

Research Insight

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Utilizing High-Throughput Phenotyping for Disease Resistance in Wheat

Benchang Zhang^{1,2}, Jinghuan Zhu², Min Fan², Weidong Wang², Wei Hua² ✉¹ College of Advanced Agricultural Sciences, Zhejiang A&F University, Hangzhou, 311300, Zhejiang, China² Institute of Crop and Nuclear Technology Utilization, Zhejiang Academy of Agricultural Sciences, Hangzhou, 310021, Zhejiang, China✉ Corresponding email: huawecau@hotmail.comMolecular Plant Breeding, 2024, Vol.15, No.5 doi: [10.5376/mpb.2024.15.0023](https://doi.org/10.5376/mpb.2024.15.0023)

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Abstract High-throughput phenotyping (HTP) has emerged as a transformative approach in the field of plant breeding, offering non-destructive, rapid, and precise quantification of a wide array of plant traits. This study explores the utilization of HTP for enhancing disease resistance in wheat. By leveraging advanced imaging technologies and automated data collection systems, HTP platforms can monitor and evaluate phenotypic variations in large wheat populations under diverse environmental conditions. The integration of various sensors, including RGB, hyperspectral, and thermal cameras, enables comprehensive assessment of disease impact and plant responses. This study highlights the potential of HTP to accelerate the identification of disease-resistant genotypes, thereby facilitating the development of robust wheat varieties. The findings underscore the importance of high-resolution imaging, data management infrastructure, and advanced analytical techniques in optimizing HTP applications for crop improvement.

Keywords High-throughput phenotyping; Disease resistance; Wheat breeding; Imaging technologies; Crop improvement

1 Introduction

Wheat (*Triticum aestivum* L.) is one of the most important cereal crops globally, serving as a staple food for a significant portion of the world's population. However, wheat production is continually threatened by various diseases, which can lead to substantial yield losses and affect food security. Enhancing disease resistance in wheat is therefore a critical objective in agricultural research. Traditional breeding methods, while effective, are often time-consuming and labor-intensive. Advanced breeding techniques, including genomic selection and high-throughput phenotyping (HTP), offer promising solutions to accelerate the development of disease-resistant wheat varieties (Juliana et al., 2018; Yates et al., 2019; Khadka et al., 2020).

High-throughput phenotyping (HTP) has emerged as a revolutionary tool in plant breeding, enabling the rapid and precise measurement of phenotypic traits across large populations of plants. HTP utilizes advanced sensor technologies, such as RGB cameras, hyperspectral sensors, and unmanned aerial systems (UAS), to collect vast amounts of data on plant health, growth, and stress responses (Araus and Cairns, 2014; Shakoore et al., 2017; Singh et al., 2019). This technology facilitates the identification of disease-resistant traits by providing high-resolution, non-destructive assessments of plant phenotypes under various environmental conditions. By integrating HTP with genomic selection, breeders can more efficiently screen and select for desirable traits, thereby accelerating the breeding cycle and improving the genetic gain for disease resistance (Juliana et al., 2018; Crain et al., 2019; Danilevicz et al., 2021).

This study provides a comprehensive overview of the latest advancements in high-throughput phenotyping (HTP) technologies and their applications in breeding disease-resistant wheat. It explores various HTP platforms and their capabilities, discusses the integration of HTP with genomic and molecular approaches, and highlights the challenges and future directions in this field. By synthesizing recent research findings, the study emphasizes the potential of HTP to transform wheat breeding programs and contribute to global food security through the development of robust, disease-resistant wheat varieties.

2 Technological Advances in High-Throughput Phenotyping

2.1 Recent developments in sensor technologies

Recent advancements in sensor technologies have significantly enhanced the capabilities of high-throughput phenotyping (HTP) in agriculture, particularly for disease resistance in wheat. Unmanned Aerial Vehicles (UAVs) equipped with various sensors have become a cornerstone of modern phenotyping. These sensors include regular RGB cameras, multispectral imaging cameras, hyperspectral imaging cameras, thermal imaging sensors, and LiDAR sensors. UAVs can collect high-resolution remote sensing data over large field trials, enabling the non-destructive estimation of plant traits such as yield, biomass, height, and leaf area index (Xie and Yang, 2020; Feng et al., 2021).

The use of UAVs offers several advantages over traditional ground-based methods. UAVs can cover larger areas more quickly and provide higher resolution images compared to satellite-based techniques. This increased throughput and frequency of data collection are crucial for identifying crops with high yield and strong stress resistance, including disease resistance (Xie and Yang, 2020). Additionally, UAV-based phenotyping has shown higher precision in assessing traits like grain yield in wheat, making it a valuable tool for early selection cycles in breeding programs (Hu et al., 2020).

2.2 Application of automated image analysis and machine learning in phenotyping

The integration of automated image analysis and machine learning (ML) techniques has revolutionized HTP by enabling the efficient processing and analysis of vast amounts of phenotypic data. Digital Image Processing (DIP) and ML methods can analyze images captured by UAVs to extract valuable information about plant traits. These technologies minimize the time and cost associated with traditional phenotyping methods, making it feasible to analyze entire crops quickly and accurately (Nogueira et al., 2023).

Automated image analysis involves the use of computer vision techniques to process images and extract phenotypic traits. Machine learning algorithms can then be applied to these data to identify patterns and make predictions about plant performance. For example, spectral indices derived from near-infrared (NIR) imaging have shown significant correlations with grain yield in wheat, indicating their potential as indirect selection traits (Hu et al., 2020). Moreover, ML models can be used to predict complex traits such as lodging in wheat, which is influenced by multiple genetic factors (Singh et al., 2019).

2.3 Case studies on the application of HTP in identifying disease resistance loci

Several case studies have demonstrated the effectiveness of HTP in identifying disease resistance loci in wheat. One notable example is the use of UAV-based phenotyping to assess lodging in wheat. Lodging is a complex trait that affects yield and quality, and traditional visual assessment methods are time-consuming and subjective. By using UAVs to capture high-resolution images and generate digital elevation models, researchers were able to quantitatively assess lodging across thousands of wheat plots. This approach led to the identification of a key genomic region on chromosome 2A associated with lodging resistance, highlighting the potential of HTP to uncover genetic factors underlying complex traits (Singh et al., 2019).

Another case study focused on the use of UAVs to monitor disease resistance in wheat breeding programs. High-resolution imaging and spectral indices were used to assess disease symptoms and stress responses in wheat genotypes. The data collected from UAVs were integrated with genomic information to identify disease resistance loci and improve the selection of resistant genotypes. This approach not only increased the efficiency of phenotyping but also provided valuable insights into the genetic basis of disease resistance (Shakoor et al., 2017; Hu et al., 2020).

The advancements in sensor technologies, automated image analysis, and machine learning have significantly enhanced the capabilities of high-throughput phenotyping in agriculture. These technologies have enabled the efficient and accurate assessment of complex traits, including disease resistance in wheat, and have the potential to accelerate crop improvement programs. The case studies presented here demonstrate the practical applications

of HTP in identifying disease resistance loci and improving the selection of resistant genotypes, ultimately contributing to the development of more resilient and productive wheat varieties.

3 High-Throughput Phenotyping in Disease Resistance Trait Identification

3.1 Leveraging HTP for rapid identification of quantitative resistance loci (QTLs)

The identification of QTLs associated with disease resistance is a critical step in breeding programs aimed at developing resistant wheat varieties. HTP technologies, such as unmanned aerial systems (UAS) and automated image analysis, have significantly accelerated this process. For instance, UAS-based phenotyping has been successfully applied to assess complex traits like lodging in wheat, demonstrating high correlations with visual estimates and broad-sense heritability (Singh et al., 2019). This approach allows for the rapid and accurate identification of QTLs, which can then be used in marker-assisted selection (MAS) to develop disease-resistant varieties (Zhu, 2024).

In another study, automated image analysis was employed to measure quantitative resistance to *Septoria tritici* blotch (STB) in wheat. This method enabled the identification of small chromosome intervals containing candidate genes for STB resistance, highlighting the power of HTP in pinpointing specific genomic regions associated with disease resistance (Yates et al., 2019). Similarly, the use of 35K Axiom Array SNP genotyping assays in conjunction with HTP allowed for the identification of novel QTLs for stem rust resistance in wheat, further validating the effectiveness of HTP in QTL identification (Pradhan et al., 2023).

3.2 Successful examples of HTP in detecting resistance to common wheat diseases

HTP has been instrumental in detecting resistance to several common wheat diseases. For example, Singh et al. (2019) demonstrated the use of precision phenotyping to identify novel loci for quantitative resistance to STB. Their study involved a replicated field experiment with 335 winter wheat cultivars, which were phenotyped using automated image analysis. This approach led to the identification of 26 chromosome intervals associated with STB resistance, some of which were novel and others that overlapped with previously known resistance intervals (Singh et al., 2019; Yates et al., 2019).

Another successful application of HTP is in the detection of resistance to *Fusarium* head blight (FHB). A meta-analysis of QTLs associated with FHB resistance identified 209 QTLs across 21 chromosomes, providing valuable markers for marker-assisted breeding (Liu et al., 2009). Additionally, HTP has been used to identify QTLs conferring high-temperature adult-plant (HTAP) resistance to stripe rust in wheat. This study identified eight QTLs significantly associated with HTAP resistance, with major loci on chromosomes 2B and 4A explaining a substantial portion of the phenotypic variation (Chen et al., 2012).

3.3 Importance of integrating phenotypic data with genotypic information for enhanced selection efficiency

The integration of phenotypic data with genotypic information is crucial for enhancing the efficiency of selection in breeding programs. HTP generates large volumes of phenotypic data that, when combined with genotypic information, can provide a comprehensive understanding of the genetic architecture of disease resistance traits. This integration allows for the identification of candidate genes and the development of more accurate genomic prediction models.

For instance, the integration of HTP data with genome-wide association studies (GWAS) has been shown to improve the identification of QTLs for disease resistance. In the case of stem rust resistance, the combination of phenotypic data from HTP with SNP genotyping assays enabled the identification of 20 reliable QTLs, including novel genomic regions that can be used in breeding programs (Figure 1) (Pradhan et al., 2023). Similarly, the use of HTP in conjunction with statistical genomic methods has been proposed to enhance the genetic improvement of longitudinal traits in crops, providing insights into plant functioning and gene activation in response to environmental stimuli (Moreira et al., 2020).

Moreover, the integration of phenotypic and genotypic data can facilitate the development of MAS strategies. For example, MAS has been successfully applied to transfer resistance genes and QTLs into elite breeding material, as

demonstrated by the transfer of rust resistance genes and QTLs for FHB resistance in wheat (Miedaner and Korzun, 2012). The use of high-throughput genotyping platforms and genomic selection further reduces the challenges associated with MAS, opening new avenues for molecular-based resistance breeding.

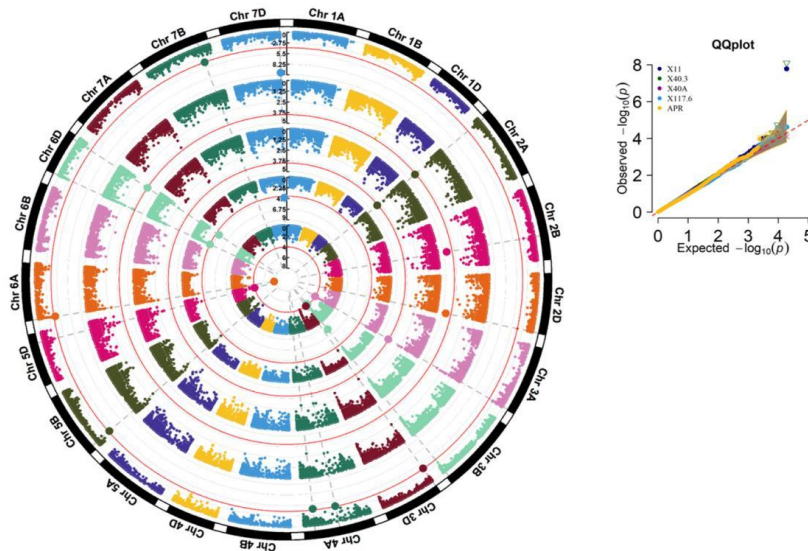


Figure 1 A circular Manhattan plot showing association of the SNP markers in the current study using MLM with their respective plot (Adopted from Pradhan et al., 2023)

Image caption: The inner to outer circle represents the marker significance for seedling (11, 40-3, 40A, and 117-6) and adult plant resistance, respectively. Significant markers above the threshold (at $-\log_{10} [P] > 4$) are highlighted as red-colored circles for each pathotype (Adopted from Pradhan et al., 2023)

HTP has proven to be a powerful tool in the identification of disease resistance traits in wheat. By leveraging HTP for rapid QTL identification, successfully detecting resistance to common wheat diseases, and integrating phenotypic data with genotypic information, breeding programs can achieve enhanced selection efficiency and develop more resilient wheat varieties.

4 Applications of HTP in Wheat Breeding for Disease Resistance

4.1 Ground-based and aerial-based HTP systems in field trials for evaluating disease response

High-throughput phenotyping (HTP) systems, both ground-based and aerial-based, have revolutionized the evaluation of disease responses in wheat breeding programs. Ground-based platforms, such as mobile phenotyping units, are equipped with various sensors to measure canopy height, temperature, and vegetation indices, providing detailed and accurate data on plant health and disease resistance (Figure 2) (Pour et al., 2021). These systems can autonomously collect data, reducing the need for labor-intensive manual measurements and allowing for the rapid assessment of large breeding populations.

Aerial-based HTP systems, particularly those utilizing unmanned aerial vehicles (UAVs), offer significant advantages in terms of coverage and efficiency. UAVs equipped with multispectral and hyperspectral cameras can capture high-resolution images of large field plots, enabling the detection of disease symptoms and stress responses across extensive areas (Haghighattalab et al., 2016; Condorelli et al., 2019). These aerial platforms can quickly assess thousands of plots, providing valuable data on disease incidence and severity, which is crucial for selecting resistant genotypes.

The integration of ground-based and aerial-based HTP systems allows for comprehensive monitoring of disease responses in wheat breeding trials. Ground-based systems provide detailed, close-up measurements, while aerial platforms offer a broader perspective, capturing spatial variability and large-scale patterns of disease spread. This combination enhances the accuracy and efficiency of disease resistance evaluations, ultimately accelerating the development of disease-resistant wheat varieties (Shakoor et al., 2017; Crain et al., 2021).

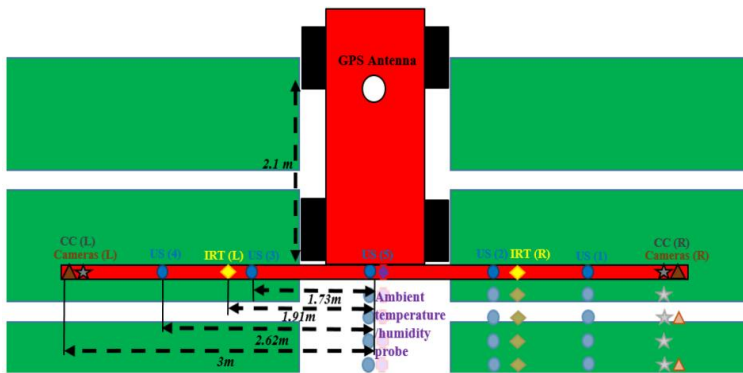


Figure 2 Schematic of developed High-Throughput Phenotyping Platform (HTPP) with utilized devices (Adopted from Pour et al., 2021)

Image caption: Utilized devices distances in the field; IRT, US and CC stand for Infra-Red Thermometer, Ultrasonic Sensor and Crop Circle, respectively (Adopted from Pour et al., 2021)

4.2 Use of HTP for real-time monitoring and prediction of disease outbreaks

HTP technologies have enabled real-time monitoring and prediction of disease outbreaks in wheat fields. By continuously collecting phenotypic data through sensors and imaging systems, HTP platforms can detect early signs of disease and monitor their progression over time. This real-time data collection is critical for timely intervention and management of disease outbreaks.

For instance, UAVs equipped with multispectral cameras can capture temporal changes in vegetation indices, which are indicative of plant health and stress levels. These indices can be used to identify areas of the field that are experiencing disease stress, allowing for targeted application of fungicides or other control measures (Haghighattalab et al., 2016; Adak et al., 2023). Additionally, the integration of HTP data with environmental and weather data can improve the prediction of disease outbreaks, enabling proactive management strategies.

The ability to monitor disease progression in real-time also facilitates the study of disease dynamics and the identification of resistant genotypes. By analyzing temporal phenotypic data, researchers can identify genotypes that exhibit stable resistance across different environmental conditions and disease pressures. This information is invaluable for breeding programs aiming to develop wheat varieties with durable disease resistance (Shakoor et al., 2017; Smith et al., 2021).

4.3 Comparison between traditional phenotyping methods and HTP platforms for precision in large-scale breeding programs

Traditional phenotyping methods in wheat breeding have relied heavily on manual measurements and visual assessments, which are time-consuming, labor-intensive, and often subjective. These methods are limited in their ability to accurately and consistently evaluate large breeding populations, particularly for complex traits such as disease resistance.

In contrast, HTP platforms offer significant improvements in precision, efficiency, and scalability. HTP systems utilize advanced sensors and imaging technologies to collect high-resolution phenotypic data, enabling the accurate measurement of multiple traits simultaneously. For example, UAV-based HTP platforms can capture detailed images of entire field plots, allowing for the precise assessment of disease symptoms and plant health (Haghighattalab et al., 2016; Condorelli et al., 2019). Ground-based platforms equipped with various sensors can provide continuous, non-destructive measurements of plant traits, reducing the need for destructive sampling and increasing the throughput of phenotyping (Pour et al., 2021).

The precision of HTP platforms is further enhanced by the use of advanced data processing and analysis techniques. Machine learning and computer vision algorithms can be applied to HTP data to automatically detect and quantify disease symptoms, reducing the potential for human error and bias (Shakoor et al., 2017; Singh et al., 2019). Additionally, the integration of HTP data with genomic information enables the identification of genetic

loci associated with disease resistance, facilitating marker-assisted selection and genomic prediction in breeding programs (Condorelli et al., 2019; Adak et al., 2023).

HTP platforms provide a more accurate, efficient, and scalable approach to phenotyping in large-scale wheat breeding programs. By enabling the rapid and precise evaluation of disease resistance, HTP technologies accelerate the development of resistant varieties and improve the overall efficiency of breeding efforts (Tolley et al., 2020; Crain et al., 2021; Danilevicz et al., 2021).

5 Challenges of High-throughput Phenotype Analysis in Wheat Disease Resistance

5.1 Technological limitations in sensor accuracy, data processing, and image resolution

High-throughput phenotyping (HTP) in wheat disease resistance faces significant technological challenges, particularly in sensor accuracy, data processing, and image resolution. The precision of sensors is crucial for capturing accurate phenotypic data, yet many current systems struggle with this aspect. For instance, a multi-sensor system developed for field phenotyping in wheat and soybean demonstrated the potential of using various sensors, including ultrasonic distance sensors, thermal infrared radiometers, and NDVI sensors, to measure crop canopy traits. However, the accuracy and resolution of these sensors can be limiting factors, affecting the reliability of the collected data (Figure 3) (Bai et al., 2016).

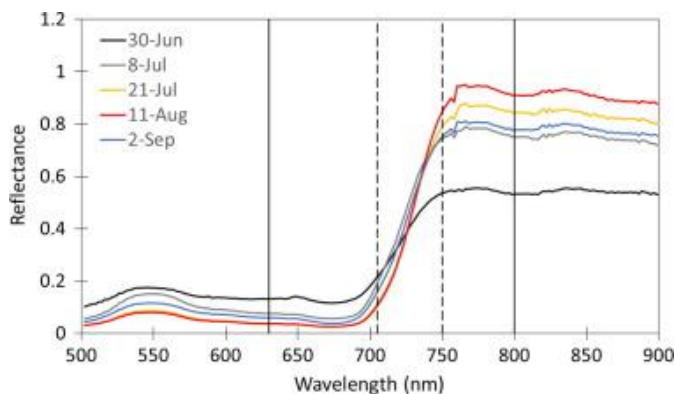


Figure 3 The average canopy reflectance spectra of the soybean plots derived from the up-looking and down-looking portable spectrometers on different dates during the season (Adopted from Bai et al., 2016)

Image caption: The solid vertical lines denote the wavelengths to calculate NDVI (630 and 800 nm) and the dashed vertical lines denote the wavelengths to calculate red-edge NDVI (705 and 750 nm) (Adopted from Bai et al., 2016)

Moreover, the integration of multiple sensors and the synchronization of their data streams require sophisticated data processing capabilities. The development of a LabVIEW program to control and synchronize measurements from all sensor modules highlights the complexity involved in managing these systems. Additionally, image resolution is a critical factor, as high-resolution images are necessary to capture detailed phenotypic traits. The use of RGB cameras and the extraction of canopy green pixel fraction as a proxy for biomass illustrate the importance of high-resolution imaging in phenotyping (Bai et al., 2016).

5.2 Data management issues: handling large-scale phenotypic datasets and integrating them with genotypic data

The management of large-scale phenotypic datasets poses another significant challenge in HTP. The sheer volume of data generated by high-throughput systems can be overwhelming, necessitating robust data storage, processing, and analysis infrastructure. For example, the use of image-based HTP platforms to monitor phenotypic variation in crops generates vast amounts of data that need to be efficiently managed and shared among researchers (Danilevicz et al., 2021).

Integrating phenotypic data with genotypic data adds another layer of complexity. The ability to correlate phenotypic traits with genetic markers is essential for advancing our understanding of disease resistance in wheat. However, this requires sophisticated data integration techniques and computational tools. The development of

automated image analysis to measure quantitative resistance to septoria tritici blotch (STB) in wheat and the association of these phenotypes with SNP markers in a genome-wide association study (GWAS) exemplify the challenges and potential of integrating phenotypic and genotypic data (Yates et al., 2019).

5.3 Overcoming variability in environmental conditions that can affect phenotypic measurements

Environmental variability is a major challenge in HTP, as phenotypic traits can be significantly influenced by changing environmental conditions. This variability can introduce noise into the data, making it difficult to accurately assess disease resistance. For instance, early plant vigor in winter wheat was found to be affected by genotypic differences, but environmental conditions also played a role, highlighting the need for methods that can account for such variability (Kipp et al., 2014).

To address this, some HTP systems incorporate environmental sensors to collect simultaneous environmental data, which can then be used to normalize phenotypic measurements. The integration of solar radiation sensors and air temperature/relative humidity sensors into a multi-sensor system for field phenotyping is an example of how environmental data can be collected alongside phenotypic data to improve the accuracy of measurements (Bai et al., 2016).

Additionally, the use of deep learning and machine learning techniques can help mitigate the impact of environmental variability by identifying patterns and correlations that may not be immediately apparent. For example, the application of deep learning on proximal imaging to score plant morphology and developmental stages in wheat demonstrated high accuracy and heritability, suggesting that advanced computational methods can enhance the robustness of phenotypic assessments under variable environmental conditions (Wang et al., 2019).

While high-throughput phenotyping offers significant potential for advancing wheat disease resistance research, it is not without its challenges. Technological limitations in sensor accuracy, data processing, and image resolution, data management issues, and the need to account for environmental variability all present significant hurdles. Addressing these challenges will require continued innovation in sensor technology, data integration techniques, and computational methods to fully realize the potential of HTP in crop breeding and disease resistance research.

6 Combining Genomic Selection with High-Throughput Phenotyping

6.1 The role of genomic-enabled prediction models in accelerating wheat breeding for disease resistance

Genomic selection (GS) has revolutionized plant breeding by enabling the prediction of breeding values using DNA polymorphisms, thus allowing for the selection of superior genotypes without the need for extensive phenotypic evaluations. This approach is particularly beneficial for complex traits such as disease resistance, which are often controlled by multiple genes with small effects. High-throughput phenotyping (HTP) complements GS by providing rapid, non-destructive measurements of phenotypic traits across large populations, thereby enhancing the accuracy and efficiency of selection processes.

The integration of HTP with GS models has shown promise in improving the predictive ability of these models. For instance, the use of secondary traits measured through HTP, such as canopy temperature and vegetation indices, has been demonstrated to increase the accuracy of genomic prediction models for grain yield in wheat by up to 70% (Rutkoski et al., 2016). This integration allows for the assessment of traits that are difficult to measure directly, thereby providing a more comprehensive understanding of the genotype-to-phenotype relationship.

6.2 Case studies integrating HTP with genomic selection to enhance prediction accuracy and breeding outcomes

Several case studies have highlighted the successful integration of HTP with GS to enhance prediction accuracy and breeding outcomes. For example, a study on wheat breeding demonstrated that the use of UAV-based phenotyping for traits such as lodging could significantly improve the accuracy of genomic predictions. The study found high correlations between digital measures of lodging and visual estimates, with genome-wide association analysis identifying key genomic regions associated with lodging resistance (Singh et al., 2019).

Another study focused on the use of HTP in early selection cycles of wheat breeding. The researchers employed UAVs to assess grain yield in different plot designs and found that aerial-based sensing provided higher precision and stronger correlations with grain yield compared to ground-based spectral sensing. This approach enabled the efficient selection of improved genotypes in early breeding stages, thereby accelerating the breeding process (Rutkoski et al., 2016).

In maize, the integration of field-based HTP with genomic data revealed time-dependent associations between genotypes and abiotic stresses, such as heat stress during flowering. This study demonstrated that combining phenomic and genomic data could significantly improve the prediction ability for complex traits like flowering times and plant height, thereby enhancing the understanding of plant-environment interactions (Adak et al., 2023).

6.3 Future trends in wheat disease resistance research using multi-environmental experiments

The future of wheat disease resistance research lies in the integration of multi-environmental experiments (METs) with advanced genomic and phenomic tools. METs allow for the evaluation of genotypic performance across diverse environmental conditions, providing insights into genotype-by-environment interactions and the stability of disease resistance traits. The use of HTP in METs can facilitate the rapid collection of phenotypic data across different environments, thereby improving the accuracy of genomic predictions and enabling the selection of genotypes with broad-spectrum disease resistance.

One promising trend is the use of envirotyping, which involves the characterization of environmental conditions at various spatial and temporal scales. By combining envirotyping with HTP and GS, researchers can better account for environmental variability and improve the estimation of genotypic performance across different environments (Smith et al., 2021). This approach can help identify genotypes that are resilient to a range of abiotic and biotic stresses, thereby enhancing the overall robustness of wheat breeding programs.

Additionally, the development of machine learning and deep learning algorithms for analyzing HTP data is expected to further advance the field. These algorithms can handle the high dimensionality and complexity of phenotypic data, enabling more accurate predictions of complex traits and the identification of novel genetic associations (Cabrera-Bosquet et al., 2012). As sensor technologies and data analytics continue to evolve, the integration of HTP with GS will become increasingly sophisticated, paving the way for more efficient and effective breeding strategies.

The combination of genomic selection with high-throughput phenotyping holds great potential for accelerating wheat breeding for disease resistance. By leveraging the strengths of both approaches, researchers can enhance the accuracy of predictions, improve the efficiency of selection processes, and ultimately develop more resilient wheat varieties. Future research should focus on the integration of multi-environmental experiments, envirotyping, and advanced data analytics to fully exploit the potential of these technologies in wheat breeding programs.

7 Innovative Wheat Disease Resistance Screening Method Using High-Throughput Phenotype Analysis

7.1 Novel image-based disease quantification approaches

High-throughput phenotyping (HTP) has revolutionized the way we quantify disease resistance in wheat by leveraging advanced imaging technologies. One of the most promising approaches involves the use of multispectral and hyperspectral imaging to capture detailed information about plant health and disease status. For instance, multispectral imaging has been successfully used to monitor barley resistance against powdery mildew by capturing reflectance data at various wavelengths, which helps in differentiating between susceptible and resistant genotypes (Kuska et al., 2018). This method allows for the early detection of disease symptoms, which is crucial for timely intervention and breeding decisions.

In another study, a vehicle-based multispectral active sensor was employed to score early plant vigor in winter wheat, demonstrating the feasibility of using spectral indices to reflect plant health accurately (Kipp et al., 2014). This approach not only enhances the speed and accuracy of phenotyping but also reduces the labor and cost

associated with traditional methods. The integration of these imaging techniques with high-throughput platforms enables the rapid screening of large populations, making it a valuable tool for modern plant breeding programs.

7.2 Use of hyperspectral and multispectral imaging to detect early disease symptoms

Hyperspectral and multispectral imaging technologies have shown great potential in detecting early disease symptoms in wheat. These imaging techniques capture a wide range of wavelengths, providing detailed spectral information that can be used to identify subtle changes in plant physiology caused by pathogen infection. For example, hyperspectral imaging combined with machine learning has been used to monitor plant phenotypes under salt stress, demonstrating its ability to detect physiological and biochemical changes non-destructively (Feng et al., 2020). This approach can be adapted to detect early disease symptoms in wheat, allowing for timely and accurate disease management.

A study comparing UAV-based RGB and multispectral imaging for phenotyping wheat stay-green traits found that multispectral indices, particularly those containing red-edge or near-infrared bands, were more effective in detecting early disease symptoms than RGB indices (Cao et al., 2021). This highlights the importance of selecting appropriate spectral bands for disease detection. The use of hyperspectral imaging to evaluate wheat chlorophyll content under drought stress further supports the utility of this technology in monitoring plant health and detecting early signs of disease (Yang et al., 2023).

7.3 Application of machine learning models for automated disease scoring and resistance evaluation

The integration of machine learning models with high-throughput phenotyping platforms has significantly advanced the automated scoring of disease resistance in wheat. Machine learning algorithms, such as support vector machines (SVM) and deep learning, can analyze complex datasets generated by imaging technologies to identify disease symptoms and quantify resistance levels accurately. For instance, SVM classification was used to identify and quantify powdery mildew in barley, demonstrating the potential of machine learning in automating disease scoring (Kuska et al., 2018; Wu, 2024).

In another study, deep learning approaches were applied to hyperspectral imaging data to segment plant and leaf regions accurately, enabling the precise measurement of physiological traits affected by salinity stress (Feng et al., 2020). This method can be extended to disease resistance screening, where machine learning models can be trained to recognize disease-specific spectral signatures and automate the evaluation process.

Furthermore, the use of ensemble feature selection methods has been shown to improve the accuracy of hyperspectral data analysis by identifying the most informative spectral features for plant phenotyping (Moghimi et al., 2018). This approach can enhance the performance of machine learning models in disease resistance screening by reducing data dimensionality and focusing on relevant features. The combination of hyperspectral imaging and machine learning provides a powerful tool for high-throughput, non-destructive disease resistance evaluation in wheat, facilitating the rapid identification of resistant genotypes and accelerating breeding programs.

The integration of novel image-based quantification approaches, hyperspectral and multispectral imaging, and machine learning models has transformed the landscape of wheat disease resistance screening. These high-throughput phenotyping methods offer unprecedented accuracy, speed, and efficiency, enabling the early detection of disease symptoms and the automated evaluation of resistance, ultimately contributing to the development of more resilient wheat varieties.

8 Future Directions

8.1 Potential advancements in HTP technology to improve accuracy and scalability in disease resistance screening

High-throughput phenotyping (HTP) technology has made significant strides in recent years, but there is still considerable room for improvement, particularly in the context of disease resistance screening in wheat. One of the primary areas for advancement is the enhancement of sensor technologies. Current HTP platforms utilize a variety of sensors, including RGB cameras, hyperspectral sensors, and computed tomography, which provide

valuable data on crop phenotypes (Danilevicz et al., 2021). However, the resolution and accuracy of these sensors can be further improved to better capture subtle phenotypic variations associated with disease resistance.

Another potential advancement lies in the integration of more sophisticated data analysis tools. Machine learning and artificial intelligence (AI) have already shown promise in processing large datasets generated by HTP platforms (Shakoor et al., 2017; Tayade et al., 2022). Future developments could focus on refining these algorithms to improve their predictive accuracy and scalability. For instance, the use of deep learning techniques could enhance the ability to identify and quantify disease symptoms from complex image data, thereby providing more accurate assessments of disease resistance (Smith et al., 2021).

The scalability of HTP systems can be enhanced by developing more robust and user-friendly data management infrastructures. Efficient data storage, retrieval, and analysis are critical for handling the vast amounts of data generated by HTP platforms. Advances in cloud computing and big data technologies could play a pivotal role in this regard, enabling researchers to scale up their phenotyping efforts without being constrained by data management challenges (Araus and Cairns, 2014).

8.2 Integration of artificial intelligence and robotics in fully automated phenotyping systems

The integration of AI and robotics into HTP systems represents a transformative step towards fully automated phenotyping. AI can be employed to automate the analysis of phenotypic data, reducing the need for manual intervention and increasing the throughput of phenotyping processes. For example, AI algorithms can be trained to recognize specific disease symptoms in wheat, allowing for rapid and accurate disease resistance screening (Juliana et al., 2018; Tayade et al., 2022).

Robotics can further enhance the automation of HTP systems by enabling the deployment of phenotyping platforms in diverse field conditions. Unmanned aerial systems (UAS) or drones equipped with advanced sensors can be used to collect high-resolution phenotypic data across large field plots, providing a non-invasive and efficient means of monitoring crop health (Singh et al., 2019). Ground-based robotic platforms can also be utilized to navigate through field plots, capturing detailed phenotypic data at the plant level (Ninomiya, 2022).

The combination of AI and robotics can lead to the development of fully automated phenotyping systems that operate continuously and autonomously. Such systems could significantly reduce the labor and time required for phenotyping, allowing researchers to screen larger populations of wheat for disease resistance traits. Additionally, the integration of AI and robotics can improve the consistency and repeatability of phenotyping measurements, leading to more reliable data for breeding programs (Goggin et al., 2015).

8.3 The role of international collaborations in advancing HTP applications for wheat breeding

International collaborations play a crucial role in advancing the applications of HTP for wheat breeding. Collaborative efforts can facilitate the sharing of resources, expertise, and data, thereby accelerating the development and deployment of HTP technologies. For instance, the establishment of global phenotyping networks can enable researchers to access diverse germplasm collections and phenotyping platforms, fostering the exchange of knowledge and best practices (Danilevicz et al., 2021).

Collaborations between research institutions, industry partners, and governmental organizations can also drive the standardization of HTP protocols and data formats. Standardization is essential for ensuring the comparability and interoperability of phenotypic data across different studies and regions. By working together, stakeholders can develop common guidelines and frameworks for HTP, promoting the widespread adoption of these technologies in wheat breeding programs (Araus and Cairns, 2014).

Furthermore, international collaborations can support the development of training programs and capacity-building initiatives. Training researchers and breeders in the use of HTP technologies and data analysis tools is critical for maximizing the impact of these innovations. Collaborative training programs can help build a skilled workforce capable of leveraging HTP for disease resistance screening and other breeding objectives (Shakoor et al., 2017).

In conclusion, the future of HTP in wheat breeding is promising, with potential advancements in sensor technologies, AI, and robotics poised to enhance the accuracy and scalability of phenotyping systems. International collaborations will be instrumental in driving these advancements, fostering the exchange of knowledge and resources, and promoting the adoption of HTP technologies worldwide. By working together, the global research community can accelerate the development of disease-resistant wheat varieties, contributing to food security and sustainable agriculture.

9 Concluding Remarks

High-throughput phenotyping (HTP) has significantly advanced wheat disease resistance research by enabling the rapid and precise measurement of phenotypic traits across large populations. This technology has facilitated the identification of disease-resistant genotypes by providing detailed and objective data on plant responses to various stressors, including pathogens and environmental conditions. For instance, automated HTP systems have been developed to assess traits such as green leaf area and green normalized difference vegetation index, which are indicative of disease resistance and stress tolerance. Additionally, drone-based HTP has proven effective in quantifying complex traits like lodging, which impacts yield and quality, thereby enhancing the accuracy and efficiency of phenotyping in large breeding nurseries. These advancements underscore the transformative role of HTP in accelerating the breeding of disease-resistant wheat varieties.

HTP holds immense potential to revolutionize future wheat breeding efforts by integrating advanced imaging technologies, machine learning, and genomic selection. The ability to non-destructively measure a wide range of traits, from morphological to physiological, allows for the comprehensive evaluation of genotypes under various environmental conditions. This integration can significantly enhance the selection process for traits associated with disease resistance, drought tolerance, and overall yield stability. For example, the use of spectral indices and remote sensing technologies in HTP enables the precise monitoring of plant health and stress responses, which are critical for developing resilient wheat varieties. Moreover, the scalability of HTP systems, such as unmanned aerial systems (UAS), allows for the high-throughput assessment of thousands of plots, making it feasible to conduct large-scale genetic studies and improve breeding efficiency.

To fully realize the potential of HTP in wheat breeding, continued research and development are essential. Future efforts should focus on enhancing the accuracy and affordability of HTP systems to make them accessible to a broader range of breeding programs. This includes the development of more sophisticated image processing algorithms and machine learning models to better analyze phenotypic data and predict complex traits. Additionally, there is a need for standardized protocols and data-sharing platforms to facilitate the integration and comparison of HTP data across different studies and environments. Collaborative efforts between researchers, breeders, and technology developers will be crucial in advancing HTP technologies and ensuring their effective application in breeding programs. By addressing these challenges, HTP can continue to drive innovations in wheat breeding, ultimately leading to the development of more resilient and high-yielding wheat varieties.

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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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