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Advances in screening and breeding for resistance to late leaf spot and rust in groundnut (*Arachis hypogaea* L.)

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Abstract

Systematic disease screening is essential for identifying resistant sources against two major foliar diseases late leaf spot (LLS) and rust responsible for severe yield losses in groundnut (*Arachis hypogaea* L.) worldwide. These pathogens reduce green leaf area, promote premature defoliation and adversely affect photosynthesis, resulting in substantial pod yield decline under favorable environmental conditions. Comprehensive screening under natural field conditions, artificial epiphytotic setups and controlled greenhouse environments has revealed wide variability in disease response among germplasm accessions, breeding lines and segregating populations. Evaluations of mini core collections, recombinant inbred lines and MAGIC populations have facilitated the identification of moderately resistant and resistant genotypes using standardized disease scoring scales. Artificial inoculation, infector rows and staggered sowing techniques help ensure uniform disease pressure and reliable phenotypic assessment. Quantitative parameters such as incubation period, lesion size, percent defoliation and area under disease progress curve (AUDPC) improve selection accuracy compared to visual scoring alone. Recent advances in molecular screening, including allele-specific and KASP markers linked to resistance loci, enable early and precise identification of resistant genotypes. The integration of phenotypic and molecular approaches enhances selection efficiency and supports the development of stable, high-yielding, disease-resistant groundnut cultivars across diverse agro-climatic regions.

Keywords: Artificial epiphytotic screening, Disease screening, Germplasm evaluation, Late leaf spot, Rust, Molecular validation

Introduction

Groundnut is an economically important oilseed crop cultivated extensively across Asia and Africa. It serves as a vital source of edible oil, protein and income for smallholder farmers. Despite its significance, groundnut productivity remains below potential levels in many developing countries due to persistent biotic stresses. Among these, late leaf spot caused by *Phaeoisariopsis personata* and rust caused by *Puccinia arachidis* are the most widespread and damaging foliar fungal diseases.

Late leaf spot appears as dark necrotic lesions on the lower leaf surface, eventually leading to chlorosis and defoliation. Rust infection manifests as reddish-brown pustules on leaves, petioles and stems. Both diseases reduce the effective photosynthetic area and interfere with assimilate partitioning, ultimately leading to poor pod filling and yield reduction (Killada *et al.*, 2023) [8]. Under favorable environmental conditions, yield losses can be severe due to accelerated defoliation and physiological stress.

Chemical control through fungicide application is effective but costly and environmentally unsustainable when applied repeatedly. Therefore, host plant resistance has emerged as the most economical, farmer-friendly and environmentally safe management strategy. The identification and utilization of resistant genotypes remain central to groundnut breeding programs worldwide.

Economic Importance and Impact of Late Leaf Spot and Rust

The widespread occurrence of foliar diseases is a major reason for low productivity of groundnut in developing regions. Disease severity directly influences plant vigor, canopy duration and pod yield. Increased infection leads to premature leaf fall, weakening of plants and reduction in assimilate translocation to developing pods (Killada *et al.*, 2023) [8].

Recent field evaluations confirmed strong negative associations between disease severity and key physiological functions, emphasizing that effective disease management is crucial for sustaining crop productivity (Killada *et al.*, 2023) [8]. Consequently, breeding for resistance remains a priority objective in many groundnut improvement programs. In environments where fungicidal control is limited by cost or accessibility, host plant resistance offers a sustainable and farmer-friendly solution. Resistant cultivars help maintain functional leaf area for a longer duration, ensuring continued photosynthesis and improved pod filling. Moreover, stable resistance reduces dependency on chemical inputs, lowers production costs and minimizes environmental impact. Therefore, integrating durable genetic resistance into high-yielding backgrounds is essential for enhancing groundnut productivity under disease-prone agro-ecological conditions.

Early Germplasm Screening and Identification of Resistance Sources

Systematic screening for foliar disease resistance began several decades ago. Subrahmanyam *et al.* (1982) [19] conducted field and greenhouse evaluations and identified genotypes such as GP-NC-343 and NC-5 with resistance to both rust and late leaf spot. Their pioneering work established the feasibility of identifying dual-resistant genotypes.

Sneh-Mathur *et al.* (1987) [17] screened 131 varieties and reported seven resistant and eleven moderately resistant entries. Waliyar *et al.* (1988) [21] reviewed resistance research and outlined strategies for identifying new resistance sources. Mosses *et al.* (1989) identified 54 accessions resistant to *Phaeoisariopsis personata*. Further large-scale germplasm evaluations strengthened resistance identification.

Dinakaran *et al.* (1992) [5] screened 28 genotypes and identified five resistant lines to both diseases. Waliyar *et al.* (1993) [22] evaluated 424 lines and identified multiple disease-resistant genotypes such as ICG-1707 and ICG-6330. Mehan *et al.*, (1996) [11] screened 979 accessions and confirmed 28 resistant lines. Field studies conducted by Roy, (2017) [15] based on 1-9 scale (Figure **1** &2) across diverse environments consistently revealed variability in disease reactions. These investigations demonstrated that resistance exists within cultivated and exotic germplasm pools, though immunity is rare and resistance is largely quantitative.

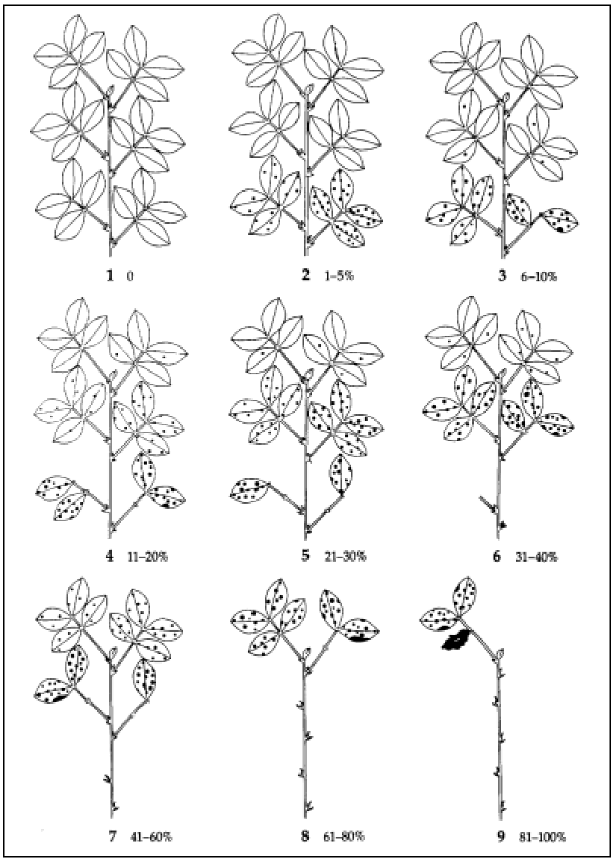

Fig 1

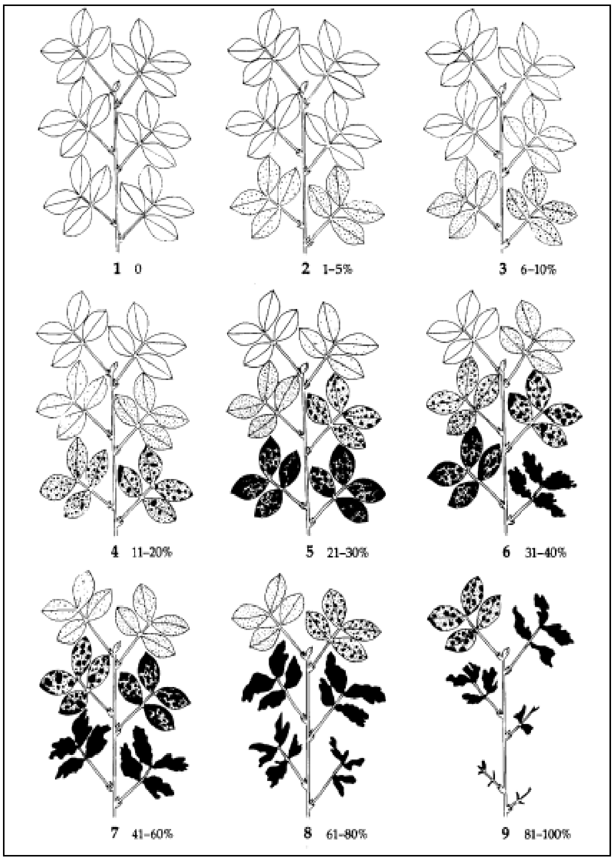

Fig 2

Modified 9- point scale (Subrahmanyam *et al.*, 1995) [18] used for field screening for reaction to late leaf spot and rust resistance

Variability, Inheritance and Genetic Parameters

Understanding genetic variability and inheritance patterns is essential for resistance breeding. Gowda *et al.* (1996) [7] studied variability in crosses and reported complex associations between productivity traits and resistance. While high productivity was sometimes associated with larger pods, resistant lines occasionally exhibited smaller pod size, indicating possible trade-offs.

Asish (2014) evaluated 816 recombinant inbred lines and reported high heritability and high genetic advance for rust and late leaf spot resistance, suggesting additive gene action and effectiveness of direct selection. Divyadharsini *et al.* (2016) [6] observed significant genetic variability among F3 cross derivatives and reported superior crosses for specific disease resistance. High heritability coupled with minimal difference between phenotypic and genotypic coefficients of variation indicated limited environmental influence (Asish, 2014). These findings confirm that resistance traits can be reliably improved through phenotypic selection.

Artificial Epiphytotic Screening and Phenotyping Advances

Reliable phenotyping is critical for identifying stable resistance. Sudini *et al.* (2015) [20] screened the peanut mini core collection under artificial epiphytotic conditions, ensuring uniform disease pressure. The use of infector rows, staggered sowing and controlled inoculation techniques enhanced disease expression.

Disease scoring based on standardized 1–9 scales became widely adopted. Quantitative parameters such as incubation period, lesion diameter and area under disease progress curve (AUDPC) improved selection precision (Mushtaq *et* *al.*, 2024) [13]. Resistant genotypes showed lower AUDPC values and reduced defoliation compared to susceptible ones.

Recent developments in visual disease estimation further improved accuracy. Cazon *et al.* (2025) [4] demonstrated that standardized area diagrams reduced subjectivity in severity estimation. Larsen *et al.* (2025) [10] emphasized objective quantification of infected leaf area to ensure consistent assessment across environments. These advancements collectively enhanced reliability and reproducibility of phenotypic screening.

Recombinant Inbred Lines and MAGIC Populations

The development of recombinant inbred lines (RILs) facilitated detailed genetic analysis of resistance. RIL populations provided stable genetic backgrounds for evaluating additive effects and heritability (Asish, 2014). MAGIC (Multi-parent Advanced Generation Inter-Cross) populations further broadened genetic diversity. Wankhade *et al.* (2021) [23] evaluated MAGIC lines and identified several entries with longer incubation periods, smaller lesion diameters and lower disease scores than resistant checks. These populations enhance recombination frequency and facilitate identification of superior allelic combinations. Such populations are valuable for dissecting quantitative resistance and identifying stable donors for breeding.

Molecular Screening and Marker-Assisted Selection

Molecular breeding has revolutionized resistance improvement. Bhawar *et al.* (2020) [3] used allele-specific primers to identify resistant germplasm lines for foliar diseases. Molecular validation reduces reliance on late-stage phenotypic screening.

Killada *et al.* (2024) [9] validated disease-linked markers across diverse genetic backgrounds, confirming their strong association with reduced foliar disease severity. Pandey *et al.* (2024) [14] developed and validated KASP markers linked to resistance genes such as LISR1, LISR2 and LRI1. These markers enabled precise identification of resistant alleles and facilitated release of improved lines combining disease resistance and desirable oil quality traits. Marker-assisted selection accelerates breeding cycles, enhances precision and supports gene pyramiding for durable resistance.

Integration of Phenotypic and Molecular Approaches

Recent studies emphasize combining artificial screening with molecular tools. Saravanan *et al.* (2025) [16] demonstrated that combining artificial field screening with molecular marker analysis enabled clear and consistent discrimination between resistant, moderately resistant and susceptible genotypes. Their study showed that phenotypically resistant lines consistently possessed resistance-linked alleles, confirming the genetic basis of observed resistance. This dual approach minimized the possibility of disease escape and ensured that selected genotypes carried stable resistance genes rather than temporary field tolerance. Such integration strengthens confidence in selection decisions and enhances reproducibility across seasons and locations.

Similarly, Bhat *et al.* (2024) [2] highlighted the significant role of Virginia-type germplasm as a reliable reservoir of resistance to both late leaf spot and rust. Their findings confirmed GPBD 4 as a stable and durable donor genotype under high disease pressure conditions. The consistent performance of GPBD 4 across environments underscores the value of well-characterized resistance donors in breeding programs. When validated molecular markers are deployed alongside rigorous phenotypic screening, breeders can effectively pyramid resistance genes, accelerate selection cycles and improve durability of resistance. This integrated breeding strategy not only enhances efficiency but also contributes to the development of high-yielding, disease-resilient cultivars adaptable to diverse agro-climatic regions.

Challenges in Resistance Breeding

Despite considerable progress in identifying resistant sources and deploying molecular tools, several challenges continue to limit the development of durable resistance to late leaf spot and rust in groundnut. Resistance to these foliar diseases is predominantly quantitative in nature and governed by multiple genes with small to moderate effects. As a result, resistance expression is often influenced by environmental conditions such as temperature, humidity and disease pressure intensity. This genotype × environment interaction complicates selection, particularly when screening is conducted under variable natural infection conditions.

Another important constraint is the occasional negative association between resistance and yield-related traits. Gowda *et al.* (1996) [7] reported that certain resistant lines were associated with undesirable agronomic features such as smaller pod size or altered shell thickness, thereby necessitating careful recombination and selection strategies. Balancing high yield potential with durable disease resistance remains a key breeding objective.

Pathogen variability and the emergence of new virulent races further threaten resistance stability. Continuous exposure of resistant cultivars to pathogen populations may exert selection pressure, leading to breakdown of resistance over time. Therefore, multi-location and multi-season testing is essential to confirm stability across diverse agro-climatic zones. Gene pyramiding, involving the accumulation of multiple resistance genes within a single genotype, offers a promising strategy to enhance durability and reduce the risk of resistance erosion.

Future Perspectives

Future resistance breeding strategies must adopt an integrated and forward-looking approach to address evolving disease challenges. Expanding germplasm exploration, including utilization of landraces and wild relatives of groundnut, may uncover novel alleles conferring enhanced resistance to late leaf spot and rust. Broadening the genetic base is crucial to avoid vulnerability associated with narrow parentage.

Strengthening multi-environment testing will ensure identification of genotypes with stable resistance across varied ecological conditions. Advances in genomic technologies, including high-density marker systems and genome-wide association studies, can further dissect the genetic architecture of resistance traits. Integration of genomic tools with high-throughput phenotyping platforms will improve precision in identifying superior lines.

Large-scale deployment of validated KASP markers can accelerate early-generation selection and facilitate gene pyramiding strategies. Marker-assisted backcrossing and genomic selection approaches may significantly reduce breeding cycle duration while improving selection accuracy.

The convergence of conventional breeding methods, validated molecular markers and objective phenotyping tools presents strong opportunities for developing high-yielding, disease-resilient cultivars. Such an integrated strategy will play a pivotal role in ensuring sustainable groundnut production and long-term management of foliar diseases under changing climatic conditions.

Conclusion

Extensive disease screening efforts have established the presence of exploitable variability for late leaf spot and rust resistance in groundnut. High heritability and additive gene effects support effective phenotypic selection, while validated molecular markers enhance precision breeding. Artificial epiphytotic screening, objective disease quantification and integration of genomic tools collectively accelerate the development of durable, high-yielding resistant cultivars. Continued integration of phenotypic and molecular strategies will be critical to sustaining groundnut productivity under increasing disease pressure.

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