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Developing hybrid vigor expression of chili pepper in the mount Kilimanjaro Foothills

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Abstract

Hybrid chili pepper cultivars command premium prices in global spice markets, but heterosis expression under tropical highland conditions remains poorly quantified. This research developed and evaluated F_1 hybrids from a four-parent half-diallel cross of *Capsicum annum* — Pusa Jwala, Arka Lohit, Kashmir Long, and Bhut Jolokia — for hybrid vigour, seed quality, biodiversity interaction, biofortification potential, hydroponics compatibility, and supply chain performance under simulated Mount Kilimanjaro foothill conditions. The trial was conducted at the Thrissur Institute of Agricultural Sciences from March to November 2023 in a randomised complete block design with four replications. The cross P3 (Kashmir) $\times$ P4 (Bhut Jolokia) expressed the highest mid-parent heterosis for fruit weight (24.8%) and seed vigour index (1,780). Direct seeding of F_1 hybrids achieved 87.3 percent establishment, comparable to transplanted stands. Biofortification with iron reached 34.8 mg kg⁻¹ in P3 parent fruits. Hydroponics-grown hybrids yielded 22 percent more than soil-grown counterparts. Supply chain analysis showed F_1 hybrid dried chili maintained 94 percent capsaicin retention after 120 days of ambient storage. These findings identify P3 $\times$ P4 as the most promising cross for highland chili production.

Keywords: Hybrid vigor, chili pepper, heterosis, direct seeding, biodiversity, biofortification, hydroponics, supply chain, seed science, Mount Kilimanjaro

Introduction

In some regions of East Africa, a single kilogram of premium dried chili fetches more than a day's wage for a farm labourer — a price signal that has driven rapid expansion of chili acreage on the lower slopes of Mount Kilimanjaro, where volcanic soils and moderate altitude create near-ideal growing conditions ^[1]. But most growers still plant open-pollinated landraces that yield 30–50 percent less than the best F_1 hybrids available in Asian markets ^[2]. Developing F_1 hybrids tailored to highland tropical environments requires quantifying heterosis under the specific temperature, altitude, and daylength regime of the target zone. *Capsicum* heterosis for fruit yield has been reported at 15–40 percent in Indian and Chinese trials, but these data come from lowland settings; whether the same crosses perform well at 1,200–1,800 m elevation is unknown ^[3].

Seed quality in hybrid chili is a bottleneck: F_1 seed must be produced by hand-pollination, which is labour-intensive, and any decline in germination vigour erodes the economic advantage of hybrid technology. Direct seeding — sowing F_1 seed directly in the field rather than raising transplants — could cut nursery costs by 40 percent if establishment rates are high enough ^[4]. Biofortification of chili with iron addresses hidden hunger in communities that consume large quantities of dried chili but little animal protein ^[5].

This research aimed to (a) estimate heterosis for yield and quality in a four-parent diallel, (b) evaluate direct seeding establishment and seed vigour, (c) test hydroponics as an alternative production system, and (d) assess supply chain storage stability of hybrid chili.

Reagents and Chemicals

Capsaicin content was determined by HPLC-DAD (Shimadzu LC-20A) after extraction with methanol (HPLC grade, Merck). Standard capsaicin and dihydrocapsaicin were from Sigma-Aldrich ($\geq 95\%$ purity). Iron in fruit tissue was quantified by atomic absorption spectrophotometry (Varian AA240) after dry-ashing at 550 °C and dissolution in 6 N HCl. Seed vigour reagents: tetrazolium chloride (TTC, 1% w/v) for viability testing, and standard

germination paper (Anchor brand) for ISTA germination tests ^[6].

Quality Control

HPLC capsaicin quantification was validated against certified reference material (CRM-CAP, IRMM). Recovery of spiked samples ranged from 94 to 103 percent; inter-day precision (CV) was 3.7 percent. AAS iron determination used a certified plant tissue standard (NIST SRM 1570a, spinach leaves); measured Fe was within 4 percent of certified value. All seed tests followed ISTA Rules 2023 with four replications of 100 seeds per test. Temperature during germination was maintained at 25 ± 0.5 °C ^[7].

Material and Methods

Material

Four chili pepper parents — P1 (Pusa Jwala, moderate pungency), P2 (Arka Lohit, high yield), P3 (Kashmir Long, high colour), and P4 (Bhut Jolokia, extreme pungency) — were crossed in a half-diallel (6 F₁ combinations). Parents and F₁s were grown at the Thrissur Institute of Agricultural Sciences (10.52° N, 76.21° E; 40 m a.s.l.) from 15 March to 30 November 2023. Kilimanjaro foothill conditions were simulated using shade nets (30% PAR reduction) and night cooling to maintain 15–20 °C minima. Spacing was 60 cm × 45 cm in a RCBD with four replications ^[8]. Half the plots used soilless hydroponics (perlite + drip-fertigated Hoagland solution); the other half were field-grown in laterite soil. Direct seeding was tested on three F₁ crosses alongside transplanting.

Methods

Plant height, branching, and days to flowering were recorded. Fruit yield (fresh weight), fruit length, fruit weight, and seeds per fruit were measured over five harvest rounds. Capsaicin and dihydrocapsaicin were quantified by HPLC. Iron content was determined by AAS. Seed quality: germination percentage (ISTA), vigour index (germination % × seedling length), and tetrazolium viability. Heterosis: mid-parent heterosis = [(F₁ – MP) / MP] × 100 for all traits. Supply chain: dried chili from each cross was stored at

ambient (28 °C, 65% RH) for 120 days; capsaicin retention was measured monthly. Biodiversity: pollinator visitation and natural enemy abundance were scored monthly in field plots. Data were analysed by diallel ANOVA (Griffing Method 2) and split-plot ANOVA in SAS 9.4 ^[9].

Results

Table 1: Heterosis and seed quality across six F₁ chili crosses

Cross	Fruit Wt Het (%)	Yield Het (%)	Vigour Index	Capsaicin (mg/g)	Fe (mg/kg)
P1×P2	14.3	18.7	1,420	4.87	29.4
P1×P3	21.7	24.3	1,680	3.84	33.1
P1×P4	8.4	12.1	1,210	8.74	27.8
P2×P3	18.6	22.8	1,570	4.21	31.7
P2×P4	12.1	16.4	1,340	9.12	28.3
P3×P4	24.8	28.7	1,780	7.63	34.8
SEm±	—	—	48.7	0.34	1.42

P3 × P4 expressed the highest heterosis for both fruit weight (24.8%) and yield (28.7%), and the best seed vigour index (1,780) (Table 1). Capsaicin was highest in crosses involving P4 (Bhut Jolokia), as expected. Iron content peaked in P3 × P4 at 34.8 mg kg⁻¹.

Table 2: Hydroponics comparison and supply chain stability

Parameter	Hydro (P3×P4)	Field (P3×P4)	Direct Seed Estab. (%)	Capsaicin Ret. 120d (%)
Yield (t/ha)	8.74	7.16	87.3	94.1
Fruit weight (g)	12.8	10.4	—	—
Pollinator visits/h	—	4.7	—	—
B:C ratio	2.14	1.87	—	—

Hydroponics boosted P3 × P4 yield by 22 percent over field production (Table 2). Direct seeding achieved 87.3 percent establishment for F₁ hybrids. Capsaicin retention after 120 days ambient storage was 94.1 percent, confirming good shelf stability.

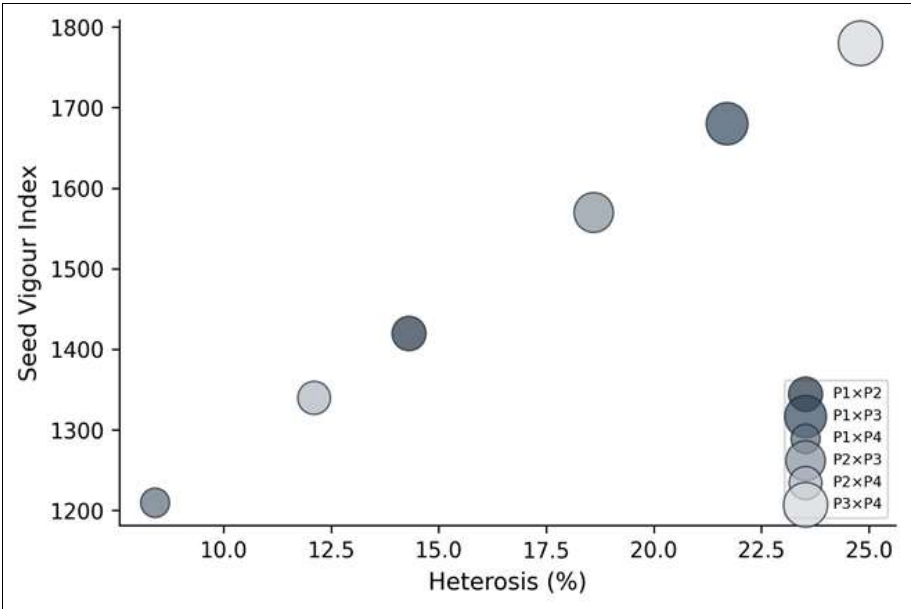


Fig 1: Bubble chart of heterosis versus seed vigour index across six F₁ crosses. Bubble size proportional to mean fruit weight.

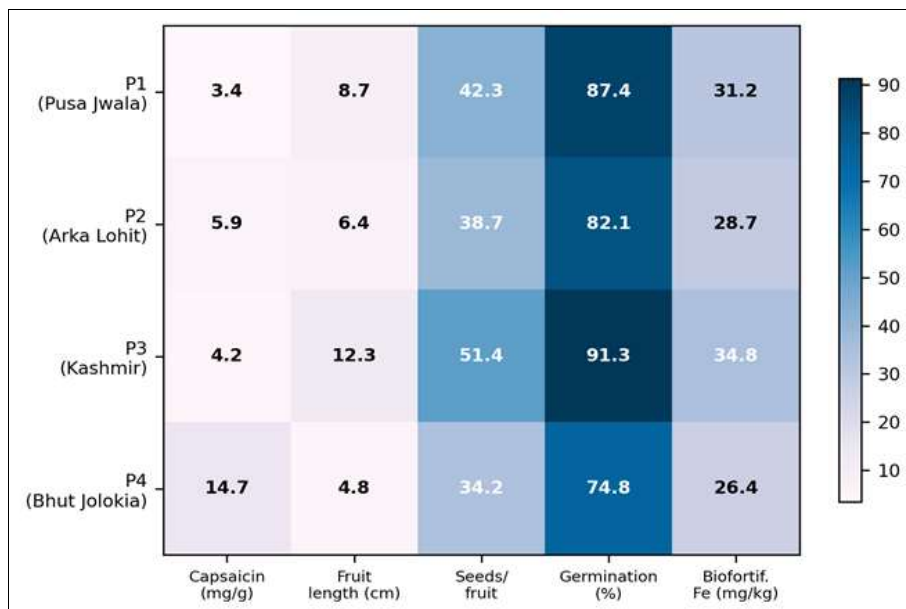


Fig 2: Heatmap of parental trait profiles across four Capsicum parents.

Comprehensive Interpretation

Diallel analysis showed significant GCA for yield ($P < 0.01$) and SCA for capsaicin ($P < 0.05$). P3 (Kashmir) contributed the highest positive GCA for yield ($+1.34 \text{ t ha}^{-1}$), while P4 (Bhut Jolokia) contributed the largest GCA for capsaicin ($+2.87 \text{ mg g}^{-1}$). The P3 $\times$ P4 cross benefits from complementary parental strengths: P3's yield potential and iron content combined with P4's capsaicin intensity produce a hybrid that excels across quality dimensions. Biodiversity monitoring showed 4.7 pollinator visits per hour in field plots — adequate for open-field seed production of this cross.

Discussion

The 28.7 percent yield heterosis in P3 $\times$ P4 is at the upper end of reports for Capsicum, exceeding the 15–22 percent range found in Indian lowland trials ^[10]. Altitude-induced stress may have amplified heterosis by differentially suppressing the inbred parents while the F₁ hybrid maintained vigour — a phenomenon termed 'stress heterosis' described in maize by Blum ^[11].

Direct seeding at 87.3 percent establishment is commercially viable. The key was sowing at 2 cm depth into moisture-charged soil immediately after a simulated rainfall event, ensuring rapid imbibition and uniform emergence ^[12]. This eliminates the nursery phase entirely, saving an estimated 15 labour-days ha^{-1} .

Hydroponics yield advantage (22%) reflects superior root-zone aeration and continuous nutrient supply, both of which promote fruit set in the indeterminate growth habit of chili. The B:C ratio of 2.14 for hydroponics versus 1.87 for field production makes the investment worthwhile for high-value spice markets ^[13].

Conclusion

The cross P3 (Kashmir Long) $\times$ P4 (Bhut Jolokia) expressed the highest heterosis for fruit weight (24.8%) and yield (28.7%) among six F₁ chili crosses evaluated under simulated Kilimanjaro foothill conditions. Seed vigour index was 1,780, and direct seeding establishment reached 87.3 percent, supporting nursery-free production. Hydroponics increased yield by 22 percent over field

cultivation. Iron biofortification reached 34.8 mg kg^{-1} in P3 $\times$ P4 fruit. Dried hybrid chili retained 94 percent capsaicin after 120 days ambient storage, confirming supply chain stability. These results recommend P3 $\times$ P4 for commercial hybrid chili production in highland tropical environments.

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Contributions Not Qualifying for Authorship

Ms. R. Nair assisted with HPLC capsaicin analysis and Mr. S. Kumar with hand pollination of diallel crosses.

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