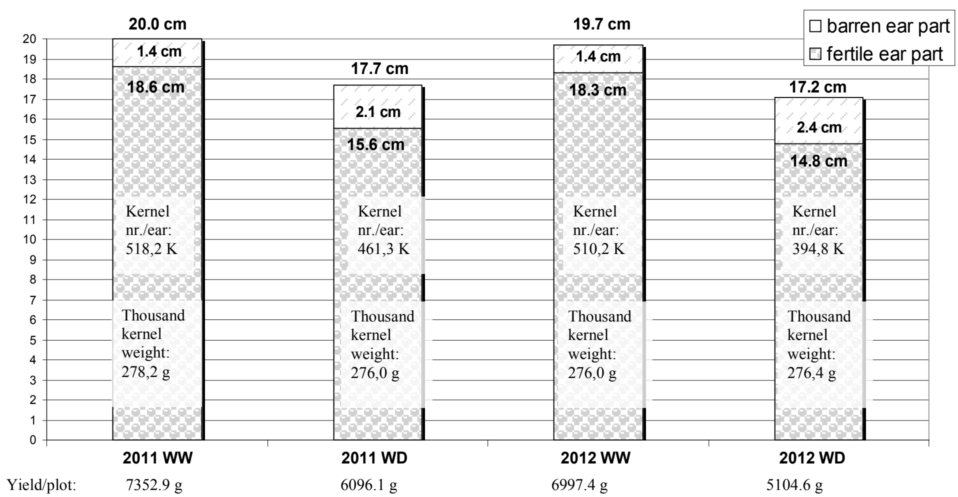
Changes in flowering date and proterandry

The hybrids examined were produced by crossing 83 different female inbred lines with a European flint tester (EFT) as pollinator. The parental lines were developed by various research institutes and companies and belong to diverse maturity groups. The number of days from sowing to male flowering was 58.25 days for the earliest flowering hybrid (EP51*EFT) and 63.25 days for the latest (B73*EFT). This 5-day difference in flowering date is considerable, but the aim of the investigations was to determine the degree of proterandry rather than to determine maturity groups. Female flowering began at the earliest on the 59th day (CO109*EFT) and at the latest on the 65th day (B73*EFT). Both male and female flowering generally began later in WD compared to WW. A comparison of the years revealed that in 2011 flowering took place later than in 2012 and the differences were considerably smaller, in some cases being insignificant.

The analysis of flowering synchrony revealed that both the year and rainfall deficiency have a substantial effect on the appearance of male and female flowers and on the interval between male and female flowering. In the more favourable year, under irrigated conditions, the length of proterandry averaged 1.23 days (ranging from 0.25 to 3 days), while in response to drought stress, in years with rainfall deficit, it averaged 1.96 days (ranging from 0.25 to as much as 6 days). Longer proterandry was induced to a lesser extent by a delay in male flowering than by the protraction of female flowering.

Analysis of yield components and grain moisture

The traits examined at harvest were the following: grain yield, kernel moisture content at harvest, thousand-kernel weight, kernel number per ear, ear length and fertilised ear length. Prolificacy was not investigated, as the genotypes tested did not produce secondary ears in any of the treatments when grown at normal plant density (65–70,000 plants/ha). Complete barrenness did not occur even in the case of drought stress. The yields per plot were adjusted to 15% grain moisture content. As expected, the grain



LSD5%: 146.6 g for yield; 6.2 g for TKW; 12.2 K for kernel number; 0.3 cm for ear and fertile ear lenght

Figure. 1. Average length of the ears and the fertile ears in 2011 and 2012

yields were significantly different for each treatment and genotype. The 2011 season was dry, but otherwise favourable, so the grain yield averaged 7352.93 g/plot on the irrigated area and 6096.14 g/plot without irrigation ($LSD_{5\%}$: 146.58 g). The weather was drier in 2012, leading to average yields of 6997.36 g on the irrigated plots and 5104.6 g under non-irrigated conditions (Figure 1).

As is clear from the data, even for the best hybrids there were significant differences in yield between the dry areas and those with optimum water supplies the WW and WD treatments. The hybrids of three lines (HMv5325, F912 and F98902) did not exhibit significant yield losses in response to drought stress, and the yield of the hybrid developed using line HMv5325 was not substantially lower than that of the highest yielding hybrid (the progeny of line B97). Consequently, the combination HMv5325*EFT could be regarded as the best hybrid, having a high yield under optimum conditions and the least decrease in yield in response to drought stress. Averaged over the two years, the highest yield was recorded for the hybrid of B97 on irrigated plots (9285.78 g).

The general mean for grain moisture content at harvest was 21.24%. Genotypic values related with the maturity groups of the parental material. The hybrids with the lowest grain moisture at harvest were combinations involving CO109 and Oh33 (17.45 and 18.33%, respectively), while those with the highest values were the hybrids of B104 and NC209 (25.66 and 25.13%, respectively). The year had a significant effect on the grain moisture at harvest. Irrigation caused a significant difference in 2011 but not in 2012. The similarity between the irrigated and non-irrigated areas could be attributed to the fact that irrigation was discontinued in both WW and WD and caused the kernels to dry at the same pace in both conditions.

Due to the similar watering conditions in WW and WD during grain filling, there was no difference in thousand-kernel weight between the treatments, nor were significant differences found over the average of the two years. Surplus yields could thus be attributed to the larger number of grain primordia developing on ears with better fertilisation, while the kernel weight remained the same.

In response to irrigation in June and July, larger ears developed on the control plots, with more kernel primordia and better fertilisation. This resulted in a higher kernel number per ear in both years compared to the non-irrigated plots. Of the two years, 2011 had better water supplies, resulting in better fertilisation than in 2012. The majority of the hybrids responded to drought stress with a reduction in the kernel number. The hybrids that produced almost the same number of kernels per ear under dry conditions as in the control environment had the following female parent components: Pa405, LH145, AS5707, FR19, F748, N25, Oh02. The hybrids of these lines had a favourable response to drought and the number of kernels per ear did not change significantly as a result of stress. As the thousand-kernel weight was almost unchanged throughout the experiment (276–278 g), the smallest yield fluctuation due to drought stress was observed for the progeny of these lines.

Acknowledgement

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Determination of the fusarium species composition of maize (Zea mays L.) kernel and stalk samples in Hungary

Csaba Szőke[1], Péter Bónis[2], Tamás Árendás[2], Árpád Szécsi[3], L.Csaba Marton[1]
[1]Maize Breeding Department, [2]Crop Production Department, Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
2462 Martonvásár, Brunszvik u. 2.
[3]Plant Pathology Department, Plant Protection Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
1022 Budapest, Herman Ottó út 15.
e-mail: szokecs@mail.mgki.hu

Abstract

As in other parts of the world, the frequency of weather extremes has increased greatly in Hungary in recent years, together with considerable differences within a single year between locations important for maize production. This means that maize production is faced with greater risks from all aspects: nutrient replacement, irrigation, plant protection. This is especially true of fusarium diseases, partly because maize is attacked primarily by xerotolerant, mesophilic species in dry years and by ombrophilic, psychrotrophic species in wet years, and partly because fusarium stalk rot is the main problem in dry years, and fusarium ear rot in wet years. Depending on which *Fusarium* species are dominant at a given location, changes can be expected in the level of infection and in the quality deterioration caused by the mycotoxins they produce. The degree of infection is fundamentally determined by the resistance traits of the maize hybrids, which can only be effectively investigated based on knowledge of the species composition of the *Fusarium* species attacking maize in Hungary. The aim of the present work was to determine whether the *Fusarium* species composition and ratio had changed with changes in the weather.

Introduction

Fusarium species are able to infect any part of the maize plant in any stage of development from germination to maturity. Infection reduces seed germination ability and the quantity of yield, while the mycotoxins they produce cause severe qualitative damage to the grain, resulting in serious risks to human and animal health. As the correlation between maize resistance and various *Fusarium* species is not clear in all cases, it is very important to know the composition of the *Fusarium* species attacking the crop at a given location. In recent decades the *Fusarium* species causing damage to maize in Hungary have been summarised by a number of authors (*Manninger*, 1967; *Békési and Hinfner*, 1970; *Mesterházy and Vojtovics*, 1977; *Fischl and Halász*, 1990; *Szécsi*, 1994; *Kizmus et al.*, 2000). The most important pathogens are considered to be *F. gra-*

minearum, *F. verticillioides* and *F. culmorum*, all of which produce mycotoxins. The major toxins of *F. graminearum* and *F. culmorum* are deoxynivalenol (DON), zearalenone (ZEN) and nivalenol (NIV), while *F. verticillioides* produces various types of fumonisin (FB1, FB2, etc.) (*Logrieco et al.*, 2002; *Bartók et al.*, 2010). Toxins are known to cause severe physiological damage (*Marasas*, 1995; *Berek et al.*, 2001; *Krska*, 2007; *Pestka*, 2010). Nevertheless, mycotoxins may play an important role in breeding for disease resistance. Several papers have reported that the toxin content is closely correlated with the level of infection, indicating that resistance to *Fusarium* species is regulated by the toxin production of the pathogen (*Perkowski et al.*, 1997; *Toldi et al.*, 2008). According to *Löffler et al.* (2011) the heritability of mycotoxin content, in the case of both *F. graminearum* and *F. verticillioides*, is similar or greater to that of ear infection. *Bolduan et al.* (2009) also found a very close correlation between the toxin contamination of *F. graminearum* and the magnitude of ear infection. These authors consider that visual evaluation is a satisfactory method in breeding for *F. graminearum* resistance. In contrast with the situation for wheat (*Van Eeuwijk et al.*, 1995), the nature of resistance to *Fusarium* species has not yet been clarified for maize, though the available data suggest that it may be similar in the two crops. Various *Fusarium* species are able to attack maize successfully under very diverse ecological conditions, and several *Fusarium* species may be present at any location. For this reason, and due to the climate change experienced in recent years, there is every justification for continuing the nation-wide determination of *Fusarium* species begun several decades ago. The results of this survey may provide an answer to whether the species composition and ratio have changed and whether new *Fusarium* species capable of attacking maize have appeared in Hungary.

Materials and methods

Stalk samples were collected in Martonvásár between 2006 and 2008, and stalk and ear samples in eight locations of importance for maize production (Martonvásár, Sárhatvan, Keszthely, Bicsérd, Kaposfüred, Maroslele, Kaba, Debrecen) between Sept. 21 and Oct. 15, 2013. The total number of samples in the years 2006–2008 was 165, while in 2013 15 ear and 15 stalk samples were collected at each location. The samples were labelled and stored frozen until processing. The stalk samples were surface-sterilised by shaking for 15 min in 1% NaOCl solution, then rinsed three times and dried to air dryness. A similar protocol will be used to process the kernel samples. The disinfected samples were placed on selective *Fusarium* media (*Szécsi*, 2004), and the isolated *Fusarium* species were determined after incubation (*Leslie and Summerell*, 2006). The statistical evaluation was performed using the modules incorporated in the MS® Excel program and the Agronomix Inc. Agrobase program.

Results and discussion

The distribution of the *Fusarium* species isolated from stalk samples in 2006–2008 is presented in *Table 1*. Averaged over the three years, the *F. verticillioides* species was found with the greatest frequency (56.36%) in Martonvásár stalk samples, followed by *F. graminearum* (23.64%) and *F. subglutinans* (12.73%). The fungus species *F. culmorum* and

Fusarium species	2006		2007		2008		Total	
	No.	%	No.	%	No.	%	No.	%
F. verticillioides	33	64.71	36	56.25	24	48.00	*93*	*56.36*
F. graminearum	12	23.53	15	23.44	12	24.00	*39*	*23.63*
F. subglutinans	6	11.76	9	14.06	6	12.00	*21*	*12.73*
F. culmorum	0	0.00	1	1.56	6	12.00	*7*	*4.24*
F. proliferatum	0	0.00	3	4.69	2	4.00	*5*	*3.03*
Total	*51*	*100*	*64*	*100*	*50*	*100*	*165*	*100*

Table 1. Distribution of Fusarium species isolated from stalk samples (Martonvásár, 2006–2008)

F. proliferatum were not detected in 2006. *F. proliferatum* was present on 4.69% of the samples in 2007, while *F. culmorum* was the more frequent of the two species (12.00%) in 2008. With the exception of *F. graminearum*, the species composition exhibited greater or lesser changes each year, while the occurrence of *F. graminearum* was around 23.5% in all three years. When the data were compared with those reported by *Békési and Hinfner* (1970) and *Mesterházy and Vojtovics* (1977) in the national survey of *Fusarium* infection on maize stalks, the two pathogens considered to be most important for stalk fusarium (*F. verticillioides* and *F. graminearum*) were found to be present in the greatest numbers in all three data series. The incidence of *F. verticillioides* (a thermophilic species) increased, while that of *F. graminearum* remained relatively constant (*Fig. 1*).

The incidence of the *F. subglutinans* species increased over the years, and this species was isolated from maize stalks at the highest frequency between 2006 and 2008. This increase can be attributed to global warming. The presence of *F. culmorum* was the most unpredictable. This may have been due to the fact that only the asexual spores of this species are capable of inducing infection, which puts them at a disadvantage compared with species having a sexual cycle (*F. graminearum*, *F. verticillioides*), where the ascospores developing in the fruiting body during the sexual phase are also able to infect the plants.

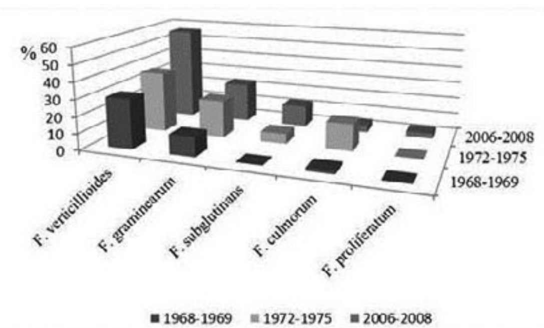

Fig. 1. Distribution of Fusarium species isolated from maize stalk samples

A national survey is planned for 2013–2014. Stalk and ear samples have already been collected this year, but have not yet been processed.

In the majority of cases the mould surrounding the nodes was white (sporodochium ring), suggesting that *F. verticillioides* will again be found in the stalk samples this year.

Acknowledgement

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Changes in the dry matter yield and chemical composition of silage maize hybrids during ripening

Zsuzsanna Tóth Zsubori, János Pintér, István Pók, L.Csaba Marton
Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences
2462 Martonvásár, Brunszvik u. 2.
e-mail: zsubori.zsuzsanna@agrar.mta.hu

Abstract

The choice of harvest date for silage maize has a substantial influence on the quantity and quality of the silage. As ripening proceeds, increasing quantities of nutrients accumulate in the ear, while the digestibility of the other plant organs gradually deteriorates. The accumulation of dry matter and changes in the chemical components, primarily the lignin and digestible organic matter content in the ear and the other plant organs, were examined together with changes in the ear ratio in plant samples taken every 10 days from flowering onwards. The proportion of the ear compared with the whole plant dry matter gradually increased to 52–60% at harvest, with a simultaneous increase in the dry matter content and digestible organic matter content. By contrast, the digestible organic matter content of the leafy stalks rose slightly at first but then exhibited a declining trend. The lignin content in the ear gradually decreased, while in the other plant organs, after a slight initial reduction, it rose considerably by the time of harvest (from 4.92 to 5.71%). The digestible dry matter yield reached its maximum at a dry matter content of 35–40%, after which it started to decline.

Introduction

The most important traits for silage maize are the dry matter yield, the ear ratio and digestibility, all of which are greatly influenced by the harvest date. According to *Weissbach and Auerbach* (1999) maize reaches silage maturity when grain filling is completed but the plant is still green, with a dry matter content of 30–35%. However, harvest is still possible without greater loss up to a dry matter content of 42%, thus ensuring maximum yield and milk production (*Darby and Lauer*, 2002). According to *Józsa* (1981), if silage maize is harvested at a dry matter content of 35–40% the proportion of the ear to the whole plant dry matter may be as much as 55–65%, which can be considered to be a very good ratio. The fodder value of maize harvested at this time is also better than when harvested earlier, while this dry matter content is required for satisfactory silage fermentation. Maize harvested at physiological maturity results in silage with a higher lactic acid content (*Irlbeck et al.*, 1993).

Digestibility depends greatly on the maturity and dry matter content of the plant, so the correct choice of harvest date is of exceptional importance. The optimum dry matter content of silage maize at harvest is around 35%, though some authors measured maximum digestibility at lower (*Argillier and Barrière*, 1996; *Kim et al.*, 1999) or higher dry matter contents (*Giardini et al.*, 1976; *Hunt et al.*, 1989). As ripening proceeds, the degradability of the stalk dry matter declines, as increasing quantities of lignin accumulate in the plant tissues, particularly in the stalks, causing a deterioration in digestibility (*Bal et al.*, 2000; *Russell*, 1986). The digestibility of the whole plant, however, improves thanks to an increase in the ear ratio (*Ettle and Schwartz*, 2003; *Gul et al.*, 2008). All in all, optimum dry matter yield, quality and milk yield can be expected at a dry matter content of between 30 and 40% (*Darby and Lauer*, 2002).

Ma et al. (2006) investigated dry matter incorporation and changes in the dry matter content at several dates after flowering and recorded the greatest dry matter accumulation at the optimum harvest date (at 35% dry matter content), between the milky and waxy ripe stages (1/3-2/3 milkline). In more northerly regions this date was 50 days and in more southerly regions 35 days after flowering. According to *Giardini et al.* (1976) the greatest dry matter yield and best chemical quality content can be expected 40–45 days after flowering (38–42% dry matter). Data in the literature suggest that the carbohydrate content of the stalk rises for a time after tasselling, then decreases from the 35th day onwards (*Dwyer et al.*, 1995). The quantity of *in vitro* digestible, insoluble carbohydrates was also found to stabilise by the 35th day after flowering (*Argillier and Barrière*, 1996).

Based on the above, the aim was thus to examine changes in the dry matter yield and chemical composition during ripening in a number of silage maize hybrids bred in Martonvásár, in order to determine the optimum harvest date.

Materials and methods

The silage maize hybrids used in the experiment were sown in Martonvásár in 4-row plots in a randomised complete block design with three replications under irrigated conditions. The genotypes examined were Mv Nutrisil, Mv Siloking, Mv Kámasil and Mv Megasil. The first samples were taken directly after flowering, on 15 July, and then at 10-day intervals until harvest (on 25 July, 5, 15 and 26 August). Three plants were cut from each replication, the ears and husks were removed and the remaining plant organs were chopped. The ears were chopped separately. The chemical composition of the chopped samples was determined using a Bruker MPA type of NIR spectrometer and the data were evaluated using INGOT calibration software. Separate plants were taken from each replication for the determination of dry matter content and ear ratio. The ears were removed, and after drying to constant weight in a drying cabinet at 105°C, the ears and the other plant organs were weighed separately. The data were then evaluated using two-way analysis of variance. The following equation was applied to calculate the digestible dry matter yield:

- IVDMY = DMT*IVDOM/100
  where IVDMY = digestible dry matter yield (t/ha), DMY = dry matter yield (t/ha) and IVDOM = *in vitro* digestible organic matter content (%). To calculate the dry matter yield per hectare, the dry matter yield per plant was multiplied by the 80,000 plants/ha plant density used in the experiment.

Results and discussion

The hybrids tested in the experiment silked between 10 and 15 July. The first sample was taken immediately after flowering. Based on data from the literature (*Ma et al.*, 2006; *Giardini et al.*, 1976) and the experience gained in earlier years, the last sampling (harvest) was timed for the 40th day after flowering. The severe drought and extremely high temperatures in summer resulted in forced ripening and in early loss of lower leaves and drying down, so the dry matter content at harvest was somewhat higher than expected. Averaged over the hybrids the dry matter content of the whole plant was 40.63%. A similarly strong year effect was noted in previous experiments (*Tóthné Zsubori et al.*, 2009).

The dry matter content in the ear increased more dynamically than that of the other plant organs (*Table 1*). The dry matter content of the leafy stalk was 17.05% at flowering, rising to 30.68% at harvest, while that of the ear was 10.94% at flowering and 50.58% at harvest. The crude weight of the ear exhibited an initial increase as the kernels developed, but dropped slightly by harvest due to drying down and the water loss of the kernels. The mass of the other plant organs only decreased slightly at first, but dropped to a greater extent between the last two sampling dates. The hybrid Nutrisil had the greatest fresh mass at harvest (1004 g/plant).

Among the chemical parameters, the digestible organic matter content gradually declined in the stalk as ripening progressed, while it increased in the ear, in agreement with data in the literature (*Bal et al.*, 2000; *Gul et al.*, 2008). When silage maturity was reached, the digestible organic matter content of the ear was almost 30% greater than that of all the other plant organs (75.18% and 57.23%). Between the last two sampling dates there was no further increase in this value; in fact, the curve fitted to the data indicated a slight reduction by harvest. The opposite tendency was noted for the lignin content, the other parameter having a major influence on digestibility, with a slight increase in the stalk and a substantial reduction in the ear in the course of ripening. At harvest the ear had a lignin content of 2.44%, while that of the stalk was 5.71%, averaged over the hybrids. Differences between the genotypes at the same sampling dates were not significant for either of the traits. Many authors consider that the digestibility of the plant organs depends less on the genotype than on the stage of maturity (*Masoero et al.*, 2006; *Ettle and Schwartz*, 2003).

Sampling date	DM* (%)		IVDOM** (%)		Lignin (%)	
	Leafy stalks	Ear	Leafy stalks	Ear	Leafy stalks	Ear
15. July	17.05	10.94	66.11	69.33	5.10	4.31
25. July	19.83	11.68	66.29	71.88	5.08	3.59
5. August	19.95	25.68	64.92	77.40	4.92	2.80
15. August	25.54	39.26	61.25	74.31	5.01	2.66
26. August	30.68	50.58	57.23	75.18	5.71	2.44
$LSD_{5\%}$	1.45	1.76	0.95	1.54	0.17	0.28

** DM= Dry matter; ** IVDOM= In vitro digestible organic matter*

Table 1. Changes in major chemical parameters during ripening (averaged over four hybrids, Martonvásár, 2013)

The ear ratio and the digestible dry matter yield are important traits for the value of silage maize hybrids (*Table 2*). The hybrid Nutrisil had the smallest ear ratio of all the hybrids tested (52.07%), but the highest digestible dry matter yield (19.32 t/ha). This could probably be attributed to the greater plant weight. At harvest the crude weight of Nutrisil was significantly greater than that of the other hybrids, with a slightly lower dry matter content in the ear and almost the same in the stalk. The dry matter yield per plant was 9.28% higher than the average for the other hybrids. The reason for the lower ear ratio was that Nutrisil produced a single, well-developed ear, while the other hybrids often produced secondary ears. However, there was no significant difference between the genotypes in the digestible organic matter of the ears and leafy stalks. Other chemical traits not detailed in the present work also have an influence on digestibility, such as the protein, starch and water-soluble carbohydrate contents. In the case of Nutrisil the ear protein content was significantly higher than that of the other hybrids.

Hybrids	Ear ratio (%)					DDMY (t/ha)				
	15 Jul	25 Jul	5 Aug	15 Aug	26 Aug	15 Jul	25 Jul	5 Aug	15 Aug	26 Aug
Nutrisil	16.16	24.15	45.06	49.75	52.07	8.03	8.51	14.63	17.71	19.32
Siloking	17.49	20.15	43.48	47.53	58.30	6.74	8.59	15.54	15.95	18.51
Kámasil	14.39	28.50	43.73	47.13	58.57	6.73	11.86	16.53	17.39	15.76
Megasil	20.96	30.70	40.48	53.02	60.43	7.91	10.26	16.09	20.37	18.09

Table 2. Changes in the digestible dry matter yield (DDMY) and ear ratio of the hybrids at various sampling dates (Martonvásár, 2013)

The ratio of the ear to the whole plant dry matter gradually increased in all the hybrids, with a steep rise between the second and third sampling dates (10 and 20 days after silking). This sudden increase in the kernel mass could be attributed to the fact that cell division in the endosperm had been completed, leading to the development of the final cell number and the start of the linear phase of grain filling, when assimilates are incorporated into the kernels in the form of starch. The intensity of this process is genetically determined and is not influenced even by high temperature (*Badu-Apraku et al.*, 1983). The digestible dry matter yield rose more steeply at first, and then to a lesser extent. In fact, a decrease was observed for Kámasil and Megasil by the harvest date. Averaged over the hybrids the changes over the whole of the period investigated could be described using the equation: $y = -0.6071x^2 + 6.5609x + 0.72$.

The results demonstrate that the quality of silage maize deteriorates at a dry matter content of over 40%. The chemical quality parameters exhibit unfavourable changes, with a rise in the lignin content and a reduction in the digestible organic matter content. This is associated with a drop in the digestible dry matter yield per hectare. An excessively high dry matter content is also unfavourable from the point of view of storage (compressibility and fermentation). Silage maize should thus be harvested at a dry matter content of 35–40%, which is reached 35–40 days after flowering in dry or droughty years for the hybrids examined in this work.

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