



RESEARCH ARTICLE

Grain yield and quality attributes of wheat (*Triticum aestivum* L.) as influenced by heat stress during the reproductive phase due to delayed sowing

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Abstract

Terminal heat stress during the reproductive stage of wheat is one of the limiting factors affecting both production and grain quality and it is expected to gradually increase under the current climate scenario. To investigate this issue, a field experiment was conducted to assess the performance of wheat varieties for grain yield and qualitative traits. Seeds were sown under two conditions: normal (November 20) and late (December 20), allowing for a comparative study of heat stress during the later stages of plant growth. The results showed that grain weight per spike and test weight progressively decreased due to high temperatures experienced during the grain-filling period, which was associated with the delayed sowing date. High temperatures during the later stages of crop growth, particularly in the reproductive phase, adversely affect both the rate and duration of grain development. This results in forced maturity, leading to shriveled, immature and small grains with poor quality parameters, including reduced protein, starch, gluten content and sedimentation value.

Keywords: grain yield; gluten; heat stress; protein; sedimentation value; starch; wheat

Introduction

Wheat (*Triticum aestivum* L.), consumed in various forms, is the most commonly grown crop in the world. In India, it is a crucial part of the diet and normally contains more protein than rice. The highest quality wheat is grown in regions with cool and moist weather during the early growing period, followed by relatively dry and slightly warm conditions to ensure proper grain ripening. However, excessively high temperatures during the grain-filling stages can accelerate the maturity of the grain. For optimal grain development, wheat requires a mean maximum temperature of about 25 °C for at least four to five weeks (30). When wheat plants are exposed to high temperatures (greater than 32 °C), this can lead to forced maturity of the grain during ripening, resulting in altered agronomic characteristics and grain quality. High temperatures shorten the duration of all developmental stages in wheat (1), thereby reducing the length of the grain-filling period. High temperatures also influence kernel weight, which can further reduce grain yield (2). When temperatures exceed the optimal range for plant growth, excessive energy is lost through transpiration. This loss of residual energy negatively impacts grain formation, ultimately resulting in lower grain yields and reduced quality parameters. Heat stress during the grain-filling stage, even for just a few days, can significantly affect protein composition. High temperatures accelerate plant metabolism,

which shortens the grain-filling period, leading to reduced starch accumulation (7). Meanwhile, protein synthesis is affected or may even decrease under heat stress, resulting in a lower protein concentration and often of lower functional quality (3). Additionally, heat stress may impact the synthesis, accumulation and assembly of gluten proteins (6). Very high temperatures, such as brief episodes exceeding 35 °C, can result in grain with weaker-than-expected dough properties (3, 5). This reduction in strength may be attributed to heat-induced alterations in the glutenin-to-gliadin ratio, as heat stress has been shown to decrease this ratio, thereby affecting the functional quality of gluten (4, 5). This change can occur due to an increased rate of monomeric protein synthesis or a reduction in glutenin synthesis (8). Seasonal variations in grain quality pose challenge for marketing and processing (9), making quality stability a crucial goal (5). Some genotypes exhibit either minimal changes or an increase in the glutenin-to-gliadin ratio (10). Such varieties are likely to maintain consistent dough properties and may serve as genetic resources for enhancing heat tolerance (15). This research aimed to evaluate the changes in grain characteristics and yield caused by heat stress during the grain-filling period in selected wheat varieties. The findings will improve understanding of how heat stress affects processing quality, aiding in the identification of heat-tolerant wheat genotypes that exhibit stable and consistent industrial quality.

Materials and Methods

Field locations

A field experiment was done for 2 consecutive years, 2021-2022, during the '*rab*' season at the Norman E. Borlaug Crop Research Centre, located within the Govind Ballabh Pant University of Agriculture and Technology in Pantnagar (U.S. Nagar), Uttarakhand, India. This centre is positioned at a latitude of 29° 03' N and a longitude of 79° 30' E, with an altitude of 243.83 m above mean sea level. It falls within the humid subtropical zone of the Shivalik range foothills of the Himalayas (20). The experimental site features silt loam soil and a chemical analysis indicated that it is rich in organic matter, has high levels of available phosphorus, medium levels of available potassium and a neutral pH (Table 1). The weather parameters recorded at the Meteorological Observatory at the Centre during the experimental period included minimum and maximum temperatures, relative humidity and rainfall (21, 22). A total of 155.2 mm of rainfall was observed during this time. The weekly mean maximum temperatures ranged from 17.6 °C to 37.1 °C, while the mean minimum temperatures varied from 3.9 °C to 20.4 °C. Additionally, the mean weekly relative humidity ranged from 62 % - 96 % at 7 AM and from 18 % to 64 % at 2 PM.

Table 1. Chemical properties of the experimental plot soil (0-15 cm depth)

Particulars	Values obtained	Method employed
Organic carbon (%)	0.69	Walkley and Black's method (23)
Av. Phosphorus (kg P ha ⁻¹)	25.79	Olsen's method (24)
Av. Potassium (kg K ha ⁻¹)	135.1	Flame Emission spectrophotometry (24)
pH (1:2.5 soil water suspension)	7.1	Beckman glass electrode pH meter (24)

Treatments and experimental design

The experiment was conducted using a split-plot design with three replications. There were two dates of sowing: D1 on November 20 (normal sowing) and D2 on December 20 (late sowing), which were considered as the main plot treatments. Additionally, ten bread wheat varieties based on popularity, heat tolerance, etc. were included as subplot treatments: V1 - PBW 343; V2 - PBW 502; V3 - UP 2425; V4 - UP 1109; V5 - UP 2565; V6 - PBW 373; V7 - UP 2338; V8 - UP 2382; V9 - UP 262 and V10 - Raj 3765. Seed was sown at a rate of 100 kg/ha with a row spacing of 22.5 cm. A recommended fertilizer dose of nitrogen and phosphorus at 120 kg/ha and 60 kg/ha respectively, was applied. The sources of fertilizer used were urea (46 % nitrogen) and di-ammonium phosphate (46 % phosphorus and 18 % nitrogen). All of the phosphorus and half of the nitrogen were applied as a basal dose, while the remaining nitrogen was applied at the crown root initiation (CRI) stage. At maturity, the crop was harvested and the plants were sun-dried for a week. Random samples of plants were selected from each plot to assess yield parameters. After sun-drying, the plants from each plot were threshed and the grains were separated and packed into jute bags. The grain material obtained from the individual plots through field experimentation was then used for laboratory studies.

Grain quality analysis

All quality parameters, including grain protein, starch, wet gluten and Zeleny content (cc), were estimated using the Infratech™ 1241 Grain Analyzer (9). Samples of grain were taken from each replicate. This analyser measures parameters in whole grains like wheat, rice,

corn, soybeans and oats quickly and efficiently without reagents or preparation. Samples are placed in a hopper, transported to a sample cell and a monochromatic light beam is transmitted to the detector. Results are displayed on an LCD screen.

Statistical analysis

The experimental data were analysed by following standard statistical procedure for split-plot design (11). The standard error of means and critical differences was computed at 5 % level of probability for significant differences only.

Results

Yield and its attributes

Grain weight per spike

Grain weight per spike was significantly influenced by heat stress at the reproductive stage due to delay in sowing among the varieties and their interaction was also significant in this regard. A review of Table 2 clearly indicates that the normally sown crop (November 20) had a higher grain weight per spike (2.02 g) compared to the late sown crop (December 20), which had a grain weight of 1.81 g. The variety UP 2565 recorded the highest grain weight per spike at 2.27 g, which was statistically comparable to UP 2425 and UP 2382. Shifting of sowing date for thirty days significantly reduced the grain weight per spike in UP 2382, UP 2338, UP 262 and UP 2565 (Table 3). Whereas other varieties produced statistically similar grain weight per spike at both dates of sowing. In normal sown condition, UP 2565 (2.51 g) exhibited significantly higher grain weight per spike than UP 2425, but under late sown condition, both these varieties had statistically the same grain weight per spike (Fig. 1A).

Test weight (1000 grain weight)

The test weight was significantly influenced by changes in sowing dates and the varieties used. The crops sown at the normal time had a significantly higher thousand grain weight (40.4 g) compared to those sown late (37.5 g). Table 2 shows the variety UP 262 had the highest test weight at 42.6 g, which was significantly greater than that of the other varieties, although it was statistically similar to UP 2425. Furthermore, varieties UP 2382, UP 2338, Raj 3765 and UP 2565 showed a significant reduction in thousand grain weight when sowing was delayed by

Table 2. Grain weight per spike (g) and test weight (g) and grain yield of wheat varieties influenced by heat stress at the reproductive stage due to delay in sowing (pooled data of 2 years)

Treatments	Grain weight per spike	Thousand grain weight	Grain yield
Sowing dates			
November 28	2.02	40.4	42.9
December 20	1.81	37.5	35.7
SEm ±	0.02	0.19	0.47
CD (5 %)	0.09	1.1	2.8
Varieties			
PBW 343	1.77	37.7	42.0
PBW 502	1.66	37.1	38.3
UP 1109	1.68	36.6	34.9
UP 262	1.84	42.6	40.8
UP 2338	1.95	37.0	40.1
PBW 373	1.80	38.7	40.9
Raj 3765	1.89	40.3	38.0
UP 2382	2.14	38.7	40.0
UP 2425	2.14	41.7	38.5
UP 2565	2.27	39.2	39.6
SEm ±	0.06	0.63	1.23
CD (5 %)	0.18	1.8	3.5

Table 3. The statistical differences in yield attributes viz., grain weight per spike (g) and thousand grain test weight (g) of wheat varieties as influenced by sowing dates (pooled data of 2 years)

Treatments	Grain wt per spike		1000 grain weight	
	Nov. 28	Dec. 20	Nov. 28	Dec. 20
Varieties				
PBW 343	1.78	1.76	38.6	36.7
PBW 502	1.67	1.65	37.6	36.6
UP 1109	1.69	1.67	37.3	35.9
UP 262	2.01	1.66	43.8	41.3
UP 2338	2.22	1.69	40.8	33.2
PBW 373	1.81	1.78	39.3	38.1
Raj 3765	1.93	1.86	41.8	38.7
UP 2382	2.40	1.87	40.8	36.6
UP 2425	2.17	2.11	42.6	40.8
UP 2565	2.51	2.03	41.2	37.2
	SEm ±	CD (5 %)	SEm ±	CD (5 %)
For comparing two varieties at the same date	0.09	0.25	0.89	2.5
For comparing two dates at same or different varieties	0.08	0.25	0.86	2.6

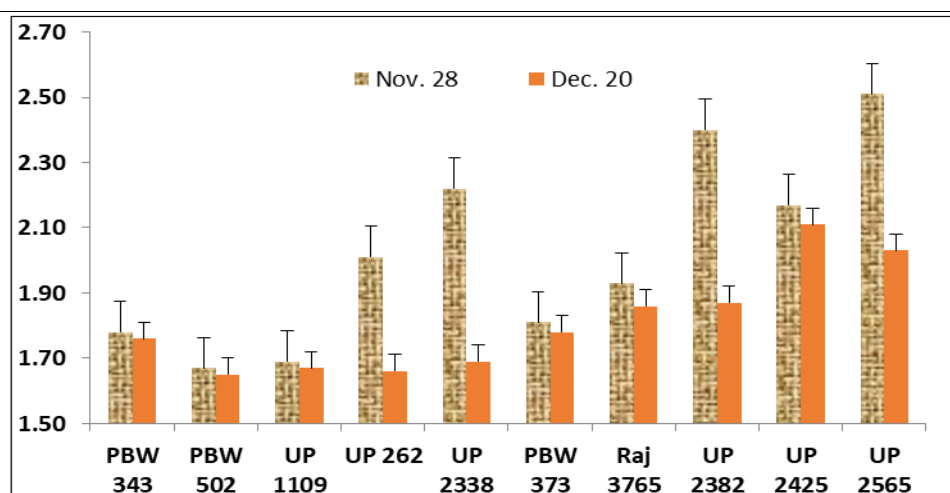


Fig. 1A. 1000 Grain wt (g) wheat varieties influenced by heat stress at reproductive stage.

thirty days (Table 3). Notably, variety UP 2338, which produced significantly bold grains compared to PBW 502 and UP 1109 under timely sown conditions, showed significantly lower thousand grain weight than all other varieties under late sown conditions (Fig. 1B).

Grain yield

Grain yield was significantly affected by changes in sowing dates and varieties; however, there was no interaction between these two factors for this parameter. A review of Table 2 clearly indicates that the normally sown crop yielded significantly more (42.9 qha^{-1}) than the late-sown crop. Among the varieties tested, PBW 343 produced the highest grain yield (42.0 qha^{-1}).

Protein

The study examined the impact of different wheat varieties and their interaction with varying sowing dates on the protein content of the grain. The results explore the effects of both sowing dates and wheat varieties on several factors, including grain yield, protein content, starch and gluten content and sedimentation value (measured in cubic centimetres, or cc) based on pooled data from two years. The variety UP 2338 showed the highest protein content at 12.78 %, which was comparable to UP 2565 and UP 262 (Table 4). Additionally, UP 2425, PBW 343, Raj 3765 and PBW 373 exhibited significantly higher protein levels. Statistical variations in grain quality traits-including protein content, starch levels, gluten

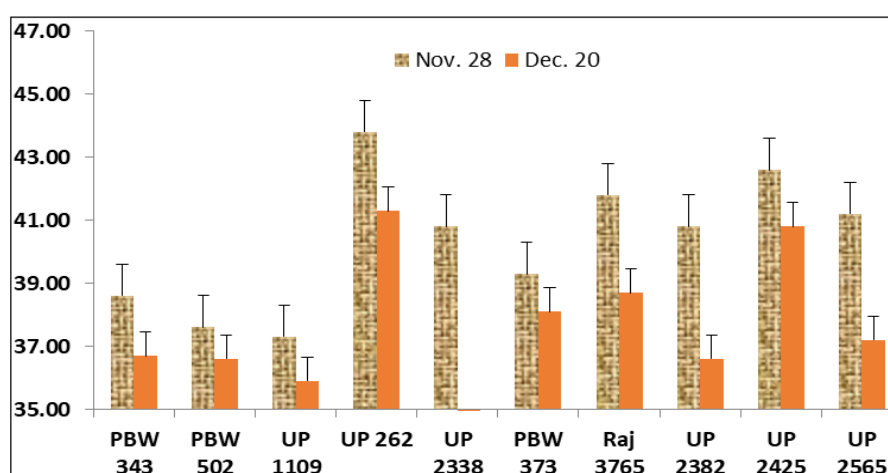


Fig. 1B. 1000 grain wt (g) of wheat varieties influenced by heat stress at reproductive stage due to delay in sowing.

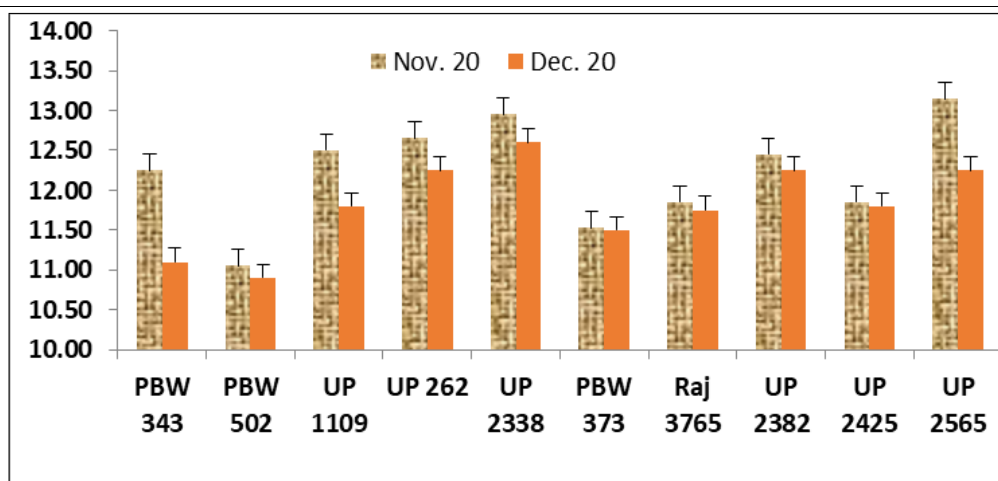
Table 4. Effect of sowing dates and varieties on protein, starch and gluten content and sedimentation value (cc) of wheat (pooled data of 2 years)

Treatments	Protein	Starch	Gluten	Sedimentation value
Sowing date				
November 20	12.11	63.8	32.2	43.0
December 20	11.82	61.8	31.2	40.0
SEm±	0.21	0.20	0.47	1.38
CD (5 %)	NS	NS	NS	NS
Varieties				
PBW 343	11.68	64.2	31.0	40.4
PBW 502	10.98	64.9	29.0	34.2
UP 1109	12.15	63.2	31.8	42.6
UP 262	12.45	63.2	32.9	44.4
UP 2338	12.78	62.3	33.9	46.8
PBW 373	11.52	64.7	30.6	37.9
Raj 3765	11.55	64.0	30.9	40.7
UP 2382	12.20	63.8	32.4	44.4
UP 2425	11.68	64.1	30.7	37.7
UP 2565	12.70	62.9	30.9	45.7
SEm±	0.14	0.14	0.37	1.03
CD (5 %)	0.41	0.4	1.1	2.9

and sedimentation value-among different wheat varieties were evaluated across multiple sowing dates, using pooled data collected over 2 years. The delay in sowing significantly reduced the protein content in the grain of PBW 343 (Table 5). In contrast, other varieties did not show any significant changes in protein content due to late sowing. Some varieties, such as Raj 3765, UP 2382 and UP 2425, actually showed an increase in protein content under late sowing conditions (Fig. 2A).

Table 5. The statistical differences between the grain quality parameters viz., protein, starch, gluten and sedimentation value of wheat varieties as influenced by sowing dates (pooled data of 2 years)

Treatments	Protein		Starch		Gluten		Sedimentation value	
	Nov. 20	Dec. 20	Nov. 20	Dec. 20	Nov. 20	Dec. 20	Nov. 20	Dec. 20
Varieties								
PBW 343	12.25	11.10	64.2	64.3	32.6	29.3	45.4	35.3
PBW 502	11.05	10.90	64.9	64.9	29.7	28.3	36.2	32.3
UP 1109	12.50	11.80	63.3	63.0	32.9	30.7	46.0	39.2
UP 262	12.65	12.25	63.4	63.4	33.1	32.8	44.8	44.0
UP 2338	12.95	12.60	62.3	62.6	34.5	33.3	49.0	44.5
PBW 373	11.53	11.50	64.6	64.9	30.9	30.5	39.3	38.6
Raj 3765	11.85	11.75	63.8	64.3	30.3	30.2	39.0	38.4
UP 2382	12.45	12.25	64.1	63.5	32.8	32.1	44.9	44.6
UP 2425	11.85	11.80	64.5	63.8	31.7	30.7	37.0	36.4
UP 2565	13.15	12.25	62.6	63.3	35.7	32.1	50.8	40.7
	SEm ±	CD (5 %)	SEm ±	CD (5 %)	SEm ±	CD (5 %)	SEm ±	CD (5 %)
For comparing two varieties at same date	0.20	0.58	0.19	0.6	0.53	1.5	1.45	4.2
For comparing two dates at same or different varieties	0.29	0.98	0.27	0.9	0.69	2.3	1.95	6.6

**Fig. 2A.** Protein content of wheat varieties as influenced by sowing dates.

Starch

The effects of different varieties and their interaction with sowing dates were significant regarding the starch content of the grain; however, the starch content was not statistically influenced by the sowing dates themselves. As shown in Table 4, crops sown at normal times accumulated statistically equal amounts of starch as those sown later. Variety PBW 502 had the highest starch content, closely followed by PBW 373. Both of these varieties showed significantly higher starch levels than all the other varieties. Varieties UP 262, UP 1109 and UP 2565 had significantly higher starch content compared to UP 2338 (Fig. 2B). PBW 343, UP 2338, PBW 373, Raj 3765 and UP 2565 showed higher starch content in late sown condition (Table 5).

Gluten

The experiment demonstrated a significant interaction between different wheat varieties and sowing dates concerning gluten content. However, the impact of sowing dates alone was found to be non-significant. Notably, the varieties UP 2565 and UP 2338, both with gluten content of 33.9 %, were comparable to UP 262 and exhibited significantly higher gluten levels than all other varieties (Table 4). The variety UP 2382 also recorded significantly higher gluten content (Fig. 2C). Varieties PBW 343 and UP 2565 displayed comparable results, showing a significant reduction in gluten content when sowing was delayed by 30 days.

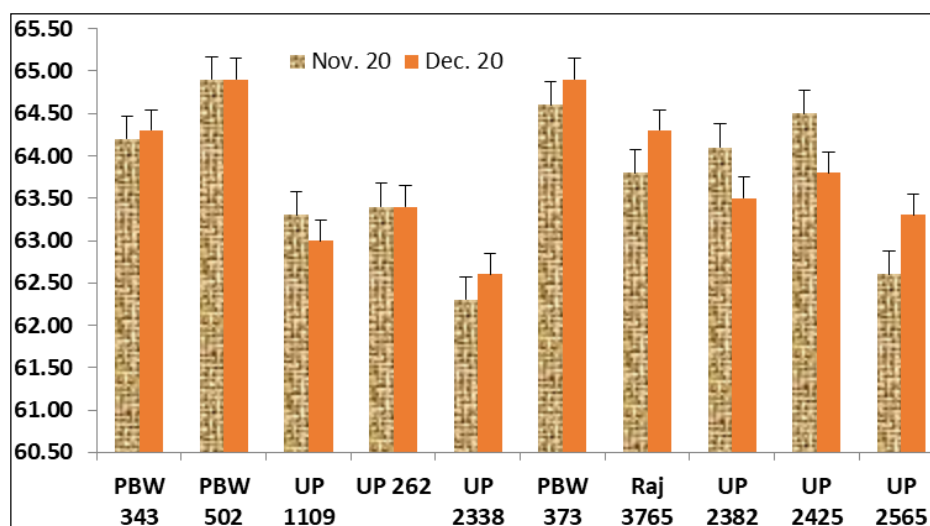


Fig. 2B. Grain starch content of wheat varieties influenced by heat stress at reproductive stage due to delay in sowing.

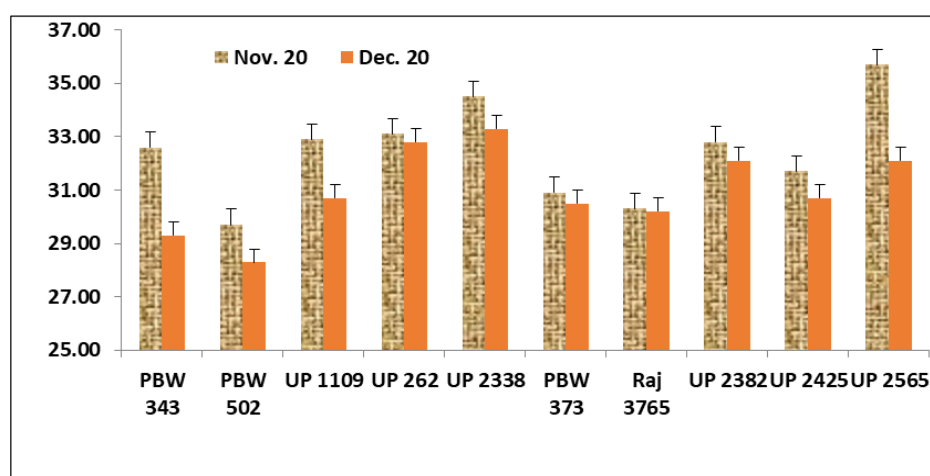


Fig. 2C. Grain gluten content of wheat varieties influenced by heat stress at reproductive stage due to delay in sowing.

Sedimentation value

The sedimentation value (mL) was significantly influenced by different wheat varieties and their interaction with sowing dates. As shown in Table 4, the normally sown crops had a significantly higher sedimentation value of 42.95 mL compared to those sown late (Table 5). Varieties PBW 373, Raj 3765, UP 2382 and UP 2425 exhibited higher sedimentation values for late-sown wheat (Fig. 2D)

Discussion

In wheat, delayed sowing exposes plants to high temperatures during the reproductive phase, causing significant morphological and physiological damage. This damage accelerates leaf senescence and reduces photosynthesis and pollen viability. Furthermore, it decreases starch biosynthesis and the reserves of water-soluble carbohydrates (WSC), while also shortening the grain-filling period (12-14). Elevated temperature from anthesis to maturity affects the grain yield by shortening the period of grain synthesis, which is crucial for accumulating photo-assimilates (17).

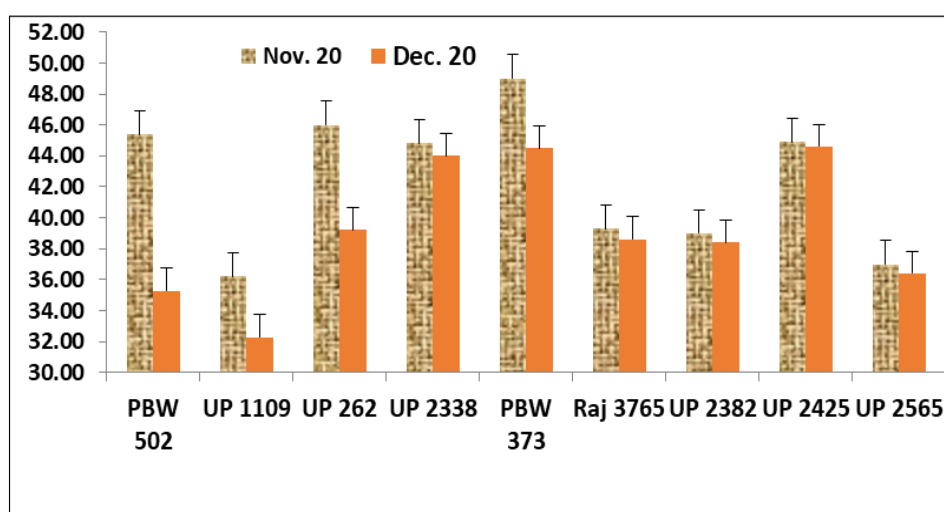


Fig. 2D. Grain sedimentation value of wheat varieties influenced by heat stress at reproductive stage due to delay in sowing.

This discussion focuses on the variations in grain yield and quality under late sowing as a result of high temperatures. Grain yield and its attributes were significantly influenced by sowing dates and varieties. A delay in sowing by 30 days resulted in a 17 % yield loss compared to normal sowing. Specifically, the loss in grain yields due to the delay from November 20 - December 20 (30 days) amounts to 32.86 kg ha⁻¹ day⁻¹. High temperatures during the post-anthesis period of the late-sown crop may contribute to this reduced yield. Previous studies have also reported yield losses due to delayed sowing. Previous studies noted similar findings (18, 19). It was observed that sowing wheat on the normal dates of November 18-20 resulted in higher grain yields compared to late sowing (December 2-5) or very late sowing (December 18-20) (20). Another study reported that late-sown crops experienced low temperatures during the early growth period, which restricted vegetative growth (21). Conversely, high temperatures during the post-anthesis period accelerated dry matter accumulation but shortened the duration of grain development, leading to a grain yield reduction of 37.5 kg ha⁻¹ per day. (An increase in temperature reduces the duration of grain growth, leading to a decline in grain weight at maturity by approximately 5 % for each 10 °C rise in temperature within the range of 17.7 - 32.7 °C (22).

"Wheat grown in hotter environments produced fewer grains and less biomass, leading to a comparatively lower yield due to elevated temperatures during the reproductive stage" (22). High temperatures can affect not only the chemical composition and metabolism of pollen but also its morphology and the rate of pollen tube growth (25). Elevated temperatures may adversely affect both the grain development rate and duration, resulting in shrivelled, immature and low-quality grains. For instance, a 2 % reduction in protein content was observed in late-sown crops (December 20) compared to normally sown crops (November 20). It was also reported that sowing on November 15 could yield better-quality crops (26). Late-sown crops experienced higher temperatures during grain development, which likely negatively impacted protein accumulation. A study similarly documented a decline in grain protein accumulation with increasing temperatures (3). It was previously reported that the correlation between protein and starch content was both negative and significant in normal as well as late-sown conditions (27). This is understandable, as the synthesis of both starch and protein occurs during grain development, leading to competition for limited energy resources. Even when adequate assimilates are available, the accumulation of starch in wheat grains can be reduced by over 30 % at temperatures between 30 °C and 40 °C, with the most critical period occurring during early grain filling (5). Enzymes involved in starch accumulation are vital targets for enhancing adaptation to warmer climates. Starch synthesis activity peaks at temperatures between 20 °C and 25 °C, while at temperatures above 35 °C, the starch synthase (SS) enzyme becomes non-functional, with 97 % of its activity lost as temperatures rise to 40 °C. SS (starch synthase) and SBE (starch branching enzymes) form hetero complexes in wheat, although the mechanisms underlying this complex formation are still not fully understood. Phosphorylation acts as a molecular switch that determines whether proteins can physically interact. This ensures tight regulation of biological processes such as signal transduction, gene expression and cell cycle progression. There is considerable genetic variation in tolerance to short periods of high temperature during

grain filling, affecting both grain yield and quality, which indicates potential breeding opportunities (5).

In wheat and barley, the process of conversion of sucrose to starch is impaired at elevated temperatures. At 30 °C, developmental processes are 1.5 to 2 times faster than at 20 °C, although the sequence of development is the same. Endosperm cell formation begins earlier at the higher temperature and ceases sooner, resulting in a final grain weight reduction of 27 %. Similar numbers of cells are found in sections of ripe grain grown at both temperatures, but cells in grain grown at 30 °C are smaller than those grown at 20 °C. Thus, while the rate of cell division increases at high temperatures, compensating for the reduced duration of division, the rate of cell enlargement does not rise sufficiently to make up for the reduced duration of cell enlargement. A reduction in gluten content was observed in late-sown crops compared to those sown on time; however, this reduction was not statistically significant (Table 3). Significant differences were noted among the varieties for both parameters. Based on experiments conducted with various wheat varieties at five locations in the Northwest Plain Zone, variation in sedimentation values between UP 2338 and PBW 502 is primarily due to genetic differences affecting protein content and gluten quality. UP 2338's higher sedimentation value indicates superior gluten strength, while PBW 502's lower value suggests weaker gluten, making each variety suited for different end-use applications.

Sowing dates did not have a significant impact on sedimentation value. Coordinated wheat experiments conducted at the five locations in the North Zone did not find significant differences in sedimentation values and are nearly the same.

Conclusion

The study concluded that among various environmental conditions, temperature is the most significant factor influencing both the yield and quality of wheat grain, particularly during the later stages of growth. Heat stress occurring from anthesis to physiological maturity affects the availability of assimilates, the translocation of photosynthates to the grain and the synthesis and deposition of starch in the developing grain. The net result is a reduction in kernel weight. While a shorter grain-filling duration is partially offset by an increased filling rate, the overall effects are quite complex. Furthermore, the extent to which different wheat varieties vary in their quality traits is influenced by the temperature during the reproductive growth phase. The interaction between variety and sowing date had a significant impact on yield and quality traits. This interaction demonstrates the potential for selecting varieties that exhibit improved stability in yield and quality under late-sown conditions. Verifying the study's results for different sowing dates and their interaction with wheat varieties will confirm that a proper combination of sowing date and variety can serve as a tool for producing quality-based cereal foods that meet consumer health benefits.

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Authors' contributions

RCN conceptualized and conducted the field studies and drafted the manuscript. HS performed the statistical analysis and contributed to drafting the manuscript. VPS contributed to the study design and coordination, while VS edited and corrected the manuscript. All authors reviewed and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interest to declare.

Ethical issues: None

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