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Insights into the Phenotypic and Genotypic Diversity of Fresh-Eating Maize Germplasm

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Abstract This study comprehensively explored the phenotypic and genotypic diversity of fresh-table maize germplasm, highlighting the importance of this diversity for breeding programs. Phenotypic assessments, such as principal component analysis (PCA) and GT biplot analysis, revealed significant variation in key traits such as grain weight, plant height, and nitrogen use efficiency, which are essential for improving yield in different environments. Meanwhile, molecular characterization using SNP markers further elucidated the genetic diversity in maize breeding germplasm and found clear distinctions between temperate and tropical/subtropical lines, as well as unique alleles that can be used for breeding. This study also introduces the latest advances in combining genotypic and phenotypic data to help identify quantitative trait loci (QTLs) to improve breeding efficiency. Despite some progress, there is still underutilized genetic diversity in current breeding programs. This study emphasizes that continued research in phenotypic and genotypic characterization, combined with advanced breeding technologies, is essential for sustainable improvement of the global maize crop.

Keywords Fresh-eating maize; Phenotypic diversity; Genotypic diversity; Hybrid breeding; Genomic selection

1 Introduction

When it comes to corn, the first thing that comes to mind is the common feed corn, but in fact, fresh corn (also known as sweet corn) is a regular on the daily table. This kind of corn can be seen all over the world. Whether it is cooked directly or made into canned or frozen food, it is very popular. But what's interesting is that although it is a staple food in many places, the way it is eaten in different regions varies greatly. Farmers especially like to grow this because they can make money - whether it is in the local market or exported, it sells well (Hansey et al., 2012; Jin et al., 2012; Hufford et al., 2021). Of course, in addition to making money, it is also very important to ensure food security. Don't think that there is everything in the supermarket now. If you really encounter some special circumstances, crops like fresh corn that can be eaten fresh and processed and preserved will play a prominent role.

Corn varieties vary widely, from taller to larger ears to different kernel compositions-and these visible differences, or phenotypic diversity, are particularly useful to breeders. After all, these traits are important for improving yields, enhancing disease resistance, or adapting corn to different climates (Yang et al., 2011; Mhoswa et al., 2016). But the real fun is at the genetic level. The DNA of different corn varieties can vary even more, from gene content to methylation patterns (Hufford et al., 2021; Shu et al., 2021). Although some varieties look similar, they can have completely different genotypes. This kind of diversity is no small matter, especially as climate change becomes increasingly apparent. Researchers have found that it is these genetic differences that can help us breed varieties that are more tolerant to drought and disease (Flint-Garcia et al., 2009; Jin et al., 2012). To put it bluntly, the future adaptability of corn may lie in its genetic diversity.

When it comes to corn for fresh consumption, you may not realize how different varieties of corn can be. Some corn cobs are extra sweet, while others are more durable in storage. Behind these visible differences, there are actually complex genetic secrets. Our study aims to find out two things: which genes control these important traits, and how environmental changes will affect corn performance. Interestingly, although genomic technology is very advanced now, the potential of many traditional corn varieties has not yet been fully explored. We will focus on analyzing the genetic variation of different corn populations, especially those key genotypes that may be helpful for breeding (Hufford et al., 2021). Of course, it is not enough to just find good genes. We also have to consider how to stably pass on these excellent traits. Especially in the context of climate change, maintaining corn diversity may become the key to improving yield and stress resistance (Flint-Garcia et al., 2009). In the final analysis, this study is to provide breeders with some new ideas to make those good traits hidden in corn genes really work. After all, to feed a growing population, it may not be enough to rely on just a few high-yield varieties.

2 Fresh-Eating Maize Germplasm

2.1 Types of fresh-eating maize

Speaking of fresh corn, you may not know that it is actually divided into several types. The most common is sweet corn, which tastes particularly sweet and tender, and many people like it. But it is not as simple as eating it fresh - most of the canned corn and frozen corn kernels in the supermarket are actually processed from sweet corn (Ruanjaichon et al., 2022; Dang et al., 2023). There is another type that you may have seen in hot pot restaurants, which is the kind of sticky glutinous corn. It is different from sweet corn, has a high starch content, and is sticky after cooking. Although it is very common in China and Southeast Asia, it is rare in the European and American markets (Ruanjaichon et al., 2022; Dang et al., 2023). Interestingly, although these two types of corn are both fresh corn, their uses and eating methods are quite different.

There are some interesting new varieties of corn on the market now, which retain the sweetness of sweet corn and have the sticky taste of sticky corn. Breeding experts do not crossbreed randomly, they focus on key characteristics such as sweetness and stickiness to improve (Ruanjaichon et al., 2022). However, it is easier said than done. To breed such varieties, you must first figure out which genes control these characteristics. Researchers now use some molecular markers, just like marking genes, so that they can find the desired traits faster (Ruanjaichon et al., 2022). Although the process is quite complicated, the result does make corn more delicious.

2.2 Geographic distribution and diversity of fresh-eating maize germplasm

Corn grown around the world is actually very different, just like people in different places have different tastes. North America is particularly fond of sweet corn, which has been grown for hundreds of years and now has countless varieties. Breeding experts have cultivated various inbred lines and hybrids, some of which have high yields and some are more disease-resistant (Hansey et al., 2012; Romay et al., 2013). Turning to Asia, the situation is different. Chinese and Southeast Asians prefer sticky corn, which is particularly popular there. Interestingly, although the planting area is not that wide, the varieties in each place are very distinctive, and they are slowly improved according to the local climate and eating habits (Ruanjaichon et al., 2022; Dang et al., 2023). You see, the same corn has developed completely different appearances in different places.

Corn in Africa is very interesting, especially in the Sahel and coastal areas of West Africa, where you can see all kinds of local varieties in all shapes and sizes. These native corns are not simple, they have a very rich genetic diversity, which is a treasure trove for breeding improved varieties that are resistant to drought and disease. Speaking of diversity, corn in the northeastern Himalayas of India is no less diverse. There are so many local varieties there that it is dizzying that the corn in each field may be slightly different. Breeding experts attach great importance to these resources, not only because they can be used to improve varieties, but more importantly, to protect these precious genetic resources. After all, who knows if the climate changes one day and the special genes of these local varieties will come in handy?

2.3 Conservation and utilization status of germplasm resources

When it comes to corn for fresh consumption, you may not know how important it is to preserve these varieties. Now countries are building germplasm banks to collect various corn seeds. The United States has done something interesting. They have conducted genetic testing on thousands of corn inbred lines (Romay et al., 2013). These data are particularly useful for breeders. Through analysis, they can find some rare gene variants and figure out which genes control important characteristics (Romay et al., 2013). Although it sounds quite technical, to put it bluntly, it is to preserve the diversity of corn. After all, no one knows when an inconspicuous local variety will help us cope with climate change or new pests and diseases.

When it comes to breeding, both old and new methods are used. For hundreds of years, farmers have known how to select good varieties for hybridization, and slowly cultivated high-yield and disease-resistant corn (Flint-Garcia et al., 2009; Nelimor et al., 2020). But now the situation is different, and scientists have begun to use some high-tech methods. In recent years, genome analysis technologies such as GWAS have been particularly popular (Ruanjaichon et al., 2022; Dang et al., 2023). Although traditional breeding has also achieved good results, the new method is indeed faster and more accurate. It can directly find genetic markers that control excellent traits, allowing breeding work to avoid many detours (Ruanjaichon et al., 2022; Dang et al., 2023). Interestingly, many breeding projects now use both old and new methods to complement each other.

3 Phenotypic Diversity of Maize

3.1 Phenotypic traits of maize and their importance in breeding

When breeders look at corn, they first focus on a few key indicators. For example, grain yield and plant height are commonplace but particularly important traits. But what's interesting is that details such as 100-grain weight and the number of cobs per plant can more accurately predict the yield (Al-Naggar et al., 2020; Naggar et al., 2020). When it comes to yield prediction, you may not think that even root characteristics are critical. Although the underground part is usually invisible, the degree of root development is closely related to the growth of corn (Tucker et al., 2019). Of course, different places may value different traits - arid areas may pay more attention to the root system, while fertile areas may pay more attention to ear characteristics. But in any case, these traits are important bases for evaluating the quality of varieties.

Breeding experts have found that some characteristics of corn are particularly "stubborn" - indicators such as ear height and thousand-grain weight are basically stably inherited by offspring (Ababulgu et al., 2016). This is very interesting, because as long as these traits are selected, the success rate of breeding new varieties can be greatly improved. But then again, just focusing on yield is not enough. Now people pay more attention to the comprehensive evaluation of various phenotypic characteristics, so as to find truly high-quality genotypes (Mhoswa et al., 2016). For example, some corn may not have the highest yield, but it is particularly resistant to drought. Combining these characteristics often breeds hybrid varieties that are both high-yielding and resistant to stress.

3.2 Methods for evaluating major phenotypic traits

To study whether corn grows well, we need to rely on field investigation and data analysis. Farmers and scientists often conduct experiments in drought or fertilizer-deficient plots, mainly looking at intuitive indicators such as corn cob size and plant height. Interestingly, the same variety may perform very differently in different plots. There are many ways to analyze data now, such as principal component analysis (PCA), which can clarify complex trait relationships (Al-Naggar et al., 2020). There is also a GT biplot analysis that is quite practical, which can simultaneously show the interaction between variety characteristics and environmental factors (Naggar et al., 2020). Although it sounds professional, it is actually to have a more comprehensive understanding of the growth characteristics of corn.

When it comes to predicting corn yields, even machine learning is now used. These algorithms are smart and can find details that we might overlook - such as the shape of the corn cob or the development of the root system, which have a significant impact on the final yield (Tucker et al., 2019). However, machine learning is not a panacea. Researchers have found that it is more effective when used in conjunction with path coefficient analysis (Mhoswa et al., 2016). This method can distinguish which traits directly affect yield and which ones work indirectly through other factors. For example, some seemingly important characteristics may just be "hitchhiking". By clarifying these relationships, you can more accurately grasp the key points when breeding.

3.3 Influence of cultivation environments on maize phenotypic diversity

How corn grows actually depends a lot on its "face" - here we are talking about the growing environment. How much water is applied, what fertilizer is applied, and the temperature will make the corn grow differently. Take the 100-grain weight as an example. The same variety of corn may differ a lot in dry land and fertile land (Al-Naggar et al., 2020). Interestingly, some characteristics are particularly "delicate". For example, the number of cobs per plant and the number of grains per row are particularly sensitive to water and fertilizer conditions (Naggar et al., 2020). Therefore, when breeding, you can't just look at the variety, you also have to consider where it will be planted in the future. After all, the same corn planted in different plots may have a very different harvest.

Corn hybridization is quite interesting. The "children" born from the same parents in different places may be very different. For example, some hybrid combinations grow particularly well in the north, with long and thick ears and full grains, but they may perform mediocrely in the south (Yu et al., 2020). This reminds breeding experts that they cannot only conduct research in the laboratory. Now they pay more and more attention to field testing, after all, corn will eventually be planted in the field. Some varieties look good in data, but when they are planted in fields with different climatic conditions, their performance may be completely different (Yu et al., 2020). Therefore, when breeding new varieties, environmental factors must be taken into account, so that good varieties with strong adaptability can be selected.

4 Genotypic Diversity of Maize

4.1 Sources of genetic variation in maize

The genes of corn are like a hodgepodge, with a lot of variations - this has become its specialty in adapting to different environments. Where do these variations come from? Some are natural mutations, some are hybridization and recombination, and some are the result of artificial selection. Take Chinese summer corn for example. Although they are all from the same ecological zone, the genetic differences between different varieties are not small (Shu et al., 2020). Interestingly, these Chinese varieties are also mixed with a lot of North American corn genes. In the past 20 years, North American germplasm has made a significant contribution to the genetic improvement of Chinese summer corn (Shu et al., 2020). However, genetic variation is also very selective. The part of the corn chromosome close to the telomeres has a lot of variation, while the area close to the centromere is relatively conservative (Figure 1). This "selective" variation pattern makes breeding work more interesting.

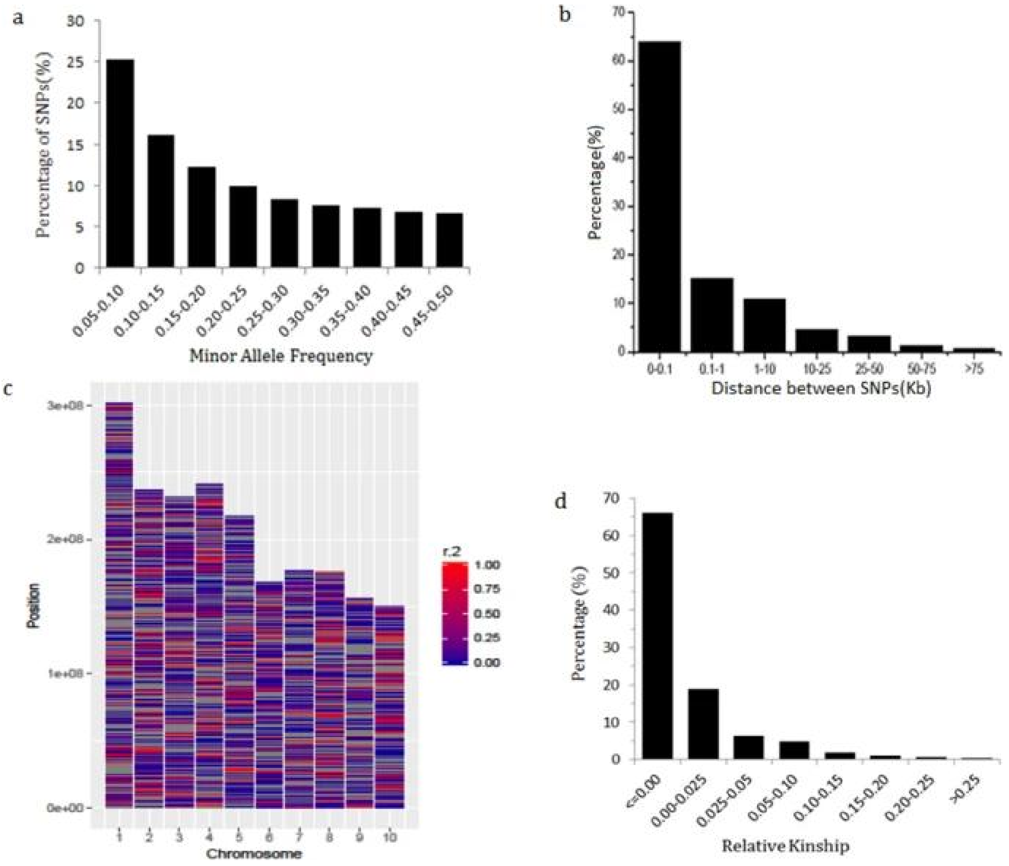

Figure 1 The molecular characteristics of maize genomes (Adopted from Shu et al., 2020)

Image caption: (a) Minor allele frequency distribution; (b) Physical distance between adjacent SNP loci; (c) LD pattern of SNP loci along each of the 10 chromosomes; (d) The Relative kinship of 490 inbreds (Adopted from Shu et al., 2020)

When it comes to genetic variation in corn, there is an interesting phenomenon - some important genetic information is not in the conventional reference genome at all. Scientists have discovered a lot of "hidden" new transcripts using RNA sequencing technology, which has added new information to the genetic diversity of corn (Hansey et al., 2012). Take one of the studies as an example, they found 1321 reliable new transcripts. Some are common in all corn varieties, but some are very "selective" and only appear in specific lines (Hansey et al., 2012). This kind of transcript variation (what experts call ePAV) is actually very important in explaining the hybrid advantage of corn (Jin et al., 2015). Simply put, it may be these genetic fragments that appear from time to time that make hybrid corn grow better than its parents.

4.2 Common tools and techniques for studying genotypic diversity in maize

There are many methods to study the genetic differences of corn now, and scientists have several "brushes" in their hands. Old technologies such as SSR markers are still very easy to use - they can clearly tell us how big the genetic differences are between different inbred lines (Patto et al., 2004). It's interesting to say that although there are new methods such as SNP and RAD-seq, SSR markers are still particularly useful in distinguishing corn varieties (Patto et al., 2004). After all, to figure out "who is who" among these inbred lines and their genetic relationships, the answers given by SSR are both intuitive and reliable. However, each technology has its strengths, and researchers usually use them in combination according to specific needs.

When it comes to studying genetic differences in corn, SNP markers are now very popular. This type of marker is very precise and can find those tiny variation points in the genome. A study used RNA-seq technology to discover more than 350 000 polymorphic sites (Hansey et al., 2012). Relying on these sites, scientists successfully classified different corn inbred lines. But SNP is more useful than that. Genotyping methods such as GBS also like to use it, after all, it can clearly show the genetic differences between corn varieties (Shu et al., 2020). Interestingly, through SNP analysis, it was found that those seemingly similar inbred lines can actually be divided into several subgroups. This shows that the genetic diversity of corn may be much richer than we imagined.

There is another trick called RAD-seq for studying corn genes, which is quite interesting. It is like using scissors to cut a specific position of DNA, and then studying the cut gene fragment (Hufford et al., 2021). Although it sounds a bit violent, the effect is unexpectedly good. Using this method, we can not only find the location of genes that control important characteristics, but also draw high-quality gene maps for different corn varieties (Hufford et al., 2021). In fact, whether it is RAD-seq or other technologies, the ultimate goal is to understand the genetic code of corn. With these tools, breeders will have more direction in improving varieties, and they don't have to rely on luck as before.

4.3 Application of genomic data in genetic research on maize

Corn genomic data is now a treasure, and breeders are scrambling to use it. What are its uses? The most practical one is that it can help us find those gene loci (QTLs) that control important traits. But interestingly, some genes have very small effects, which cannot be found by previous technologies. Now with the tool of association population, the situation is different (Flint-Garcia et al., 2005). It can cover most of the genetic variation in corn varieties, and even those QTLs with subtle effects can be found. Although the effect of a single small effect gene is limited, the sum of them is very significant. This is particularly helpful for analyzing complex traits, and it also allows breeding work to better utilize the genetic diversity of corn.

Speaking of corn breeding, scientists recently discovered an interesting phenomenon. By comparing the genomes of temperate and tropical corn, they found more than 1 100 artificially selected regions (He et al., 2017). These places are not simple. Some control key genes, while others affect gene expression. Interestingly, these selected regions are often related to important characteristics. For example, sucrose transport and oil storage, which directly affect the quality of corn (He et al., 2017). Although traditional breeding is also effective, this method starting from the genetic level makes the improvement work more targeted. After all, knowing where the "switch" is, it is much easier to adjust.

Maize genome research has recently developed a new method-onstructing a pan-genome and a pan-transcriptome. This is not simply putting together several genomes, but it can discover many new sequences that cannot be found by traditional methods (Jin et al., 2015). Interestingly, some genes are regulated in a very special way and may be affected by distant eQTLs. By analyzing RNA-seq data from different inbred lines, researchers have found a large number of such new genes (Jin et al., 2015). Although their specific functions have yet to be studied, they are likely to affect various traits of maize. These findings provide new ideas for breeding work and may help us breed better maize varieties. After all, the more you know about a gene, the more possibilities for improvement.

5 Phenotypic and Genotypic Association Studies in Maize

5.1 Overview of phenotype-genotype association analysis

To figure out why corn grows like this or that, scientists now love to play "matching" - matching the traits they see with the genes. It sounds simple, but there are many factors to consider in actual operation: the influence of the genes themselves, the role of the environment, and the complex interaction between them (Wallace et al., 2014). There is a particularly interesting study. They found more than 5 000 corn inbred lines and observed 41 different traits. Guess what? Nearly 4 800 gene variants related to these traits were found (Wallace et al., 2014). This number is really amazing, indicating that the genetic background of corn is much more complicated than we thought. However, this also brings new challenges to breeding work - after all, finding truly useful genes is like looking for a needle in a haystack.

Breeding experts now attach great importance to genotype data, and there is a reason for this. Take one study, for example, they genotyped 2 815 maize inbred lines and found more than 680 000 SNP markers (Romay et al., 2013). This number may sound a bit abstract, but it is actually very useful. These markers can help us discover those rare genetic variants and clarify the relationship between different populations (Romay et al., 2013). Although it sounds very technical, to put it bluntly, it is to conduct GWAS analysis more accurately. After all, to improve maize varieties, you must first understand its genetic background. With this data, breeding work can avoid many detours.

5.2 Application of genome-wide association studies (GWAS) in fresh-eating maize

GWAS has become a standard tool for corn genetic research now, thanks to the rapid development of sequencing technology. Corn is particularly suitable for this analysis method because it has many genetic variations and linkage disequilibrium decays quickly (Xiao et al., 2017). In fact, there have been quite a few achievements in this area in the past decade. Researchers have found that as long as the statistical model is used correctly, GWAS can match genetic variation with actual traits (Shikha et al., 2021). For example, the results shown in Figure 2 not only found the gene regions that control common traits, but also found many sites related to complex traits such as drought resistance and disease resistance. Although each discovery may be just a small piece of the puzzle, when accumulated, you can see the whole picture.

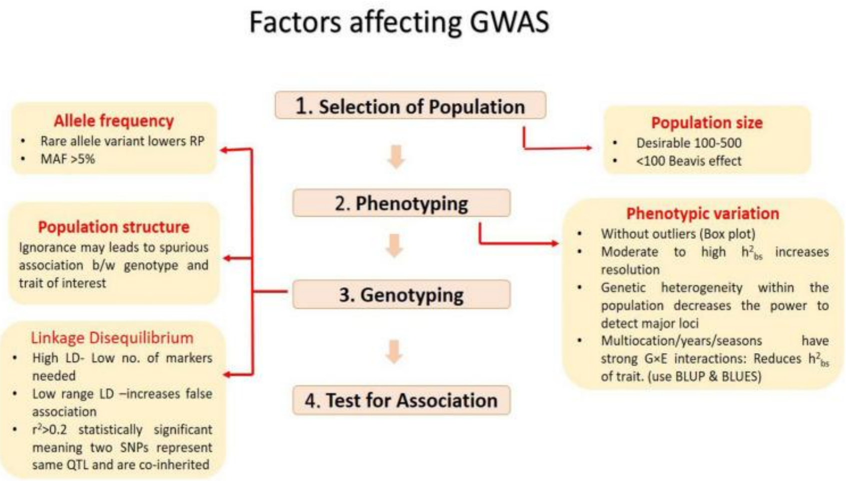

Figure 2 Factors affecting GWAS accuracy and resolution at successive stages (Adopted from Shikha et al., 2021)

Recently, GWAS has come up with a new trick called XP-GWAS. This method is quite interesting. Instead of testing a large number of samples, it only selects those individuals with extreme performance for study (Yang et al., 2015). Take the number of rows of corn kernels as an example. This method can still find related gene variants and analyze complex QTLs. Although it sounds like lazy, it is actually particularly suitable for species with limited genetic testing resources (Yang et al., 2015). Think about it, traditional GWAS often tests hundreds or thousands of samples, while XP-GWAS only needs to focus on the extreme parts at both ends. It saves money and the effect is not discounted. It is particularly useful for studying quantitative trait genes.

5.3 Analysis of key traits and their associated control genes

When it comes to corn breeding, what everyone is most concerned about is yield. But what's interesting is that although yield itself is important, traits that seem not directly related, such as plant height and ear length, can explain a lot of problems (Rahman et al., 2018). We did an experiment and selected 15 different genotypes of corn. The results showed that the differences between these traits were particularly obvious. Of course, not all traits are so useful, but the two indicators of thousand-grain weight and number of grains per ear are indeed very stable, with surprisingly high heritability, and breeders should pay special attention to them. Speaking of which, through path analysis, we found that some traits have a direct impact on yield, which is quite unexpected. But then again, these findings are nothing new, and similar studies have been done in previous years.

Speaking of genomic analysis, there is now an interesting method called whole-genome mediation analysis. Simply put, it is to connect genotype and phenotype through the "middleman" of transcriptome data (Yang et al., 2022). Of course, this method has a premise assumption-gene expression plays a mediating role in this process. 736 such mediator genes have been found in corn research, but the most surprising thing is that some genes can actually affect multiple traits at the same time. Although this method still needs more verification, it has to be said that integrating different omics data can indeed help us better understand complex traits. You see, this provides a clear direction for subsequent functional verification.

6 Gene-Environment Interaction Effects on Maize Traits

6.1 Definition of gene-environment interaction and its effect on maize traits

Anyone who is engaged in seed breeding knows that the same variety may perform very differently in different places. This is the so-called gene-environment interaction (GEI), which means that different genotypes respond differently to environmental changes (Boer et al., 2007; Li et al., 2022; Singamsetti et al., 2022). For example, in the case of corn, some varieties have a sharp drop in yield under drought conditions, but another variety may be quite resistant. However, the most troublesome thing is that this interaction not only affects yield, but also changes indicators such as grain moisture content. Once environmental factors such as temperature and soil moisture change, the performance of the variety will fluctuate, which brings a lot of headaches to breeding work. Of course, it is not completely unsolvable, but it does take more effort to breed varieties with wide adaptability.

When it comes to QTL research in corn, there is a very interesting phenomenon. You may find that the same yield-related QTL performs very well under drought conditions, but it may be completely different in another environment (Boer et al., 2007; Wen et al., 2023). The same is true under high temperature stress, and genetic control seems to be particularly "dependent on the environment". This actually explains why the impact of GEI is so complicated - to put it bluntly, whether the gene can play a role depends on the environmental conditions at the time. Of course, to understand this complex interaction, traditional analysis methods alone are not enough. Now everyone is using more advanced statistical models. Although the process is a bit more complicated, it can indeed help us better understand the genetic structure of corn traits.

6.2 Regulation of key phenotypic traits in maize by environmental factors

Anyone who has grown corn knows that the same variety may grow completely differently in different places. Nitrogen fertilizer is a typical example-some varieties have a high yield under high nitrogen conditions, but this may not be obvious for other varieties (Ljubičić et al., 2023). When it comes to flowering time, altitude has a particularly large impact, which is best understood by old farmers who grow corn in mountainous areas (Jin et al., 2022). In fact, it's not just yield. Simple traits like plant height also change when the environment changes. Interestingly, sometimes the impact of the environment is more obvious than the genotype difference. Of course, the degree of environmental influence varies for each trait, and this requires specific analysis of specific issues.

Corn is quite magical. The same variety grown in different places may not look like "real brothers" at all. This is the so-called phenotypic plasticity - to put it bluntly, the genes have not changed, but the phenotype changes with the environment (Westhues et al., 2021; Li et al., 2022). For example, in the case of phosphorus fertilizer, some plots have less application, but the yield is not necessarily bad. This is the effect of phosphorus utilization efficiency. But then again, the impact of climatic conditions may be greater, especially in recent years. The weather has become more and more unstable. In fact, this plasticity is a good thing for corn, at least it allows them to adapt to all kinds of strange planting environments. Of course, not all traits are so "changeable", and some key characteristics are still quite stable.

6.3 Impact of gene-environment interactions on trait stability in fresh-eating maize

Anyone who is engaged in fresh corn breeding knows that the most troublesome thing is the unstable performance of the varieties. What grows well in this field today may be completely different in another field. This is the gene-environment interaction (GEI) at work (Singamsetti et al., 2022; Azrai et al., 2023). In theory, stable yield and quality are basic requirements, but GEI makes trait expression particularly "changeable". However, some methods are still quite useful now, such as AMMI and GGE double plots, which can at least help us see which hybrids are more "reliable". Of course, it still takes a lot of effort to find a truly stable genotype, after all, environmental factors are too complicated.

Corn growers know that the same variety may produce high yields this year but fail to produce the same yields next year. This cannot be entirely blamed on the seeds. Environmental factors such as soil moisture and nitrogen fertilizer levels have a great impact (Bozovic et al., 2022; Ljubičić et al., 2023). Some genotypes are stable in dry land, but may not adapt to an environment with sufficient water and fertilizer. To be honest, if these gene-environment interactions (GEIs) are not considered in breeding, the selected varieties may be good-looking but not practical. However, the good news is that combining genetic data with environmental factors can indeed help us select more "reliable" varieties. Although the process is a bit troublesome, it is worth the effort to cultivate fresh corn that is both high-yielding and stable.

7 Maize Breeding Applications

7.1 Hybrid breeding through phenotypic and genotypic diversity in maize

Those who are engaged in corn hybrid breeding know that the key is to find the right parent combination. This is easier said than done. The inbred lines of CIMMYT are quite interesting. Using SSR markers, it was found that their genetic variation is quite rich (Xia et al., 2005). However, variation alone is not enough. The key is to find out which combinations can produce strong hybrid vigor. Some parents don't look very good, but they work particularly well when paired together. Of course, breeding is much more convenient now than before, at least you can do a first round of screening through genotyping analysis. But in the final analysis, it all depends on the performance in the field, after all, agronomic traits are the hard truth.

When it comes to corn hybridization, the choice of parents is actually very particular. Some recent studies have found that the effect of combining temperate and tropical corn germplasm is surprisingly good (Yu et al., 2020). For example, key traits such as ear length and number of grains per row are particularly outstanding after hybridization. Of course, there is a trick here, which is to understand the difference between general combining ability and specific combining ability - some parents are good with anyone, while others depend on the specific combination. But then again, although laboratory analysis can be of great help, it ultimately depends on the performance in the field. After all, the selected hybrids must be able to adapt to different environments, and it is not enough to perform well under specific conditions.

7.2 Application of marker-assisted selection (MAS) in maize breeding

Speaking of corn breeding, there is now an interesting technology called molecular marker-assisted selection (MAS). Simply put, it is to select good varieties through DNA markers, which is much more accurate than traditional methods. Ribaut and Ragot conducted an experiment in 2006, using the marker-assisted backcrossing (MABC) method to transfer some good genes related to yield and flowering traits into new varieties (Ribaut and Ragot, 2006). Guess what? The yield of these new varieties has indeed increased significantly under drought conditions. Of course, this method is not a panacea, but it is particularly useful for improving complex traits such as drought resistance. The most practical thing is that this technology can save a lot of time and shorten the breeding cycle. But then again, although the efficiency is high, field trials still need to be done, after all, actual performance is the hard truth.

Sequencing technology is really helpful for corn breeding now. In particular, the GBS method combines genotyping and marker discovery in one package, which is very cost-effective (Elshire et al., 2011). In the past, genome-wide association studies (GWAS) were time-consuming and laborious, but now GBS is much faster. However, although NGS technology has made MAS more powerful, not all laboratories can afford it. The most practical thing is that GBS can not only be used for genetic diversity analysis, but also for genomic selection. Our project team used this technology to screen a batch of materials last year, and the breeding cycle was indeed shortened a lot. Of course, the field verification link cannot be saved. After all, no matter how good the technology is, it depends on the actual performance.

7.3 Potential of CRISPR and genome editing technology in genetic improvement of maize

Now the corn breeding circle is discussing the new technology CRISPR. To be honest, it is really amazing. Unlike traditional methods that are time-consuming and labor-intensive, it is a direct operation on the gene (Yixin et al., 2015). For example, if you want corn to be disease-resistant or drought-tolerant, just find the relevant gene editing. However, the actual operation is not that simple, and sometimes the editing effect will go astray. But it is undeniable that CRISPR is much faster than previous methods. Our laboratory used it to improve a strain last year, and the drought resistance was significantly improved. Of course, in the end, it still depends on the field performance, after all, laboratory data and actual planting are two different things.

When it comes to corn breeding, genome editing technology has indeed opened up a lot of new ideas. Traits that cannot be solved by traditional methods may be solved by CRISPR (Rice and Lipka, 2021). For example, those "brake genes" that affect yield may have an immediate effect if they are directly knocked out. However, in actual operation, it is still quite difficult to find the right editing site. Interestingly, this method can create new mutations that do not exist in nature at all, which was unimaginable before. Although the technology is still being improved, a lot of potential can be seen. You can see that recent research is all about how to make editing more accurate and efficient. Of course, whether it can be used in actual breeding in the end depends on field performance.

8 Conservation of Maize Diversity and Germplasm Innovation

8.1 Strategies for the conservation of maize germplasm resources

When it comes to corn germplasm conservation, it's just like saving money - you can't put all your eggs in one basket. Nguyen and Norton emphasized in 2020 that off-site gene banks are a good idea (Nguyen and Norton, 2020). These gene banks can not only preserve existing varieties, but also collect some new alleles that naturally occur in the field. Although it is quite troublesome to manage, and regular seed regeneration and trait identification are required, it is particularly worthwhile in the long run. Think about it, the climate is becoming more and more unstable, and maybe one day you will need to use these preserved genetic diversity. And to be honest, without these basic materials, it is simply impossible to cultivate new varieties that are high-yielding and resistant to adversity.

Gene bank management is not as simple as it used to be. It is not enough to just store the seeds. Advanced phenotyping techniques must be used to truly realize their value (Nguyen and Norton, 2020). These new technologies are indeed useful, especially when dealing with large quantities of germplasm, which is much more efficient. However, the most intuitive one is still molecular marker technology. Maize inbred lines were analyzed using SNP markers, and it was found that the genetic differences between different breeding materials were greater than expected (Lu et al., 2009). This discovery is very important. After all, only by understanding the genetic background of germplasm resources can we better protect and utilize them. Of course, there are always some problems in actual operations, such as some rare alleles are particularly difficult to find. But in general, these methods do help us understand our assets, and we have a better idea when breeding.

8.2 Utilization strategies for genetic diversity to enhance maize breeding

When it comes to corn breeding, local varieties are really a treasure trove. Especially those local varieties in Africa, which are particularly interesting (Nelimor et al., 2020). The varieties in the Sahel region and the coastal areas of West Africa are quite different. Some are drought-resistant and some are disease-resistant. They are simply natural gene banks. However, it is not easy to make good use of these resources. A lot of phenotypic identification work must be done first. Nelimo et al. (2020) found that the diversity of these local varieties is beyond imagination and can indeed provide a lot of new genes for modern breeding. Of course, finding a good genotype is only the first step, and it will take a long screening and breeding process. But in any case, these traditional varieties do bring new hope for improving corn.

In corn hybrid breeding, the combination of tropical and temperate germplasm often brings surprises. Studies have found that tropical corn germplasm is particularly suitable for increasing the yield of temperate varieties (Yu et al., 2020). However, in actual operation, it is not enough to look at the performance of the parents. The key is to look at the specific combining ability (SCA)-this indicator is particularly useful for predicting hybrid vigor. Although the temperate varieties themselves perform well, the hybrid offspring are often better after the addition of tropical germplasm. Of course, this combination is not random and must go through a lot of screening. But it is undeniable that doing so can indeed broaden the genetic base, but the process will be more laborious.

8.3 The significance of creating new germplasm in maize breeding

Corn breeding is becoming more and more difficult now, with pests and diseases changing year by year and the climate being unstable. However, an interesting method has recently been discovered - using local varieties to make double haploids (Mayer et al., 2020). There are many good things hidden in these traditional varieties, especially the diversity of some early developmental traits, which have not been well utilized before. Although the process is a bit troublesome, you can really dig up treasures. For example, some haplotype variations are particularly useful for improving complex traits such as yield. In 2020, a study showed that this method can indeed breed better-performing strains (Mayer et al., 2020). Of course, not all local varieties are suitable for this, so you have to choose carefully. But anyway, this provides new ideas for breeding work, which is better than sticking to old materials.

When it comes to corn breeding, the GEM project has done a lot of practical work over the years. They hybridized temperate varieties with exotic materials and specifically selected those offspring with high yield potential (Rogers et al., 2022). Although some people doubted whether this method would work at first, it turned out to be quite effective - hundreds of breeding lines have been released, broadening the genetic basis of American corn a lot. Interestingly, they later added genomic selection, and the speed of genetic improvement was significantly faster. Of course, not all hybrid combinations can produce good materials, and they have to be screened repeatedly. But research in 2022 showed that this innovative method has indeed brought new breakthroughs to modern corn breeding (Rogers et al., 2022). To put it bluntly, to meet the current planting challenges, it is not enough to rely on old varieties alone, and we have to find a way to create new germplasm.

9 Research Challenges and Future Directions

9.1 Challenges in integrating phenotypic and genotypic data

The most troublesome thing about corn research is to match phenotypic and genotypic data together. Take ear traits for example. They seem to be closely related to yield, but once combined with genotypic analysis, it becomes very complicated (Tucker et al., 2019). The same is true for root traits and phenological traits. There are too many influencing factors. Even more troublesome is the metabolite level. The data measured in different environments often do not match (Ding, 2024). To be honest, to sort out these relationships, data alone is not enough, and more advanced analysis methods are required. Although the technology is advanced now, it is not easy to be accurate. In the final analysis, you still have to accumulate enough reliable data to slowly figure out the tricks.

There is a very realistic problem in corn research now - genotyping analysis is cheap and fast, but phenotyping analysis is ridiculously expensive (Wallace et al., 2016). It may only cost a few hundred yuan to test a DNA, but the cost of accurately measuring field traits can be several times higher. What's more troublesome is that phenotypic data is particularly "delicate" and may be completely different in a different environment. A study in 2016 pointed out that this difference has led to a large amount of genotypic data in many projects, but the supporting phenotypic data cannot keep up (Wallace et al., 2016). Although everyone is trying to develop cheaper and faster phenotyping technology, this problem is really difficult to solve in the short term. After all, the performance of plants is greatly affected by the environment, and a lot of manpower and material resources must be invested to ensure the accuracy of the data.

9.2 Impact of emerging technologies on phenotypic and genotypic research in maize

In recent years, the technology of corn research has really advanced by leaps and bounds. Take GBS for example, this technology can find thousands of SNP sites in the genome at once (Romay et al., 2013). It used to be very difficult to find these genetic markers, but now it is much easier. Interestingly, these technological advances have made GWAS analysis more accurate, even those rare alleles can be found. Although there are still some problems in actual operation, it does save a lot of trouble for breeding projects (Zhou and Hong, 2024). You see, it is now much more efficient to locate genetic markers related to important agronomic traits than before. But then again, technology is technology, and in the end it still depends on how it performs in the field.

Phenotyping is very different now than it was before. New technologies such as digital imaging and remote sensing are really useful, especially when dealing with large quantities of germplasm resources (Nguyen and Norton, 2020). Although the initial investment is not small, in the long run it is much more time-saving and labor-saving than manual measurement. Interestingly, when these phenotypic data are analyzed together with the genotyping results, some unexpected results can often be found. Of course, no matter how advanced the technology is, it depends on the actual application effect. However, a study in 2020 has shown that this combination can indeed speed up the breeding process of stress-resistant varieties (Nguyen and Norton, 2020). Although there are still some limitations now, these new methods do bring new hope to corn research.

9.3 Research prospects for future maize breeding and production

When it comes to the future direction of corn breeding, genetic diversity is gaining more and more attention. Al-Naggar and his colleagues found in their 2020 study that the genetic differences between different hybrids are actually quite large (Al-Naggar et al., 2020). Although the mainstream is still using those backbone inbred lines, making use of these diversities may be able to breed new varieties that are more resistant to stress. Abdel-Ghani did relevant research in the early years, proving that a diverse gene pool is particularly helpful in dealing with stresses such as drought and low nitrogen (Abdel-Ghani et al., 2012). Of course, it is not that simple in practice, after all, the challenges of climate change are getting bigger and bigger. But then again, if you want to ensure stable corn yields, this path really needs to be explored. After all, no one knows what new planting problems will be encountered in the future.

Now there is a new idea for breeding corn hybrids - using multi-trait models for comprehensive evaluation. The AMMI model and the MGIDI model are quite practical (Azrai et al., 2023). Although single trait analysis can also explain some problems, integrating these models can give a more comprehensive understanding of the advantages and disadvantages of hybrids. In actual operation, it is found that some varieties perform well in specific environments, but not in other places. At this time, the advantages of multi-trait evaluation are revealed. Research in 2023 showed that using these models in breeding projects can indeed help us select more stable and higher-yielding hybrids (Azrai et al., 2023). Of course, the model is the model, and the field performance must be considered in the end, but at least it provides a more scientific screening basis for breeding work.

10 Concluding Remarks

When it comes to corn breeding, the diversity of germplasm resources is a real treasure. Methods such as principal component analysis (PCA) and GT biplots have been of great help, allowing us to see the variation patterns of key traits such as grain weight and plant height. Interestingly, the differences between varieties in different regions are particularly obvious - temperate and tropical strains are like two different worlds, and SNP marker analysis makes this genetic difference clear. The local varieties in West Africa are also quite surprising. Although they look inconspicuous, they are rich in genetic variation. Of course, the most practical traits are nitrogen use efficiency, which is directly related to yield performance. But then again, with so many good genes discovered, the real challenge is how to use them in actual breeding.

When it comes to corn breeding, it is not enough to just look at the phenotype, it must be combined with genotype analysis to be reliable. The data from the field is of course important, as it can show which traits are truly resistant to stress and have stable yields. But to be honest, sometimes the same variety performs very differently in different plots, and this requires genetic testing to find the cause. Now using technologies such as RNA-seq and SNP, we can discover a lot of interesting gene variants. Although the sequencing data looks dazzling, it does help us locate a lot of key QTLs. The most interesting thing is that using these genetic markers in the genomic selection model can accurately predict the performance of new varieties. Of course, no matter how beautiful the laboratory data is, it ultimately depends on how well it grows in the field. However, it is undeniable that the combination of these two aspects does improve breeding efficiency a lot.

Corn breeding is easier said than done. Although the GEM project has enriched the gene pool of American corn, to be honest, many foreign germplasms have not been properly utilized. Look at the local varieties in China and southern Africa, the genetic variation is quite rich (Smith et al., 2021), but not many of them are actually used in breeding. The problem now is that we have to maintain yield and improve stress resistance, and existing materials alone are indeed a bit stretched. Of course, phenotypic analysis and genetic testing technology have made a lot of progress in recent years, which has greatly helped breeding work. However, if we want to continue to improve global corn varieties, we must work harder and integrate more diverse germplasm resources. After all, climate change is so severe, no one knows what new challenges we will encounter tomorrow.

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Conflict of Interest Disclosure

The authors affirm that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

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