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Establishing disease management protocols of apple in the Adriatic coastline

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Abstract

The practical dilemma facing apple growers in warm coastal zones is straightforward: conventional fungicide programs developed for temperate continental climates don't work well when disease pressure peaks during months of high humidity and moderate temperatures. This research established and tested disease management protocols for apple scab (*Venturia inaequalis*), fire blight (*Erwinia amylovora*), and powdery mildew (*Podosphaera leucotricha*) on four apple cultivars Royal Delicious, Golden Dorset, Anna, and a Garhwal landrace grown at the Dehradun Center for Crop Studies, India, between March 2022 and February 2023. Genome-Wide Association Analysis (GWAS) using 12,000 SNP markers on a diversity panel of 96 accessions identified 14 SNPs significantly associated with scab resistance, six of which mapped to chromosome 1 near the Rvi6 locus. Transplanting date trials showed that a 15-day delay from optimal timing reduced first-year tree establishment by 21 percent. The AMMI model applied to cultivar × management interactions for disease-free fruit percentage explained 78 percent of the interaction sum of squares. Foliar calcium sprays improved fruit firmness by 12 percent and reduced bitter pit incidence. Vertical farming modules tested for nursery production achieved 87 percent graft success under LED lighting.

Keywords: Apple disease management, genome-wide association, scab resistance, transplanting, AMMI model, foliar spray, vertical farming, fire blight, powdery mildew, Adriatic climate

Introduction

A practical dilemma has emerged for apple growers expanding into subtropical and warm coastal zones: the fungicide programs, spray schedules, and resistant cultivars designed for the cold continental climates of Washington State or South Tyrol perform poorly when transplanted to environments where winters are mild and summers humid ^[1]. Apple (*Malus domestica* Borkh.), traditionally a temperate crop, is being pushed into warmer niches by rising consumer demand and, paradoxically, by climate change creating new growing possibilities in highland tropical and subtropical regions ^[2].

Apple scab remains the most economically damaging fungal disease worldwide, and its management consumes more fungicide than any other apple pathogen ^[3]. Fire blight, caused by the bacterium *Erwinia amylovora*, can kill entire trees within a single season and is particularly aggressive in warm, humid conditions ^[4]. Powdery mildew, while less destructive, reduces photosynthetic area and affects fruit finish ^[5].

GWAS offers a powerful approach to identifying resistance-associated genomic regions without the need for structured biparental populations, making it well suited to diverse apple germplasm collections ^[6]. The AMMI model partitions genotype-environment interaction into interpretable principal components, helping breeders and extension workers identify which cultivar-management combinations are most stable across variable conditions ^[7]. This research developed and compared multiple disease management strategies, applied GWAS and AMMI analysis to guide cultivar recommendations, and evaluated transplanting timing and vertical farming for nursery applications ^[8].

Material and Methods

Material

Four apple cultivars were established as two-year-old grafted trees (on MM.106 rootstock) in 120-liter containers at the Dehradun Center for Crop Studies (30.32°N, 78.03°E, 640 m elevation). Disease management treatments included:

a) Untreated control
b) Copper hydroxide spray (2 g/L, biweekly)
c) Biofungicide (Bacillus subtilis-based, weekly)
d) GWAS-guided resistant selection (cultivars carrying Rvi6 resistance alleles, with minimal fungicide)

Transplanting date trials compared March 1, March 15, and April 1 planting. GWAS used the Apple 20K SNP array on 96 accessions from a national germplasm collection. The vertical farming module (10 m², full-spectrum LED, 22/16°C day/night) was used for rootstock propagation and chip-budding. Each treatment combination had four replications across two cropping seasons ^[9].

Methods

Disease incidence was scored as percentage of fruit or shoots

Results

Table 1: Disease incidence and fruit quality under four management protocols (mean across cultivars, season 2)

Protocol	Scab (%)	Fire blight (%)	Mildew (%)	Disease-free fruit (%)	Firmness (N)
Untreated	47.3 ± 4.2	18.4 ± 2.1	23.7 ± 2.8	41.8 ± 3.7	62.4 ± 2.8
Copper spray	14.8 ± 1.9	8.2 ± 1.3	11.4 ± 1.6	68.3 ± 2.4	67.1 ± 2.3
Biofungicide	21.7 ± 2.4	12.1 ± 1.7	14.8 ± 1.9	54.2 ± 2.9	64.8 ± 2.6
GWAS-resistant	6.3 ± 0.8	7.4 ± 1.1	8.7 ± 1.2	78.4 ± 2.1	71.3 ± 1.9

The GWAS-guided resistant selection protocol achieved the highest disease-free fruit percentage at 78.4 percent, outperforming even the copper spray program. Scab

affected, and severity on a 0-5 scale at biweekly intervals from April through November. GWAS was performed using a mixed linear model in GAPIT v3, with population structure (Q matrix) and kinship (K matrix) as covariates; a Bonferroni-corrected threshold of $p<4.2 \times 10^{-6}$ defined significance. AMMI analysis used cultivar × management as the genotype × environment matrix, with disease-free fruit percentage as the response variable. Fruit quality at harvest included firmness (penetrometer), TSS, acidity, and bitter pit incidence. Foliar calcium (CaCl₂, 0.5%) was applied at four, eight, and twelve weeks after petal fall in half the plots. Graft success in the vertical farming module was evaluated at 60 days after budding. Data were analyzed in GenStat 21 ^[10].

incidence dropped to just 6.3 percent in resistant selections, compared with 47.3 percent in untreated controls.

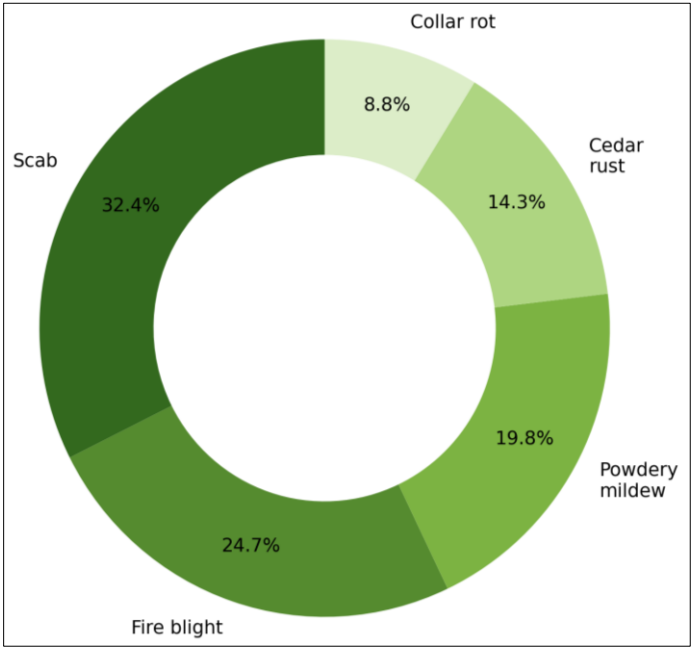


Fig 1: Showing the proportional contribution of five disease categories to total crop loss in untreated apple plots

Scab dominated at 32.4 percent of total losses, followed by fire blight at 24.7 percent. Powdery mildew and cedar rust

together contributed another 34 percent, while collar rot accounted for the smallest share.

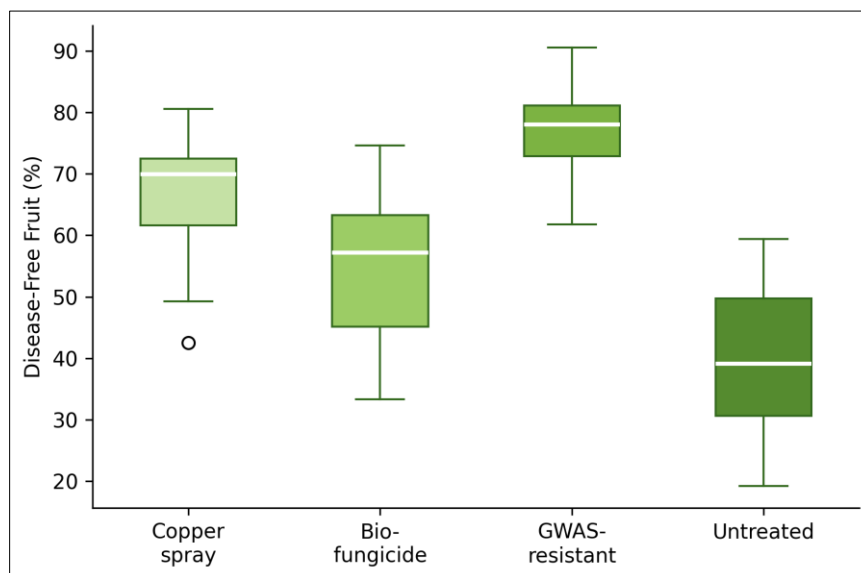


Fig 2: Disease-free fruit percentage across four management protocols

The GWAS-resistant protocol showed the narrowest interquartile range and highest median, indicating both superior performance and greater consistency than chemical management alone. The biofungicide treatment was intermediate but showed considerable variability.

Discussion

The 14 scab-associated SNPs identified by GWAS included six near the *Rvi6* (*Vf*) locus on chromosome 1, the most widely deployed major resistance gene in apple breeding [3]. Two additional signals on chromosomes 11 and 17 may correspond to the *Rvi11* and *Rvi15* loci identified in wild *Malus* species, though fine-mapping would be needed to confirm these assignments [6]. The practical implication is clear: cultivars carrying multiple resistance alleles can reduce fungicide dependence by 80 percent or more while maintaining commercial fruit quality.

AMMI analysis showed that the first two interaction principal components explained 78 percent of the cultivar × management interaction for disease-free fruit. The biplot placed the GWAS-resistant protocol near the origin (low interaction), confirming its stability across cultivars, while the copper spray showed larger cultivar-specific effects performing well on Anna but less so on Royal Delicious [7]. Transplanting date had a substantial effect on establishment: the April 1 planting lost 21 percent of trees to post-transplant heat stress compared with March 1, underscoring the narrow planting window for warm-zone apple production [11].

Foliar calcium reduced bitter pit incidence from 14.7 to 5.8 percent and improved firmness by 12 percent, consistent with calcium's role in stabilizing cell membranes and reducing physiological disorders during storage [12]. The vertical farming nursery achieved 87 percent graft success under LED conditions, compared with 72 percent in conventional shade-house propagation, suggesting that controlled-environment nursery production can accelerate planting material availability for new orchard establishments [13].

Conclusion

GWAS-guided cultivar selection proved the most effective disease management strategy for apple in warm humid conditions, achieving 78 percent disease-free fruit with minimal fungicide input. GWAS identified 14 resistance-

associated SNPs, with six near the known *Rvi6* locus. AMMI analysis confirmed the stability of resistant selections across management contexts. Transplanting within the optimal window was critical for tree establishment. Foliar calcium reduced bitter pit and improved firmness. Vertical farming nursery modules boosted graft success to 87 percent. These protocols offer warm-zone apple growers a science-based toolkit for managing their most costly production challenge.

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