



RESEARCH ARTICLE

Optimal window for seed physiological and harvestable maturity to maximise seed yield and quality in little millet (*Panicum sumatrense* Roth. ex Roem. & Schult.)

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Abstract

Little millet (*Panicum sumatrense* Roth. ex. Roem. and Schult.) is a climate-resilient, nutrient-rich crop, yet information is limited to its seed physiological maturity and optimal harvest stage, which are the key determinants for maximising seed yield and quality. Experiments across two post-rainy seasons with three cultivars (TNAU 63, Gujarat vari 1, DHLM 36-3) examined seed development and maturation behaviour from anthesis to maturity. Seed formation commenced at 7 days after anthesis (DAA) in year 1 but varied in year 2 (7–14 DAA) across cultivars. Seeds were harvested at weekly stages up to 56–63 DAA and assessed for seed physiological and biochemical traits. Seed moisture content declined and hardness increased steadily with maturity, while test weight, seed yield, germination, vigour and storability peaked between 35–42 DAA, marking physiological maturity at 35 DAA across cultivars in year 1 and at 35 DAA (Gujarat vari 1) or 42 DAA (TNAU 63, DHLM 36-3) in year 2, reflecting temperature-driven variation. Polynomial regression analysis further validated this maturity window. At physiological maturity, little millet panicles turn straw to dark brown, while the oval seeds appear smooth, shiny and streaked in shades of greenish brown to yellow. Upon de-husking, the seeds exhibited a brownish depression at the hilar end, which serve as reliable morphological marker of physiological maturity in little millet. Early harvests produced immature, low-vigour seeds, while delayed harvests reduced seed yield and quality due to seed shattering and deterioration. The study identifies 35–42 DAA as the physiological and harvestable maturity window for high-quality seed production in little millet.

Keywords: harvest stage; little millet; morphological indicators; physiological maturity; polynomial regression; seed yield; seed quality

Introduction

Little millet (*Panicum sumatrense* Roth. ex. Roem. and Schult.) is one of the important minor millet crops, a hardy cereal cultivated extensively in the tropics and subtropics, particularly across South and Southeast Asia. It serves as a staple food for low-income groups in several countries. Belonging to the genus *Panicum*, little millet has been domesticated in India approximately 4000–5000 years ago, with its origins traced primarily to the Eastern Ghats (1, 2). It has historically been cultivated in India, Myanmar, Nepal and Sri Lanka (3), with India as the leading producer, accounting for more than 98 % of global area of production (4). Taxonomically, little millet is a tetraploid species ($2n = 4x = 36$) with a basic chromosome set of $x = 9$ (5). Little millet bears a panicle type inflorescence consisting of spikelets with two florets, a lower sterile floret and an upper bisexual floret each with a distinct lemma and palea (6). Morphologically, the crop is short in duration and hardy, growing 30–90 cm tall with hollow-jointed stems and linear leaves up to 50 cm long.

Agronomically, little millet is climate-resilient, adapted to low-fertility soils, drought-prone arid and semi-arid regions and even conditions of waterlogging. It exhibits high water-use efficiency,

tolerance to salinity and resistance to many pests and diseases (2, 7, 8). These characteristics render little millet as a promising crop for enhancing food and nutritional security in marginal environments. However, factors such as poor harvesting, drudgery in processing and palatability issues have limited its large-scale adoption (9, 10). Nutritionally, little millet grains are rich in protein, dietary fibre, polyunsaturated fatty acids (PUFAs), iron (Fe), zinc (Zn), phosphorus (P) and magnesium (Mg), with low glycemic index and high antioxidant activity (11, 12). Limited studies have focused on its morpho-agronomic diversity, quality traits and genetic resources (13–15), though landraces from regions such as the Eastern Ghats, particularly Koraput recognised as a global agricultural heritage site holds immense potential (8, 16).

The production of high-quality seeds is central to achieving the full genetic potential of improved varieties and ensuring robust crop stands. Seed quality encompassing viability, vigour, uniformity and storability depends critically on the timing of harvest relative to seed developmental stages. Harvesting seeds too early or too late can compromise seed performance, reduce germination and shorten storage life (17, 18). Thus, identifying physiological maturity and optimum stage of harvest is a key challenge in seed production,

especially for under-studied crops like small millets. In small millets, delayed harvesting often results in seed shattering, which not only lowers overall yield but also reduces seed quality and storability due to seed deterioration in the field under prolonged exposure to unfavourable environmental conditions (19). Given the scarcity of systematic studies on physiological and harvestable maturity in small millets and the cultivation of little millet across diverse seasonal and environmental conditions, there is a need to precisely define the optimum harvest stage for different cultivars and seasons. This study aims to characterise seed physical and physiological traits over time, to identify the physiological maturity point and optimal harvest window for little millet seed production.

Materials and Methods

Experimental material and design

Field experiments were carried out during the late post-rainy seasons of 2018-19 (year 1) and 2019-20 (year 2) at ICAR-Indian Institute of Millets Research (ICAR-IIMR), Hyderabad (17.3850° N, 78.4867° E; 505 m above mean sea level (AMSL)), India. The experiment was laid out in a randomised complete block design (RCBD) with 3 replications using three little millet cultivars: TNAU 63 (C1), Gujarat vari 1 (C2) and DHLM 36-3 (C3). The details of these three cultivars are presented in Table 1. In the year 1, after 100 % anthesis, seed formation was first observed at 7 days in all the three cultivars. Accordingly, the experimental treatments were eight harvest stages (HS) scheduled at weekly intervals from 7–56 days after anthesis (DAA) to monitor seed development and maturation during year 1 (Table 2). However, in the year 2, only in cultivar C2 the commencement of seed formation was seen at 7 DAA, whereas in cultivars C1 and C3 the seed formation was recorded at 14 DAA and accordingly eight harvests were scheduled from 14–63 DAA for cultivars C1 and C3 (Table 2).

Seed yield, moisture content, hardness and test weight

Seed weight per plant was determined from five randomly selected and tagged plants at each harvest stage, with values expressed in grams (g). Seed yield per net plot (6 m²) was extrapolated for every 100 plants and recorded in g. At each harvest stage, fresh seed

samples were collected to assess moisture content using a digital seed moisture meter (FARMEX-MT-PRO, FARMCOMP, Agro-electronics, USA), calibrated specifically for millet seeds. Moisture content was expressed as a percentage.

Seed hardness was evaluated using a digital hardness tester (Pharmag Instruments Ltd., India), which applies tension or compression to individual seeds through a force gauge. Four replications of 10 seeds each were selected from each sample for testing and the mean hardness was reported in kilogram force (kgf).

Test weight was determined by counting and weighing four replicates of 100 seeds from each sample, with results expressed in milligrams (mg).

Seed germination, seedling growth and vigour indices

Seed germination assays were performed following the protocols outlined by the International Seed Testing Association (20) for little millet. Germination counts were taken on the 7th day and seedlings were subsequently evaluated for growth and vigour characteristics. Germination percentage (G) was calculated as the proportion of normal seedlings obtained per 100 seeds. From these, 10 normal seedlings were randomly selected for measurement of growth traits.

Root length was measured from the collar region to the tip of the primary root, while shoot length was recorded from the collar region to the tip of the first leaf. Seedlings exhibiting abnormal morphology were excluded from analysis. For seedling dry weight, the selected seedlings were oven-dried at 80 °C for 24 hr, cooled in a desiccator for 30 min and weighed to obtain the mean dry weight.

Seedling vigour indices were computed using the following formulae: (i) seedling vigour index-I (SVI-I) = {germination (%) × seedling length (cm)}; (ii) seedling vigour index-II (SVI-II) = {germination (%) × seedling dry weight (g)}. Field emergence (FE) was assessed for both fresh and accelerated-aged seeds. Fifty seeds per replication were sown in four replications within cement pots (45 cm diameter) filled with a mixture of red and black soils at 1 : 1 ratio. After 20 days, the number of seedlings with leaves emerging above the soil surface was recorded and field emergence was expressed as a percentage.

Table 1. Pedigree details of little millet cultivars TNAU 63, Gujarat vari 1 and DHLM 36-3

Sl. No	Cultivar	Pedigree	Institute - developed	Year - released	Maturity (Days)	Average yield (q/ha)	Areas of adaptation	Special features
1	TNAU 63	Selection from germplasm MS 2369	TNAU, Coimbatore	1997	90–100	11–12	Tamil Nadu, Karnataka, Gujarat	High grain and fodder yield
2	Gujarat vari 1	Pure line selection from local germplasm var. 8-3	Waghai, NAU, Navsari	1980	80–85	15–16	Gujarat	Non-lodging medium tall plants, yellow lustrous grains, purple pigmentation on stem and panicles, blunted tips in tillers
3	DHLM 36-3	Co-4 x Paiyur - 2	ARS, Hanumanmatti, UAS, Dharwad	1978	95–100	14–16	National	Late maturing variety

Table 2. Mean number of days required after anthesis (DAA) at different harvest stages (weekly intervals) after stage-I (at visibility of seed setting) to stage-VIII (at visibility of over-matured seeds) in little millet cultivars TNAU 63, Gujarat vari 1 and DHLM 36-3 across years

Harvest stages (HS)	TNAU 63 (C1)		Gujarat vari 1 (C2)		DHLM 36-3 (C3)	
	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
I	7	14	7	7	7	14
II	14	21	14	14	14	21
III	21	28	21	21	21	28
IV	28	35	28	28	28	35
V	35	42	35	35	35	42
VI	42	49	42	42	42	49
VII	49	56	49	49	49	56
VIII	56	63	56	56	56	63

Seed storability

The accelerated ageing (AA) test was conducted in an ageing chamber (Memmert-HPP 108/749, Germany) following a standard procedure (21). Seeds sampled from each stage of maturation were subjected to accelerated ageing under controlled conditions. A single layer of seeds was placed on stainless steel mesh racks inside the chamber, which maintained a relative humidity of 90 % at a temperature of $44 \pm 1^\circ\text{C}$. Seeds were exposed to these conditions for predetermined duration of 96 hr, established through prior standardisation specific to little millet.

α -Amylase enzyme activity in seedlings

Dry seeds (0.2 g) were placed in Petri plates lined with moistened filter paper to facilitate germination. Samples were harvested on the 2nd, 4th and 7th day of germination. α -amylase activity was determined using the 3,5-dinitrosalicylic acid (DNS) method as described (22). The samples in triplicates were homogenised with 15 mL of ice-cold buffer (20 mM phosphate buffer, pH 6.9) in pre-chilled mortar and pestle kept over crushed ice. An amount of 100 mg of polyvinyl pyrrolidone was added during homogenisation. The homogenate was then centrifuged at $20000 \times g$, 4°C for 10 min. The supernatant (200 μL) was then mixed with 300 μL distilled water and 500 μL of phosphate buffer. After which, 1 mL starch solution (1 % w/v) was added and thoroughly mixed and incubated for 3 min at 25°C . The reaction was then brought to a halt by adding 2 mL DNS reagent (3, 5-dinitrosalicylic acid, 1 % w/v), the test tubes were then incubated in a boiling water bath for 5 min. The test tubes were cooled down and vortex mixed after addition of 20 mL distilled water. Finally, the absorbance of the resultant solution was measured at 530 nm in an ultraviolet-visible (UV-VIS) spectrophotometer. Enzyme activity was expressed as the number of micromoles of maltose released per min by the total enzyme extract obtained from 100 seeds (after converting the sample weight into number of seeds using test weight of respective harvest stage) ($\mu\text{mol} \cdot \text{min}^{-1} \cdot 100 \text{ seeds}^{-1}$).

Electrical conductivity

Leachate conductivity was determined by soaking 1 g of seed from each harvest stage in 100 mL of deionised water at 25°C for 24 hr. Following incubation, the electrical conductivity of the seed leachate was measured using a conductivity meter (Systronics model-306, India). Results were expressed as micro-siemens per centimeter per gram of seed ($\mu\text{S} \cdot \text{cm}^{-1} \cdot \text{g}^{-1}$).

Statistical analysis

Data was subjected to arc-sine transformation when required and analysis of variance (ANOVA) was performed using a randomised complete block design in Statistix software (version 8.1). The second-order polynomial regression models using Microsoft Excel were plotted to determine the optimum harvest stage for achieving maximum seed yield, vigour and storability.

Results and Discussion

Seed maturity in little millet is often non-uniform within and among panicles as well as across plant populations. This variability makes it essential to identify the precise stage of physiological and harvestable maturity window to harvest high-quality seed while minimising yield losses from shattering at delayed harvests. Harvestable maturity emphasises the readiness of the seed

population rather than individual seeds. Across two post-rainy seasons, the days to anthesis among three millet cultivars ranged from 57–76 days after sowing (DAS), reflecting variability both among cultivars and across seasons. In the 1st year, all three cultivars recorded seed development within 7 DAA. However, in the 2nd year, this timeline diverged: cultivar C2 maintained seed formation at 7 DAA, while cultivars C1 and C3 exhibited a delay, with seed development commencing only at 14 DAA (Table 2). This variation highlights the differential physiological responses of plants to seeds under changing climatic parameters across years during two post rainy seasons (Fig. 1). Such delay in seed development from one year to another could be attributed to the temperature stress effect during anthesis and seed formation in the crop. During the transition from seed development to maturation, a series of coordinated cell divisions and differentiations occur, with distinct processes at the stages of early reserve accumulation and late maturation drying. From fertilisation onward, seeds undergo sequential physiological and biochemical changes until they become independent of the mother plant following the cessation of nutrient supply (23). During seed maturation, the developing seeds progressively acquire desiccation tolerance, with a visible transition of the panicles from green to brown, which indicates physiological drying. At maturity, the seeds exhibit a characteristic streaked appearance with coloration ranging from green to yellowish-brown, an oval shape and a smooth, lustrous surface. The de-husked seeds display a distinct brownish depression at the hilar end, that serves as a morphological marker for physiological maturity and time of harvest (Fig. 2). Importantly, desiccation tolerance is acquired during early maturation, with the underlying protective mechanisms being activated in orthodox seeds (24).

Seed moisture, hardness and yield

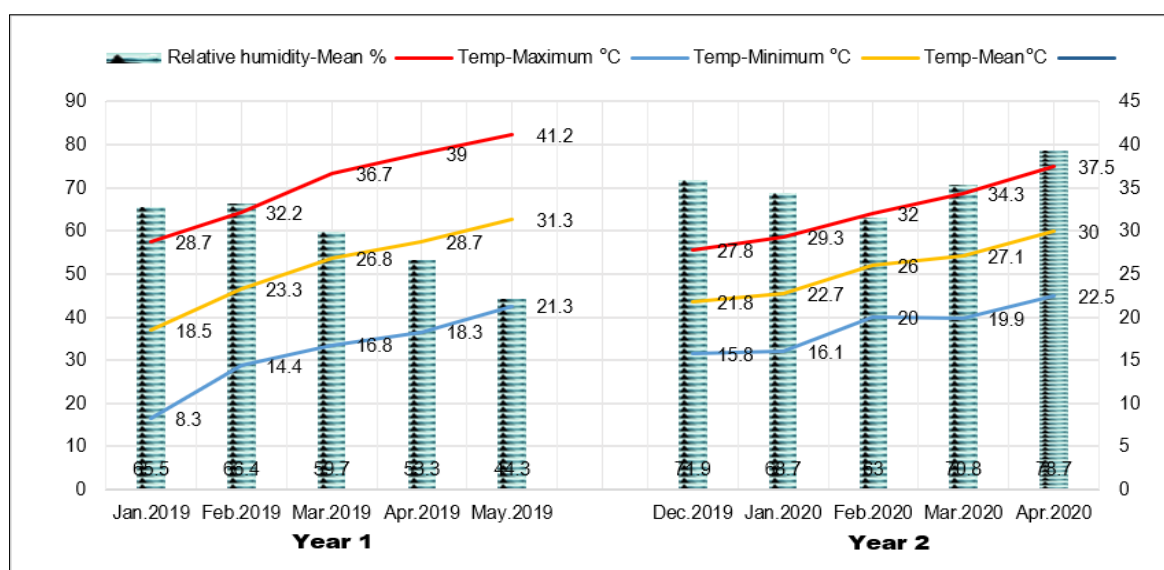
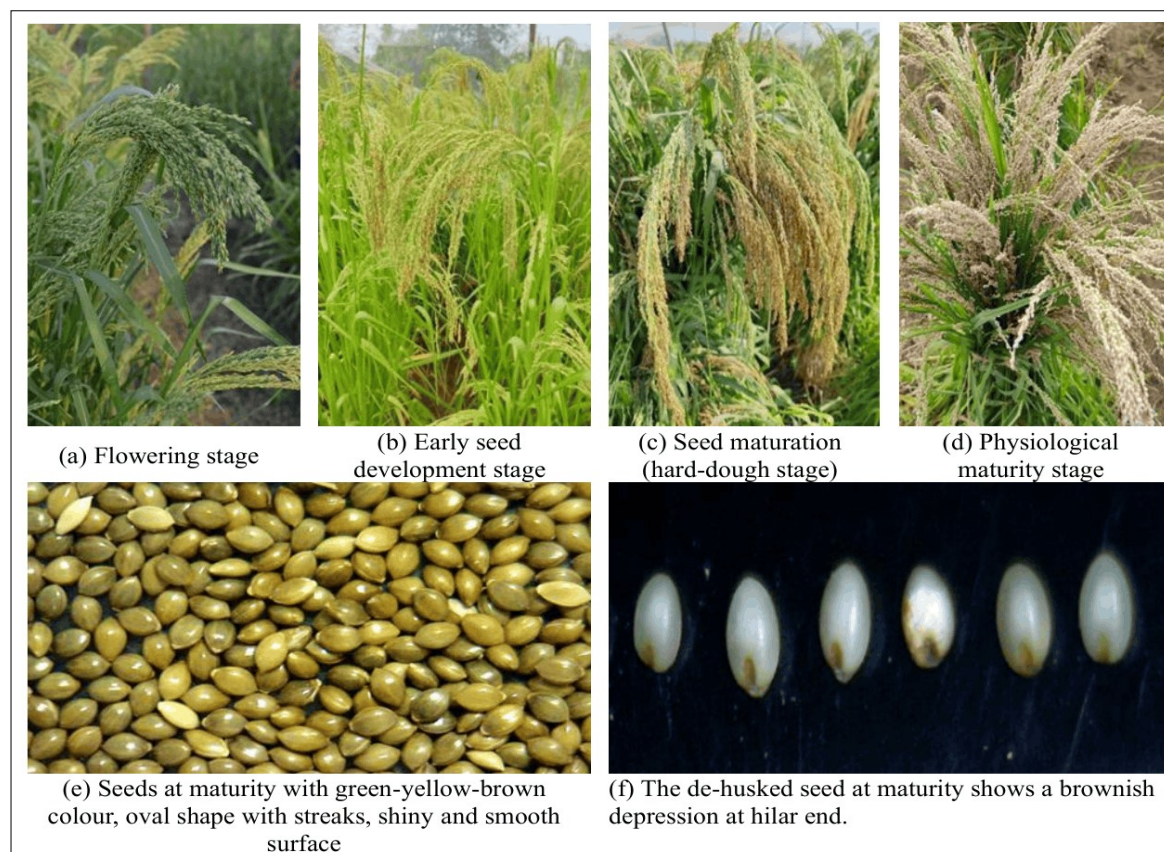
The seed agronomic traits such as moisture content (MC), seed hardness (HN), seed test weight (STW) and seed yield were significantly influenced by HS across cultivars and years, reflecting the progressive nature of seed maturation and subsequent deterioration with late harvests.

Moisture content declined steadily with advancement in seed development and maturation across all three cultivars in both years (Table 3). Early harvests (7–14 DAA) recorded higher MC (18–20 % in year 1 and 13–14 % in year 2), which was closely associated with lower seed weight, poor germination, higher electrical conductivity and reduced seedling vigour, indicating physiologically immature status of the seeds. As maturation progressed, MC decreased gradually and stabilised around 11–12 % between 35 and 42 DAA, signifying the attainment of physiological maturity. Beyond this stage, further reduction in MC did not result in improvement of seed quality; instead, over-drying and increased hardness at later harvests (49–63 DAA) was associated with deterioration in vigour and field emergence. This pattern reflects the typical desiccation phase of seed development, wherein maturation drying enhances storability up to an optimum threshold, beyond which aging-related damage becomes evident (25, 26). Among the cultivars, DHLM 36-3 consistently recorded slightly lower MC across stages, suggesting faster drying rates, possibly due to genotypic differences in seed coat characteristics, as also reported in proso millet (27). The seed desiccation phase during maturation is critical for the acquisition of desiccation tolerance until optimal stage, during which protective molecules such as LEA (late embryogenesis

Table 3. Effect of different stages of seed development and maturation on seed moisture, hardness and test weight in little millet cultivars across years

Harvest stages	MC (%)						HN (kgf)						STW (mg)					
	C1		C2		C3		C1		C2		C3		C1		C2		C3	
	Yr. 1	Yr. 2	Yr. 1	Yr. 2	Yr. 1	Yr. 2	Yr. 1	Yr. 2	Yr. 1	Yr. 2	Yr. 1	Yr. 2	Yr. 1	Yr. 2	Yr. 1	Yr. 2	Yr. 1	Yr. 2
I	20.3	13.4	18.4	13.7	18.2	13.3	1.09	1.12	1.03	1.05	1.22	1.15	159	234	142	209	211	263
II	18.8	13.0	17.6	13.2	17.5	12.9	1.15	1.17	1.08	1.12	1.26	1.19	173	238	155	213	223	276
III	18.0	12.5	16.6	12.8	16.5	12.4	1.19	1.23	1.15	1.16	1.29	1.25	185	245	172	219	232	282
IV	17.0	12.0	15.4	12.3	15.7	12.0	1.23	1.28	1.18	1.20	1.34	1.30	197	250	184	229	243	288
V	15.8	11.5	14.3	11.8	14.8	11.6	1.27	1.33	1.24	1.25	1.37	1.35	212	257	193	239	261	300
VI	14.8	11.0	13.4	11.2	14.0	11.1	1.30	1.38	1.27	1.29	1.43	1.38	194	246	179	225	248	291
VII	13.9	10.4	12.2	10.7	13.1	10.7	1.34	1.43	1.29	1.35	1.47	1.45	182	242	166	220	237	288
VIII	12.9	10.0	11.0	10.2	11.7	10.2	1.36	1.48	1.33	1.39	1.50	1.49	168	237	156	216	225	281
Mean	16.4	11.7	14.9	12.0	15.2	11.8	1.24	1.30	1.20	1.23	1.36	1.32	184	244	168	221	235	284
SEM ±	0.09	0.05	0.08	0.06	0.23	0.03	0.00	0.007	0.005	0.004	0.009	0.004	1.33	2.39	1.19	0.46	1.49	1.31
CD (5 %)	0.47	0.27	0.44	0.30	1.18	0.17	0.03	0.03	0.03	0.02	0.04	0.02	6.67	11.9	5.96	2.30	7.42	6.55

C1: cultivar TNAU63; C2: cultivar; Gujrat vari 1; C3: cultivar DHLH36-3; MC: moisture content; HN: hardness; STW: seed test weight; Yr.1: year 1; Yr.2: year 2.

**Fig. 1.** Temperature and relative humidity during two post rainy seasons of crop experimentation.**Fig. 2.** Little millet panicles and seeds at different stages of seed development and maturation.

abundant) proteins accumulate (28).

Seed test weight (STW) showed a clear stage-dependent trend across cultivars and years, reflecting progressive dry matter accumulation followed by post-maturity losses (Table 3). At early harvests (7–14 DAA), STW was markedly lower due to incomplete endosperm development and high moisture content, which coincided with poor germination, low seedling dry weight and higher electrical conductivity, owing to physiological immaturity of the seeds. From 21–35 DAA, STW increased steadily as reserve deposition intensified, paralleling reductions in moisture content, improved membrane integrity and enhanced seedling vigour. Maximum STW was consistently recorded between 35 and 42 DAA, corresponding with physiological maturity, minimum solute leakage and peak germination and vigour across cultivars, with C3 (DHLM 36-3) recording the highest seed weight, followed by C2 and C1, reflecting genotypic differences in assimilate partitioning efficiency. At later harvests (49–63 DAA), STW declined uniformly across cultivars, likely due to respiratory losses, shriveling and field weathering, which also coincided with increased electrical conductivity and reduced germination and field emergence. These results confirm that although seed size is genotype-dependent, optimal seed weight is achieved only within a narrow harvest window around physiological maturity, after which deterioration outweighs further gains in dry matter (28–30).

Seed yield per plant and per plot followed patterns closely aligned with morphological and physiological seed traits (Fig. 3). At early harvests (7–14 DAA), yields were low due to incomplete seed set and inadequate dry matter accumulation. Across the years, seed yield increased progressively with advancing maturity and reached a maximum between 35 and 42 DAA window, coinciding with physiological maturity, at which peak test weight, superior germination and minimum electrical conductivity were recorded. Thus, indicating that the optimal harvestable maturity of little millet falls within 35–42 DAA as well. Harvestable maturity is the stage or window when about 80 % of the seed population has reached physiological maturity, making it the optimal point for harvesting to balance seed yield, quality and storability. This stage reflects population readiness rather than individual seed maturity. Cultivar C1 consistently recorded higher yield compared to C2 and C3, indicating its genetic potential with greater source-sink strength and reproductive efficiency, whereas C3, despite larger seed size, produced relatively lower yield. In year 1, yields were comparatively lower than year 2, likely due to high temperature stress (Fig. 1) impairing photosynthesis and reproductive tissue integrity, resulting in poor seed set (31). Delayed harvests beyond 49 DAA until 63 DAA caused a rapid decline in yield, attributed to seed shattering, bird damage and prolonged exposure to field conditions (19, 32). In accordance with these results, the second order polynomial regression analysis predicted the seed yield in all the three cultivars, where the maximum seed yield per plant was recorded at 35 DAA in all the three cultivars C1 ($R^2 = 0.86$), C2 ($R^2 = 0.69$) and C3 ($R^2 = 0.90$), respectively in year 1. Beyond 35 DAA, the yield began to decline gradually; however, at subsequent harvests the reduction became more rapid due to accelerated seed drying and shattering. In a different trend during year 2, the cultivars C1 and C3 recorded maximum yield at 42 DAA, with R^2 values of 0.91 and 0.88, respectively (Fig. 3). In the case of C2, highest seed yield ($R^2 = 0.97$) was recorded at 35 DAA similar to year 1, thereafter slight reduction in seed yield at 42 DAA however the differences between these two

stages were non-significant, emphasising the importance of harvesting at the appropriate maturity stage to maximise seed recovery (28).

Seed germination, vigour and viability

Seed physiological quality parameters, including germination (G), field emergence (FE), seedling growth, vigour indices and electrical conductivity (EC), showed a strong and consistent variation across different HS across cultivars and years. At early harvests (7–14 DAA), seeds exhibited low germination (< 75 %), poor seedling growth, lower field emergence and high EC values ($39.4\text{--}43.8 \mu\text{S}\cdot\text{cm}^{-1} \cdot \text{g}^{-1}$), reflecting immature embryos, incomplete reserve accumulation and weak membrane integrity (Fig. 4–7). As harvest was delayed to intermediate stages (21–28 DAA), gradual improvements were observed in germination, shoot and root length, seedling dry weight and vigour indices, coinciding with declining moisture content and enhanced membrane stabilisation, indicating progressive physiological development.

Maximum physiological performance was attained between 35 and 42 DAA window across cultivars and years, where seeds recorded peak germination (up to 97 %), highest vigour indices, superior field emergence (82–94 %) and minimum EC values ($21.7\text{--}31.6 \mu\text{S}\cdot\text{cm}^{-1} \cdot \text{g}^{-1}$). This stage coincided with maximum dry matter accumulation and efficient reserve mobilisation, resulting in vigorous seedling establishment signifying the attainment of right stage of physiological maturity, as widely reported in cereals and millets (19, 33–37). Further, the polynomial regression analysis supported these findings, predicting the highest germination and vigour at 35 DAA in year 1 across all three cultivars (C1–C3). In year 2, however, the model indicated different optimal timings: maximum germination and vigour was predicted at 42 DAA in C1 ($R^2 = 0.83$) and C3 ($R^2 = 0.81$), while cultivar C2 reached its peak at 35 DAA ($R^2 = 0.84$) (Fig. 6). Improved field emergence at 35–42 DAA reflects enhanced reserve mobilisation, enzymatic activity and seedling growth potential (38). The strong association between germination and field emergence indicates that laboratory vigour parameters reliably predict field performance, as reported in pearl millet and other cereals (39). Across cultivars, year 2 recorded comparatively higher yields and superior physiological traits, likely due to reduced temperature stress during flowering and seed filling (Fig. 1).

In year 1, the germination and vigour declined rapidly after 35–42 DAA window due to accelerated moisture loss from high temperatures, membrane deterioration and temperature induced oxidative stress, leading to faster ageing. While in year 2, the quality remained stable up to 42 DAA and declined thereafter due to over maturity, field weathering and gradual membrane disintegration, but at a lower rate as compared to year 1. Delayed harvests (49–63 DAA) resulted in a consistent decline in germination, vigour and field emergence, accompanied by rising EC values, indicating membrane degradation and onset of seed deterioration. The reduction in physiological quality at delayed harvests is attributed to respiratory losses, oxidative damage and prolonged field exposure, which compromise membrane integrity and metabolic efficiency (40, 41). Overall, the inverse relationship between EC and germination, along with the synchronisation of maximum vigour with peak dry weight, confirms that seed physiological quality is maximised within a narrow harvest window and declines rapidly when harvesting is either premature or delayed (28, 42).

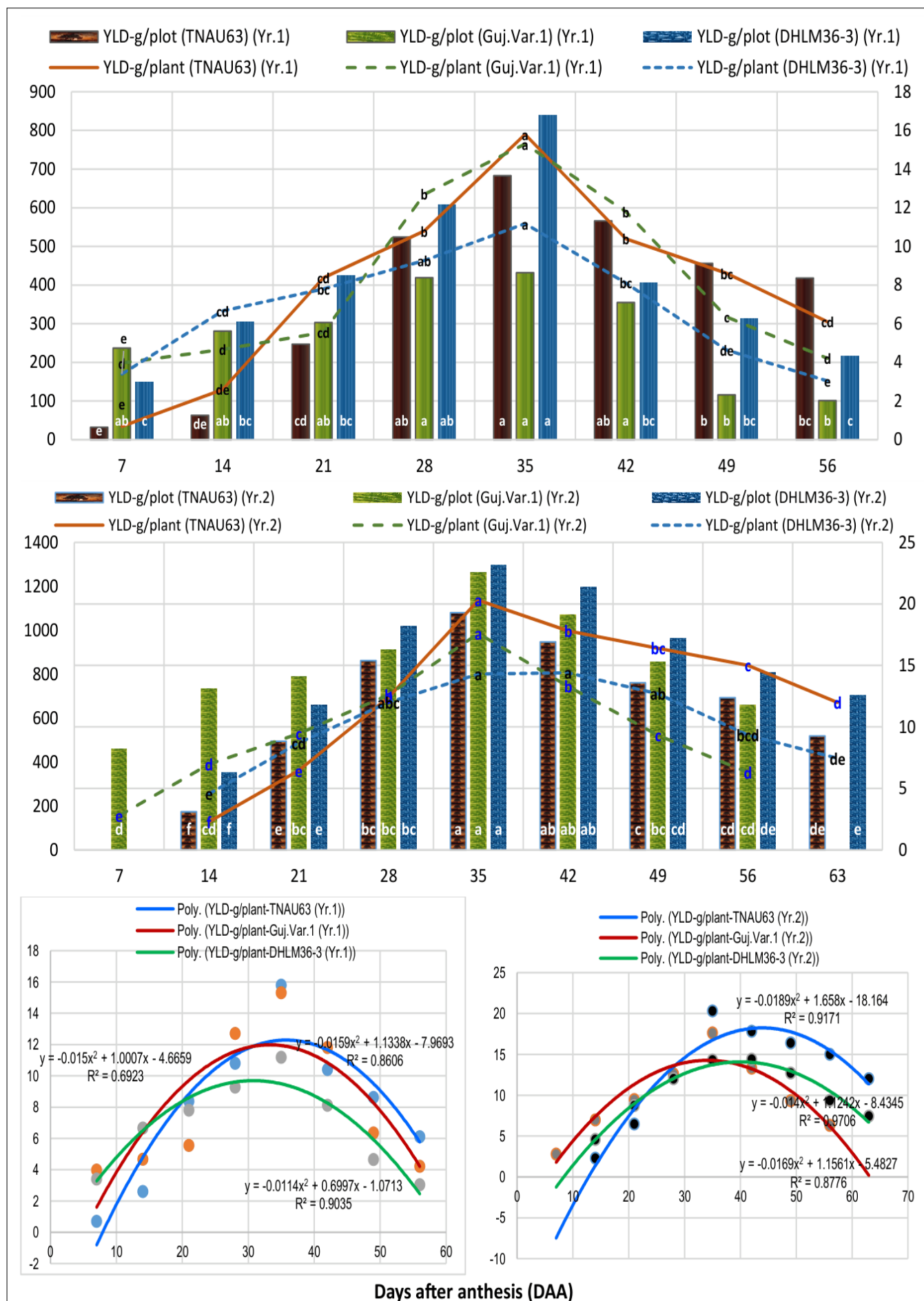


Fig. 3. Effect of different stages of seed development and maturation on yield (YLD) during year 1 (Yr.1) and year 2 (Yr.2) in three cultivars of little millet and the polynomial regression analysis for seed yield traits against days after anthesis (DAA) during year 1 (Yr.1) and year 2 (Yr. 2) in three cultivars of little millet.

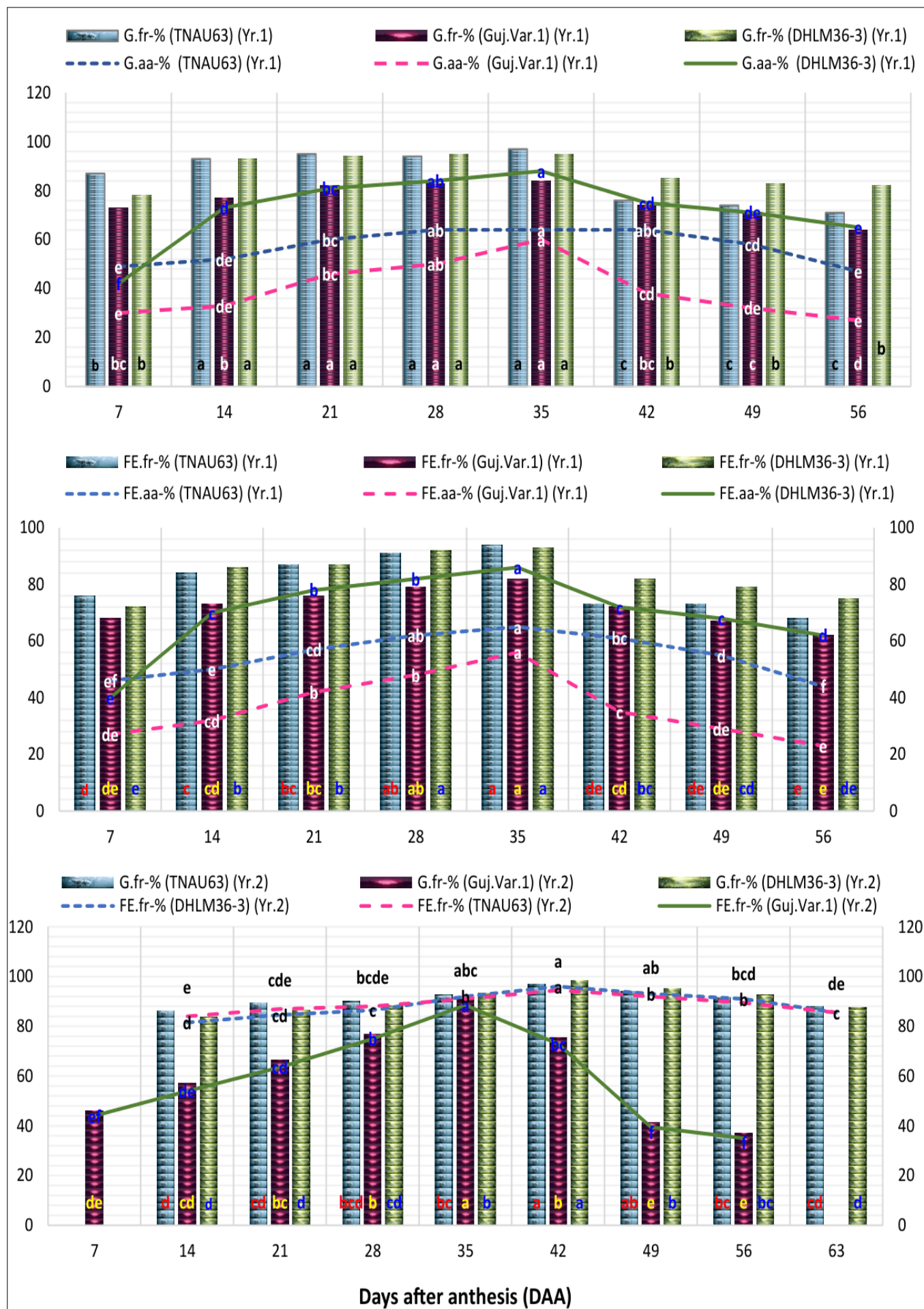


Fig. 4. Effect of different stages of seed development and maturation on germination (G), field emergence (FE) traits of fresh (fr.) and aged (aa) seeds during year 1 (Yr.1) and fresh (fr.) seeds during year 2 (Yr.2) in three cultivars of little millet.

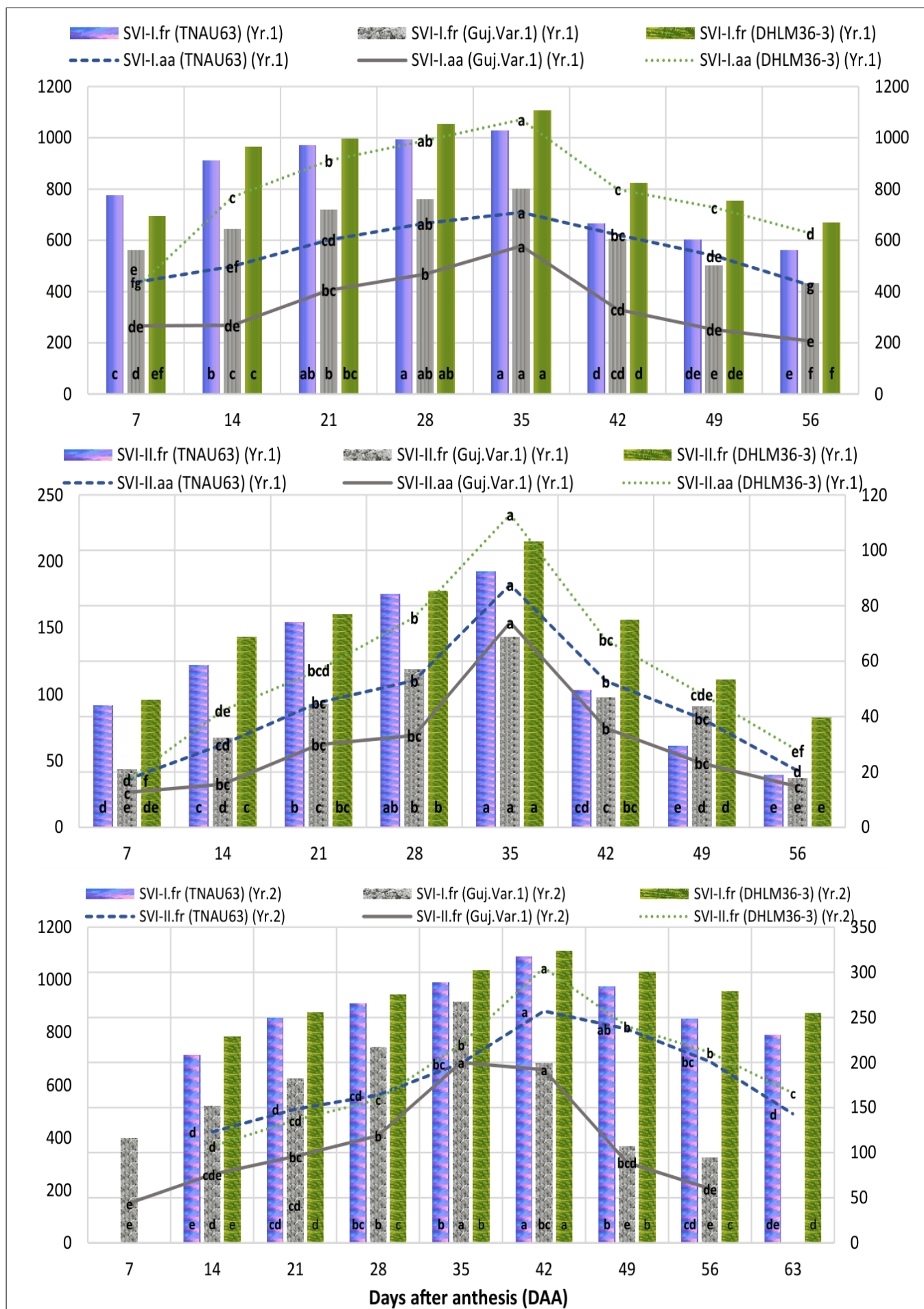


Fig. 5. Effect of different stages of seed development and maturation on seedling vigour index (SVI-I and SVI-II) traits of fresh (fr.) and aged (aa) seeds during year 1 (Yr.1) and fresh seeds during year 2 (Yr.2) in three cultivars of little millet.

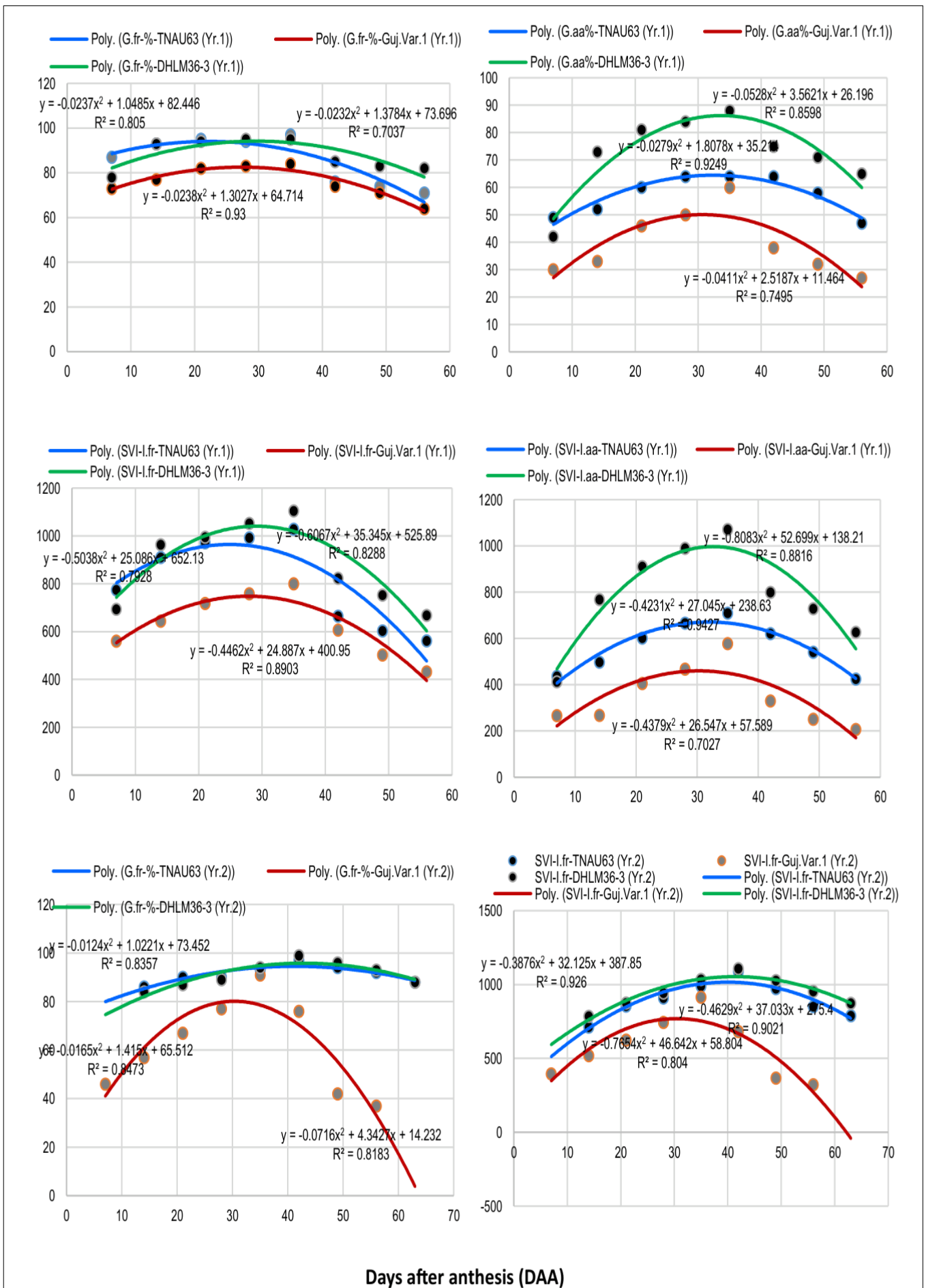


Fig. 6. Polynomial regression analysis for seed germination (G) and seedling vigour index (SVI-I) against days after anthesis (DAA) during year 1 (Yr.1) in fresh (fr.) and aged (aa) seeds and year 2 (Yr. 2) in fresh (fr.) seeds for three cultivars of little millet.

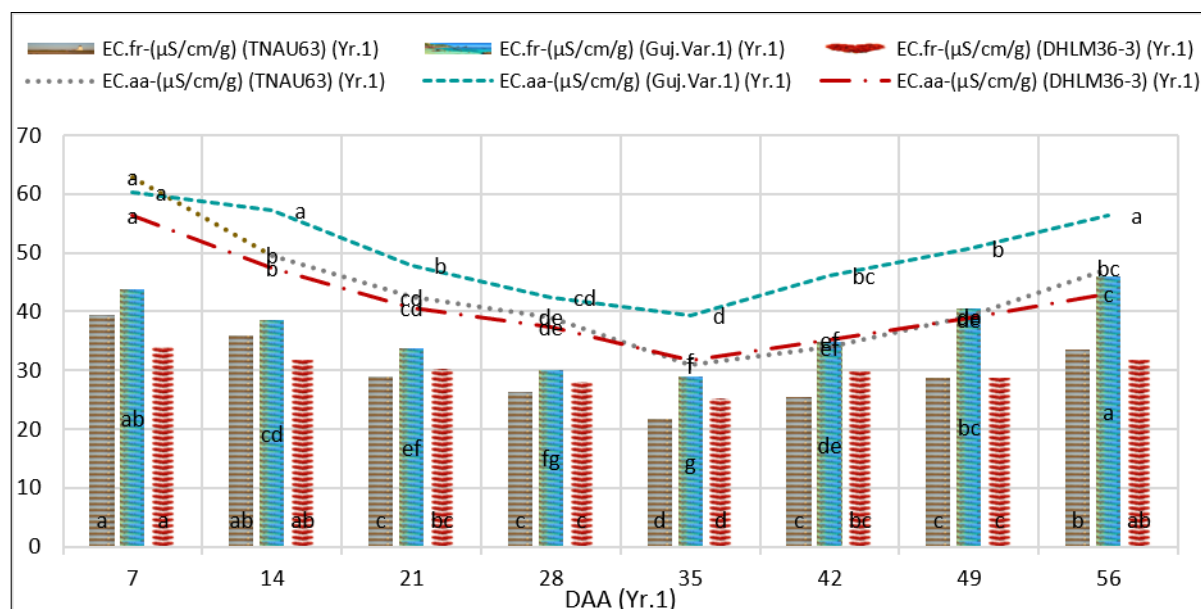


Fig. 7. Effect of different stages of seed development and maturation on electrical conductivity (EC) of fresh (fr.) and aged (aa) seeds during year 1 (Yr.1) in three cultivars of little millet.

α -amylase activity

α -amylase activity in little millet cultivar DHLM 36-3 (C3) varied prominently with HS and germination duration, reflecting the biochemical readiness of seeds for reserve mobilisation. Seeds harvested at early stages (7–14 DAA) exhibited minimal enzyme activity on the 2nd day after imbibition, consistent with incomplete development of the aleurone layer and enzyme synthesis machinery (Fig. 8). Similarly, low enzyme activity in immature seeds has been reported in foxtail and proso millet (19, 43, 44). As maturation advanced from 21–35 DAA, α -amylase activity increased significantly, reaching maximum levels on the 4th day of germination. This increase coincided with peak germination, seedling vigour and low EC values, indicating effective reserve mobilisation. The enhanced enzyme activity reflects activation of GA₃-mediated signaling pathways that induce α -amylase synthesis to hydrolyse starch into soluble sugars required for early seedling growth (45–47). Although the highest α -amylase activity

was recorded at later harvests (49–56 DAA), these seeds did not exhibit superior germination or vigour, suggesting that enzymatic potential alone cannot compensate for structural and membrane deterioration associated with degenerative changes during post-maturity. It clearly indicates that physiologically mature seeds have well-developed enzyme systems capable of rapid activation upon imbibition, ensuring efficient reserve mobilisation. Comparable trends were observed in sorghum and pearl millet, where α -amylase activity was strongly correlated with seed maturity and vigour (48–50). Across germination days, α -amylase activity increased from day 2 to day 4 and stabilised or declined slightly by day 7, likely due to substrate depletion and feedback inhibition as soluble sugars accumulated (51–53). The association between α -amylase activity and seed quality traits observed in this study corroborates earlier reports that seeds with higher enzymatic competence exhibit faster germination, better emergence and higher vigour (26, 54, 55).

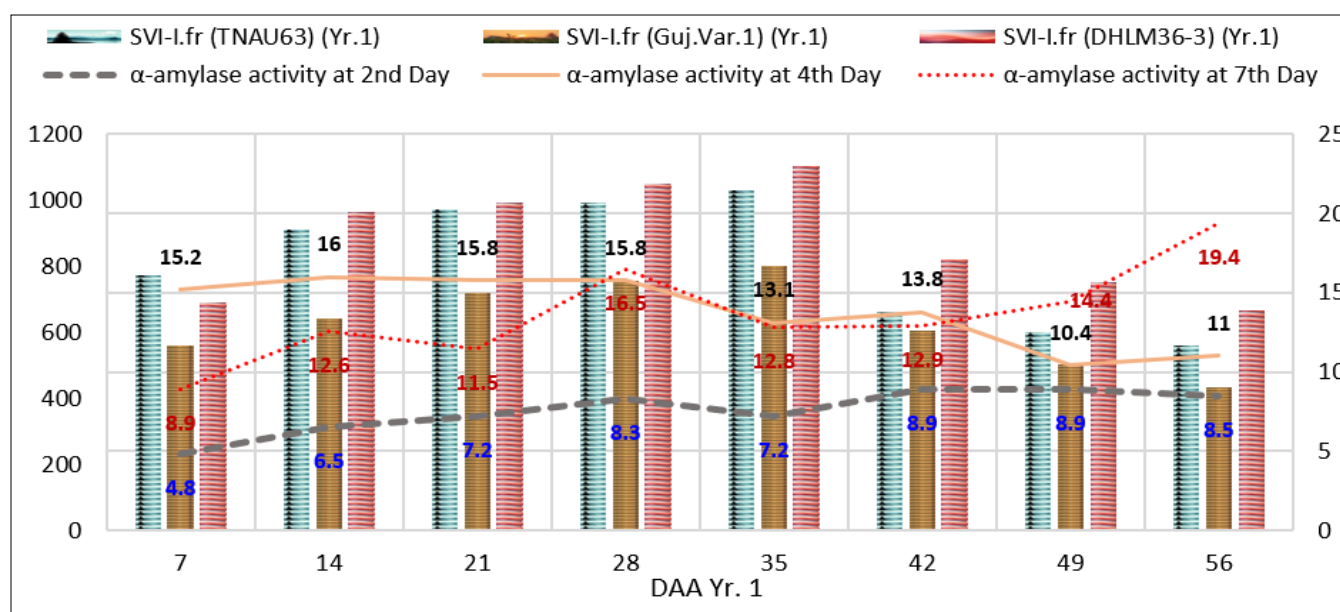


Fig. 8. Influence of different stages of harvest on α -amylase activity ($\mu\text{mol maltose} \cdot \text{min}^{-1} \cdot 100 \text{ seeds}^{-1}$) on 2nd, 4th and 7th day of seed germination, along with seedling vigour index I (SVI-I) of fresh (fr) seeds during year 1 (Yr.1) in little millet cultivar DHLM36-3.

The present study, conducted across 2 years and three cultivars of little millet, revealed distinct patterns in seed formation and maturation. In year 1, all cultivars initiated seed formation uniformly at 7 DAA. In contrast, year 2 showed variability: Gujarat vari 1 recorded seed formation at 7 DAA, whereas TNAU 63 and DHLM 36-3 were delayed until 14 DAA. Seed moisture content declined progressively with maturity, while seed test weight, hardness, germination, vigour indices, field emergence, α -amylase activity and storability increased up to a critical stage. At full maturity, seeds exhibited optimal moisture balance, maximum dry matter content and high germination capacity (19, 23). In terms of morphological indices, the seeds develop desiccation tolerance, accompanied by a colour shift of little millet panicles from green to dark brown. While the seeds with oval smooth and shiny body, show a streaked green to yellow brown colour. More importantly de-husked seeds exhibited a distinct brownish depression at the hilar end (Fig. 2). Similar morphological indices for physiological maturity were also recorded in other small millets (19, 36, 37). In year 1, physiological maturity was attained at 35 DAA, with all cultivars recording significantly higher seed yield and quality traits. In year 2, however, Gujarat vari 1 reached peak seed quality at 35 DAA, while TNAU 63 and DHLM 36-3 attained it at 42 DAA. Early harvests produced immature, low-vigour seeds, whereas delayed harvests resulted in deterioration and reduced seed quality (56). This deterioration occurred as seeds remained exposed in the field, where adverse summer weather conditions accelerated damage and loss. Thus, harvesting at the right time is essential for producing high-quality seed with superior field performance and storage potential (57). Similar patterns of seed development and quality enhancement at physiological maturity have been reported in other pearl millet and other small millets (19, 36, 58, 59).

Overall, physiological maturity in little millet ranged between 35–42 DAA depending on cultivar and year, establishing this interval is suitable for the harvestable maturity window as well. At this window of physiological and harvestable maturity, seed harvesting and handling characteristics are optimal to minimise field losses of seeds. However, in practice, the timing of harvestable maturity varies across genotypes and under varying climatic conditions.

Conclusion

The study demonstrates that the stage of harvest influences seed yield, seed physiological quality performance and biochemical quality attributes in little millet. Physiological maturity, indicated by peak seed quality traits, occurred at 35 DAA across cultivars in year 1, but in year 2 at 35 DAA for Gujarat vari 1 and 42 DAA for TNAU 63 and DHLM 36-3 due to temperature-driven variation. The findings establish 35–42 DAA as the optimal window for physiological and harvestable maturity in little millet. Polynomial regression analysis further validated this maturity window. Early harvests produced immature, low-vigour seeds, whereas delayed harvests resulted in seed shattering and quality deterioration. At physiological maturity, little millet panicles turn straw/dark brown and seeds are oval, smooth, shiny and streaked from greenish brown to yellow. By defining a precise maturity window, this research provides a practical basis for improving seed production, quality assurance and breeding programs. The identified harvest window serves as a reliable guideline for seed producers to schedule harvesting for maximising seed recovery, vigour, storability and planting value

while minimising losses due to shattering and field weathering. Future research should focus on validating these maturity indicators across a wider range of agro-ecological zones and genetic base and on exploring the physiological and molecular mechanisms underlying cultivar-specific and environment-induced variation in seed maturity and quality acquisition.

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Authors' contributions

KN, GKN and VR conceptualised, designed and conducted experiments. KN, DIK and NM performed data analysis and interpretation. KN, NM, MVM, SBC and TSC prepared the manuscript. KN, GKN, NM, VR, MVM, SBC, DIK and TSC reviewed and approved the final manuscript for submission. All authors have read and approved the manuscript.

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During the process of manuscript writing, the authors used Microsoft Copilot and ChatGPT as language-assisting tools to improve grammar, paraphrase sentences, and enhance readability. After using these tools, the authors reviewed and edited the content as needed, and they own the research findings and related scientific content of the publication.

References

- de Wet JM, Brink DE, Rao KP, Mengesha MH. Diversity in kodo millet, *Paspalum scrobiculatum*. *Econ Bot.* 1983;37(2):159–63. <https://doi.org/10.1007/BF02858779>
- Ganapathy KN. Genetic improvement in little millet. In: Millets and sorghum: biology and genetic improvement. 2017. p. 170–83. <https://doi.org/10.1002/9781119130765.ch6>
- Prasada Rao KE, de Wet JM, Gopal Reddy V, Mengesha MH. Diversity in the small millets collection at ICRISAT. In: Riley KW, Gupta SC, Seetharam A, Mushonga JN, editors. *Advances in small millets*. New Delhi: Oxford & IBH Publishing Co. Pvt. Ltd.; 1993.
- Mohan MM, Vijayakumar A, Bang-Berthelsen CH, Mudnakudu-Nagaraju KK, Shetty R. Millets: journey from an ancient crop to sustainable and healthy food. *Foods*. 2025;14(10):1733. <https://doi.org/10.3390/foods14101733>
- Saha D, Gowda MC, Arya L, Verma M, Bansal KC. Genetic and genomic resources of small millets. *Crit Rev Plant Sci.* 2016;35(1):56–79. <https://doi.org/10.1080/07352689.2016.1147907>
- Nagaraja TE, Nandini C, Bhat S, Parveen SG. Artificial hybridization

- techniques in small millets: A review. *Front Plant Sci.* 2023;14:1112117. <https://doi.org/10.3389/fpls.2023.1112117>
7. Dwivedi SL, Upadhyaya HD, Senthilvel S, Hash CT, Fukunaga K, Diao X, et al. Millets: genetic and genomic resources. 2012. <https://doi.org/10.1002/9781118100509.ch5>
 8. Matere BS, Bhavsar VV, Mate PA, Girase VS. Genetic variability of yield and its components in little millet (*Panicum sumatrense* L.) local germplasm. *J Agric Res Technol.* 2022;47:258–63. <https://doi.org/10.56228/JART.2022.47303>
 9. Arunachalam V, Rengalakshmi R, Kubera Raj MS. Ecological stability of genetic diversity among landraces of little millet (*Panicum sumatrense*) in South India. *Genet Resour Crop Evol.* 2005;52(1):15–9. <https://doi.org/10.1007/s10722-005-6693-4>
 10. Patil KB, Chimmad BV, Itagi S. Glycemic index and quality evaluation of little millet (*Panicum miliare*) flakes with enhanced shelf life. *J Food Sci Technol.* 2015;52(9):6078–82. <https://doi.org/10.1007/s13197-014-1663-5>
 11. Indirani K. Review on nutritional profiles and health benefits of little millets–India. *Int J Res Eng Sci.* 2021;9(11):7–11.
 12. Vetriventhan M, Azevedo VC, Upadhyaya HD, Nirmalakumari A, Kane-Potaka J, Anitha S, et al. Genetic and genomic resources and breeding for accelerating improvement of small millets: current status and future interventions. *The Nucleus.* 2020;63(3):217–39. <https://doi.org/10.1007/s13237-020-00322-3>
 13. Asharani VT, Jayadeep A, Malleshi NG. Natural antioxidants in edible flours of selected small millets. *Int J Food Prop.* 2010;13(1):41–50. <https://doi.org/10.1080/10942910802163105>
 14. Nambi VA, Eganathan P, Philip PM. Proximate and mineral composition analysis of little millet collected from some millet growing areas in Tamil Nadu. *Indian J Plant Genet Resour.* 2012;25(2):189–91.
 15. Pasha VK, Ratnavathi CV, Ajani J, Raju D, Manoj Kumar S, Beedu SR. Proximate, mineral composition and antioxidant activity of traditional small millets cultivated and consumed in Rayalaseema region of South India. *J Sci Food Agric.* 2018;98(2):652–60. <https://doi.org/10.1002/jsfa.8510>
 16. Panda D, Palita SK. Potential of underutilized wild crops in Koraput, Odisha, India for improving nutritional security and promoting climate resilience. *Curr Sci.* 2021;120(6):989–96. <https://doi.org/10.18520/cs/v120/i6/989-996>
 17. Ekpong B, Sukprakarn S. Seed physiological maturity in dill (*Anethum graveolens* L.). *Agric Nat Res.* 2008;42(5):1–6.
 18. Pollock BM, Roos EE. Seed and seedling vigor. In: Kozlowski TT, editor. *Seed biology*. Vol. 1. Importance, development and germination. 1972. p. 314–87. <https://doi.org/10.1016/B978-0-12-424301-9.50012-7>
 19. Kannababu N, Hariprasanna K, Venkateswarlu R, Sangappa C, Das IK, Tara Satyavathi C. Determining physiological maturity for optimal seed quality and reduced yield loss in foxtail millet (*Setaria italica* (L.) P. Beauv.). *Plant Sci Today.* 2025;12(4). <https://doi.org/10.14719/pst.11195>
 20. International Seed Testing Association. International rules for seed testing. Bassersdorf: ISTA; 2015.
 21. Delouche JC, Baskin CC. Accelerated aging techniques for predicting the relative storability of seed lots. *Seed Sci Technol.* 1973;1:427–52.
 22. Bernfeld P. Amylases, α and β . In: Colowick SP, Kaplan NO, editors. *Methods in Enzymology*. New York: Academic Press; 1955. p. 149–58. [https://doi.org/10.1016/0076-6879\(55\)01021-5](https://doi.org/10.1016/0076-6879(55)01021-5)
 23. Marcos-Filho J. Fisiologia de sementes de plantas cultivadas. 2nd ed. Londrina: Abrates; 2015. p. 659.
 24. Leprince O, Pellizzaro A, Berriri S, Buitink J. Late seed maturation: drying without dying. *J Exp Bot.* 2017;68(4):827–41. <https://doi.org/10.1093/jxb/erw363>
 25. Kermode AR. Regulatory mechanisms involved in the transition from seed development to germination. *Crit Rev Plant Sci.* 1990;9(2):155–95. <https://doi.org/10.1080/07352689009382286>
 26. Bewley JD, Bradford KJ, Hilhorst HWM. *Seeds: physiology of development, germination and dormancy*. New York: Springer; 2012. <https://doi.org/10.1007/978-1-4614-4693-4>
 27. Yadav VP, Yadav S, Yadav P, Yadav AK. Increase efficiency in millets agrarian and nutritional food from bio-fortified hybrids: A review. *Plant Arch.* 2025;25(2):1535–44. <https://doi.org/10.51470/PLANTARCHIVES.2025.v25.no.2.223>
 28. Groot SPC. Seed maturation and its practical implications. *Seed Sci Technol.* 2022;50(2):141–51. <https://doi.org/10.15258/sst.2022.50.1.s.08>
 29. Reddy BV, Ramesh S, Reddy PS. Sorghum breeding research at ICRIAT: goals, strategies, methods and accomplishments. *Int Sorghum Millets Newsl.* 2004;45:5–12.
 30. Tonapi VA, Varnavasiappan S, Navi SS, Ravinder Reddy C, Karivatharaju TV. Effect of environmental factors during seed development and maturation on seed quality in *Sorghum bicolor* (L.) Moench. *Plant Arch.* 2006;6(2):515–9.
 31. Djanaguiraman M, Priyanka AS, Vaishnavi SJ, Perumal R, Ciampitti IA, Prasad PV. Impact of drought and high-temperature stresses on growth and development stages, physiological, reproductive and yield traits in pearl millet. In: *Pearl millet: a resilient cereal crop for food, nutrition and climate security*. 2024. p. 249–76. <https://doi.org/10.1002/9780891184034.ch9>
 32. Ellis RH, Pieta Filho C. The development of seed quality in spring and winter cultivars of barley and wheat. *Seed Sci Res.* 1992;2(1):9–15. <https://doi.org/10.1017/S0960258500001057>
 33. Kannababu N, Amasiddha B, Venkateswarlu R, Das IK, Prabhakar B, Tonapi VA. Determination of physiological and harvestable mass maturity of seeds during rainy and post-rainy seasons in barnyard millet (*Echinochloa frumentacea* (Roxb.). *Range Manag Agrofor.* 2023;44(1):84–94. <https://doi.org/10.59515/rma.2023.v44.i1.10>
 34. Copeland LO, McDonald MB. *Principles of seed science and technology*. New York: Springer; 2012.
 35. Lainufar PA, Qadir A, Suhartanto MR, Ardie SW. Harvesting criteria and drying methods to improve the quality of foxtail millet seeds (*Setaria italica* (L.) P. Beauv.). *J Trop Crop Sci.* 2021;8(3). <https://doi.org/10.29244/jtcs.8.03.168-176>
 36. Kannababu N, Ganapathy KN, Venkateswarlu R, Tara Satyavathi C. Physiological and harvestable maturity of seeds in finger millet (*Eleusine coracana* (L.) Gaertn.). *Range Manag Agrofor.* 2024;45(2):290–9. <https://doi.org/10.59515/rma.2024.v45.i2.14>
 37. Kannababu N, Deepika C, Venkateswarlu R, Hariprasanna K, Das IK. Physiological maturity of seeds in kodo millet (*Paspalum scrobiculatum* L.) cultivars across seasons. *Plant Physiol Rep.* 2023;28(2):209–20. <https://doi.org/10.1007/s40502-023-00725-9>
 38. Gough RE. *Seed quality: basic mechanisms and agricultural implications*. Boca Raton: CRC Press; 2020. <https://doi.org/10.4324/9781003075226>
 39. Mahanthesh M, Pahuja SK, Puneeth Raj MS, Devvart, Mor VS, Santosh Korav. Seed quality parameters assessment in pearl millet (*Pennisetum glaucum*). *Pharma Innov J.* 2022;11(7):2769–72.
 40. Matthews S, Powell AA. Electrical conductivity vigour test: physiological basis and use. *Seed Test Int.* 2006;131:32–35.
 41. Sharma P, Sardana V, Kandhola SS. Effect of sowing dates and harvesting dates on germination and seedling vigor of groundnut (*Arachis hypogaea*) cultivars. *Res J Seed Sci.* 2013;6(1):1–5. <https://doi.org/10.3923/rjss.2013.1.15>
 42. Senthil Raj R, Ahamed AS, Sujatha K, Ragupathi KP. Biochemical

- changes during seed development and maturation in little millet cv. CO (Samai) 4. *J Pharmacogn Phytochem*. 2021;10(1):703–6. <https://doi.org/10.22271/phyto.2021.v10.i1j.13406>
43. Ravindran P, Kumar PP. Regulation of seed germination: the involvement of multiple forces exerted via gibberellic acid signaling. *Mol Plant*. 2019;12(1):24–6. <https://doi.org/10.1016/j.molp.2018.12.013>
 44. Dutta P, Dey P, Bhowmik A. Changes in α -amylase activity and carbohydrate metabolism during seed germination in small millets. *Seed Res*. 2021;49(2):103–10.
 45. Fincher GB. Molecular and cellular biology associated with endosperm mobilization in germinating cereal grains. *Annu Rev Plant Physiol Plant Mol Biol*. 1989;40:305–46. <https://doi.org/10.1146/annurev.pp.40.060189.001513>
 46. Yamaguchi S. Gibberellin metabolism and its regulation. *Annu Rev Plant Biol*. 2008;59:225–51. <https://doi.org/10.1146/annurev.arplant.59.032607.092804>
 47. Nonogaki H, Bassel GW, Bewley JD. Germination—still a mystery. *Plant Sci*. 2010;179(6):574–81. <https://doi.org/10.1016/j.plantsci.2010.02.010>
 48. Varier A, Vijayakumar KR, Dadlani M. Changes in α -amylase and protease activities during germination and early seedling growth in maize and rice. *Indian J Plant Physiol*. 2010;15(2):164–70.
 49. Srivastava P, Pandey R, Kumar R. Relationship between α -amylase activity, seed vigor and germination in sorghum under varying maturity levels. *Seed Sci Technol*. 2018;46(1):133–42.
 50. Krishnan R, Shankaraiah V, Rani M. Seed maturity and α -amylase dynamics during germination in pearl millet hybrids. *Indian J Plant Physiol*. 2022;27(2):332–40.
 51. Sasaki T, Matsumoto T, Itoh K. Regulation of hydrolytic enzyme expression during cereal seed germination. *Plant Mol Biol*. 2005;59(3):463–74.
 52. Taiz L, Zeiger E, Møller IM, Murphy A. *Plant physiology and development*. 6th ed. Sunderland: Sinauer Associates; 2015.
 53. Liu Y, Hu Y, Han C. Regulation of α -amylase activity and starch degradation during seed germination: recent advances. *Plant Physiol Rep*. 2019;24(4):465–76. <https://doi.org/10.1104/pp.18.01533>
 54. Basra AS, editor. *Handbook of seed science and technology*. New York: Haworth Press; 2006.
 55. Rajendran K, Vignesh R. Seed maturity and its relation to germination and vigor in small millets. *Seed Res*. 2021;49(1):11–17.
 56. Somrug K, Timsuksai P, Jenweerawat S. Influence of harvesting date on yield and seed quality of indigo (*Indigofera tinctoria* L.). *King Mongkut's Agric J*. 2020;38(4):426–33.
 57. Aswin C, Geetha R, Sujatha K, Vanniarajan C, Sivakumar T. Effect of harvesting time on the seed quality of hybrid maize (*Zea mays* L.) COHM 8. *Agric Sci Dig*. 2023;43(5). <https://doi.org/10.18805/ag.D-5748>
 58. Ramakrishnan V, Nirmalakumari A, Mohanasundaram T. Seed quality development in finger millet. *J Cereal Res*. 2017;9(1):56–62. <https://doi.org/10.1080/15427528.2019.1709596>
 59. Yadav OP, Singh DV, Gupta SC. Seed maturity and quality in pearl millet. *Indian J Agric Sci*. 2011;81(8):734–9.

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