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RESEARCH ARTICLE

Assessment of genetic variability and principal component analysis (PCA) for rice starch viscosity traits

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Abstract

Rice quality evaluation has a significant impact in deciding consumer acceptability and its suitability for various end-user applications. In the current investigation, we have evaluated a set of 54 genotypes, including landraces, popular varieties and basmati accessions for amylose content and viscosity profile to study the significant componential traits that contribute to rice quality through PCA. These cultivars were also categorized based on stability ratio, a valuable attribute that signifies rice's stability and cooking quality. Significant variation was found between the cultivars for all traits evaluated in the study. PCA was employed in the study to investigate the relationship between viscosity profile parameters and AAC and their contribution to phenotypic variability among these accessions. PCA analysis revealed the maximum contribution of the first two PCs (76.41 %) to the total variability. Further, based on their stability ratio, the accessions were categorized into three stability groups: 12 cultivars in the high, 39 in the medium and 3 in the low stability ratio category. The study led to the identification of traits that contributed the maximum to the observed variation and these findings can be efficiently utilized in determining the rice quality and the end-use application of rice starch. The findings of this study can be directly applied in breeding programs to improve consumer-preferred rice varieties by selecting high-quality parental lines and targeted genetic improvement and in food processing industries by tailoring rice for specific end-use applications and optimizing processing conditions based on viscosity parameters.

Keywords: AAC; PCA; RVA; stability ratio; viscosity

Introduction

Rice is the world's most significant food crop, a widely cultivated staple food and has been a dietary staple for millions of people for centuries. Rice quality assessment is vital for ensuring food security and is increasingly essential in safeguarding consumer well-being and promoting agricultural and economic development. Understanding its physiochemical properties is critical to assuring quality, processing and utilization. Rice quality is mainly assessed by four characteristics: milling quality, appearance, nutrition, cooking and eating quality (1). The texture of the cooked rice and its eating quality are the two essential criteria that can significantly impact the consumer preference for a rice variety (2, 3). Pasting properties are essential functional attributes of starch (4) and it can be explained as the phenomenon that follows gelatinization and is characterized by granular expansion, molecular component extrusion and complete disruption of starch granules (5, 6). Rapid Visco Analyser (RVA) is an essential instrument for analyzing the pasting properties of rice starch and can provide information about rice starch's stability and other characteristics (7). It is regarded as a quick and repeatable analytical method to evaluate the ability of rice flour and starch to gelatinize and undergo retrogradation during heating and cooling cycles (8). Different rice varieties can also be distinguished using RVA, such as *japonica*, *indica*, aromatic and glutinous types, based on their unique pasting profiles (9). The various viscosity parameters like peak viscosity (PV), trough viscosity (TV), breakdown viscosity (BDV), setback viscosity (SBV), final viscosity (FV), peak time (PT) and pasting temperature (PTem), are leading indicators for evaluating rice cooking quality. These parameters of RVA provide insights into the thermal, textural and retrogradation behaviour depending upon the starch paste viscosity parameters analyzed during RVA. Recent investigations have highlighted the importance of RVA in rice starch characterization, a study examined the pasting behaviours in 21 Asian rice varieties, emphasizing the difference between indica and japonica types (10) and another study explored how heating modes affect rice starch pasting and morphology (10, 11).

The stability ratio is another critical derived parameter from RVA pasting profiles. It is the proportion between the viscosity at the start of cooling and the viscosity at the peak of heating. It is a valuable indicator of the resistance of rice starch to shearing and thermal breakdown during processing. A higher stability ratio suggest greater stability under high-temperature and shear conditions. It signifies that the starch granules are more resistant to disintegration and maintain their viscosity even under high temperatures and shear stress (12). Recent studies have analyzed how different starch properties influence thermal and mechanical stress resistance during processing. For instance, a systematic comparative analysis of starches from various rice varieties demonstrated that pasting behaviours, assessed through RVA profiles, play a crucial role in determining starch stability, cooking quality and end-use application (13).

The multivariate analysis technique, Principal component analysis (PCA), helps reduce redundancy and find patterns in datasets by classifying genotypes and revealing similarities between variables (14). 'Eigenvalues' assess how each component contributes to the overall variance, whereas the coefficients in eigenvectors show how much each original variable influences each principal component. The main advantage of PCA is its ability to evaluate each dimension's contribution to the variations observed within a dataset (15). PCA has successfully identified genetic variations in starch viscosity traits, characterizing the diversity of grain nutritional and physicochemical quality, characterizing the diversity of grain phytic acid content and determining the genetic diversity in rice crops (16-19).

This study assessed 54 rice accessions for their starch viscosity parameters and apparent amylase content (AAC). We deciphered the significant componential traits that contribute to rice grain quality through PCA. Further, the genotypes were categorized into different grain quality groups based on their stability ratio obtained from viscosity parameters.

Material and Methods

Plant Material

A total of 54 genotypes, including breeding lines, popular varieties, landraces and Basmati accessions, were employed in the study; the rice materials were planted during *Kharif* or wet season (May-October), 2018-19 under hot and tropical humid climatic conditions at the research farm of ICAR-Indian Agricultural Research Institute (ICAR-IARI), New Delhi (Latitude 28°38' 23", Longitude 77°09' 27"E and Altitude 228.61 m above mean sea level). The sandy loam soils of the ICAR-IARI experimental fields in New Delhi have a pH of 7.4-7.8, which is neutral. All the accessions were grown in an augmented randomized complete block design (20). These lines were harvested at maturity, air-dried and stored at room temperature until analysis. A laboratory dehusker (Satake Corp., Japan GF6434) was used to turn the paddy samples into brown rice and a lab milling equipment (Satake Corp., Japan, TM05C) was then used to turn them into white rice. Approx. 20 gm of each sample was subsequently processed in the Foss Cyclotec ball mill until they passed through a 1-mm sieve, yielding flour for further analysis.

Determination of apparent amylose content (AAC)

An iodometric technique based on measuring the amylose-iodine complex was used to analyze the AAC of isolated rice samples. Exactly 50 mg of homogenized rice flour was used and suspended in 500 µL ethanol. After that, 4.5 mL of 1N sodium hydroxide was added and the tubes were heated to 100 °C for 15 min in a boiling water bath. The solution was cooled in an amber-coloured tube, a 500 µL aliquot was taken and 200 µL of iodine solution and 100 µL of glacial acetic acid were combined. The intensity of the blue colour developed in the solution when it was incubated for 20 min at room temperature in the dark and was measured by recording the absorbance at 620 nm in a spectrophotometer (Labomed, Inc UVD-3200). The percentage of AAC in each sample was determined by utilizing a standard curve created concurrently using rice samples with known amylose contents.

Determination of viscosity profiles

The pasting characteristics of rice flour samples were assessed in 13 min using the AACC 2000a operational methodology using RVA (RVA 4500, Perten Instruments AB, Sweden). About 3.5 g of flour (based on 11 % moisture) from each rice sample was weighed separately into the aluminium canister and mixed with 25 mL of distilled water. The heat-hold-cool temperature cycle in the RVA is set as follows: Heating for 1 min at 50 °C; heating at 95 °C linearly raised over 3 min and 42 sec; heating at 95 °C for 2 min and 30 sec, cooling for 3 min and 48 sec to reach 50 °C and then remaining there for 2 min. Each sample was analyzed in triplicates and the RVA curve was used to determine the viscosity parameters PV, TV, BDV, SBV, FV, PT and PTem. All viscosity parameters were reported in Rapid Visco Amylograph Units (RVU). The ratio of final viscosity to hold viscosity was used to compute the setback ratio, while the stability ratio was determined by dividing the trough viscosity by the peak viscosity.

Statistical analysis

Microsoft Excel computed descriptive statistics, such as each trait's minimum and maximum range and mean.

Principal component analysis (PCA)

PCA was conducted with STAR 2.0.1. (IRRI 2014 a,b) tool exhibiting total variation for starch parameters, including AAC and seven RVA parameters, were derived separately. PCA was used in the present study as it reduces the dimensionality while maintaining maximum variance, allowing the identification of key traits contributing to rice quality. The correlation structure among the component traits was degraded to components, leading to variance observed within the components. Significant PCs were determined as the components having eigenvalues exceeding one. The genotype factor coordinates were calculated for the obtained PCs. The contribution of each variable to each PC was computed using the factor-variable correlations (factor loadings). The most influential traits were determined based on their relative contributions to the first PC, second PC and so forth. The genotype contribution to PCs was utilized to scatter them to determine the genotypes linked to those variables and calculate the overall variability of the data.

Results

Genotypic variance for the parameters of the viscosity profile and AAC

The evaluated set showed wide variation for AAC and the seven viscosity profile parameters were analyzed. The AAC of milled rice flour varied from 12.85 % (Muskan) to 34.41 % (Kamlesh), with a mean of 25.1 ± 0.7. Significant variation was also shown by the viscosity profile parameters, with TV showing variation from 1224 (Jhulhat) RVU to 4487 RVU (Sitwa Dhan) with an average of 2360.9 ± 85.2 and PV ranging from 1427 RVU (Jhulhat) to 5683 RVU (Sitwa Dhan) with a mean of 2939.6 ± 121.5 RVU. A notable difference was also observed in the case of BDV, where SBV varied from 642 RVU (Langphou) to 4518 RVU (Aziz Beoul), with a mean value of 2697.6 ± 105.1 RVU, whereas the range was 37 RVU (Swarna Sub1) to 2682 RVU (Japonica), with an average of 585.3 ±73.8 RVU. The FV marked deviation from 2051 RVU (Langphou) to 7736 RVU (PB-1728) with an average of 5058.5 ± 148.8 RVU. PT varied from 5.44 Minutes (Japonica) to 7.00 Minutes (Pratikshya, Jhulhat, Aziz Beoul, Jayati and Chandana) with an average of 6.08 ± 0.05 RVU, while PTem displayed a range of 76.23 °C (Premium Egyptian rice) to 94.8 °C (Chandana) with a mean of 87.74 ± 0.58 °C (Table 1). All the cultivars' evaluated traits are provided in the supplementary table (ST1).

Stability ratio

The rice cultivars were divided into three different pasting types by stability ratio. It might be helpful in choosing rice cultivars according to the desired paste qualities and starch processability attributes. The categories of high stability ratio (> 0.95), medium stability ratio (0.65-0.95) and low stability ratio (< 0.65) included 12 cultivars in the high, 39 in medium and 3 in the low stability ratio category (12). The details of the 54 cultivars based on the categorization are mentioned in Table 2.

Association among the AAC and RVA pasting traits

The magnitude and direction of correlation among the traits studied are depicted in Table 3. AAC showed a significant positive correlation with FV (0.37; p<0.01) and SBV (0.44; p<0.001), while it depicted a negative correlation with BDV (-0.30; p<0.05). While PV displayed a strong positive correlation with TV (0.80; p<0.001), BDV (0.70; p<0.001) and FV (0.34; p<0.05), there was a negative correlation with PT (-0.49; p<0.001) and PTem (-0.67; p<0.001). The TV showed a strong positive correlation with FV (0.72; p<0.001) and was negatively correlated with PTem (-0.41; p<0.01). The BDV in the present investigation was found to be strongly significant, showing a negative correlation with SBV (-0.50; p<0.001), PT (-0.57; p<0.001) and PTem (-0.60; p<0.001). The results showed that FV was strongly positively correlated with both PT (0.35; p<0.01) and SBV (0.83; p<0.001). A significant positive connection was observed between SBV and both PT (0.63; p<0.001) and PTem (0.52; p<0.001).

Principal component analysis

PCA was carried out to understand how AAC and viscosity profile parameters contributed to the total phenotypic variability of these 54 rice accessions. Different traits contribute to the overall variance calculated for each component. For the analyzed traits, PC1 and PC2, each having more than one eigenvalue, were considered the most significant PCs. These two PCs together explained a maximum of 76.41 % of the total variation among the accessions (Fig. 1). PC1 explained 43.96 % of the cumulative variance of the two significant PCs, while PC2 explained 32.46 % of the total variation. Factor loadings

Table 1. Descriptive statistics of the AAC and viscosity profile parameters of the accessions used for analysis

	AAC (%)	**PV** (RVU^3)	**TV** (RVU)	**BDV** (RVU)	**FV** (RVU)	**SBV** (RVU)	**PT** (Minutes)	**Ptem** (°C)
Mean ± SE	25.1 ± 0.7	2939.6 ± 121.5	2360.9 ± 85.2	585.3 ± 73.8	5058.5 ± 148.8	2697.6 ± 105.1	6.08 ± 0.05	87.74 ± 0.58
Minimum	12.85	1427	1224	37	2051	642	5.44	76.23
Maximum	34.41	5683	4487	2682	7736	4518	7	94.8

AAC- Apparent amylose content; PV- Peak Viscosity; TV- Trough Viscosity; BDV- Breakdown viscosity; FV- Final viscosity; SBV- Setback Viscosity, PT- Peak time; Ptem- Pasting temperature

Table 2. Categorization of accessions based on the stability ratio

Stability ratio	**Cultivars**
High Stability Ratio (>0.95)	Sambha Mahsuri, Manaswini, Nagina 12, Jayati, PB-1728, Kamlesh, Rajendra Basmati, Buta Baber, Improved Samba Mahsuri, Swarna Sub 1
Medium stability ratio (0.65-0.95)	Pant Dhan 10, Bala Koun. PB-1509, Sah Pasand, Kudrat-3, DV 85, PB-1609, PB-1121, Swarna, Narendra Usar Dhan III, Kalanamak, T. Basmati, Muskan, Sonasal, Chimbalate Basmati, MTU 1010, Sitwa Dhan, CSR 23, ASG-ST-34, Pusa-33, Pratikshya, Urvashi, Pusa Sugandha 3, Sharbati, Basmati-370, Ananga, Gouri, Tilak Chandan, Jhulhat, Ranbir Basmati, Chandana, Kanak, Bhubana, Aziz Beoul, Khara Munga, Bala, Kasturi, Langphou, PB-1401, Pusa-1342
Low stability ratio (<0.65)	Premium Egyptian Rice, Japonica, Lalat

Table 3. Correlation matrix showing the strength and direction of relationships between variables

	AAC	**PV**	**TV**	**BDV**	**FV**	**SBV**	**PT**	**PTem**
AAC	1.00	-0.11	0.10	-0.30*	0.37**	0.44***	0.23	0.01
PV	-0.11	1.00	0.80***	0.70**	0.34*	-0.164	-0.49***	-0.68***
TV	0.10	0.80***	1.00	0.13	0.72***	0.21	-0.16	-0.41**
BDV	-0.30*	0.70***	0.13	1.00	-0.28*	-0.50***	-0.57***	-0.60***
FV	0.37**	0.34*	0.723***	-0.28*	1.00	0.83***	0.35**	0.14
SBV	0.44***	-0.16	0.214	-0.50***	0.83	1.00	0.63***	0.52***
PT	0.23	-0.49***	-0.16	-0.57***	0.35**	0.63***	1.00	0.64***
PTem	0.01	-0.68***	-0.41**	-0.60***	0.13	0.52***	0.64***	1.00

*Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1*

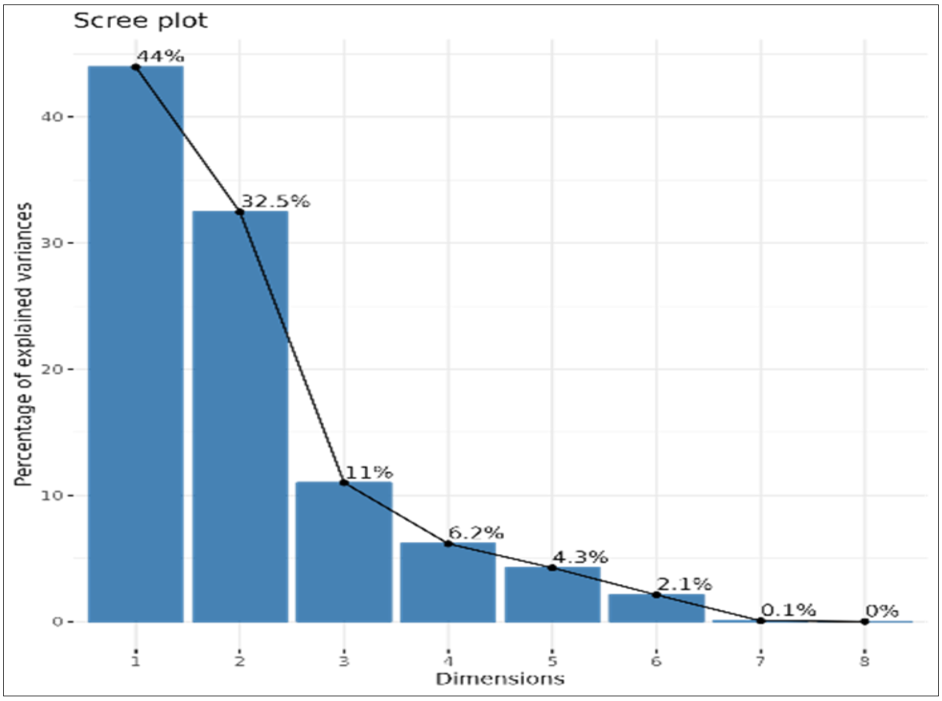

Fig. 1. Scree plot showing the percentage of explained variances. With the first contributing 43.96 % of the variability in the data and the second contributing 32.46 %, the two components together account for 76.41 % of the overall variability.

or factor-variable correlations indicated that except for PV, TV and BDV, all the other traits positively influenced PC1. Similarly, all but PTem exhibited a positive influence on PC2.

The PCA results provide valuable insights into how different traits influence rice quality and processing characteristics. Since PC1 (43.96 %) is primarily influenced by traits such as SBV, FV, PT and PTem, it suggests that gelatinization, retrogradation behaviour and texture stability are key factors in differentiating rice varieties. Higher values of these traits indicate starches that are more resistant to breakdown, which is beneficial for applications requiring firmer rice texture or processed rice products with better stability (21). On the other hand, PC2 (32.46 %), where all traits except PTem had a positive influence, highlights variations in viscosity-related properties like BDV and TV. These factors determine how rice starch responds to heat and shear forces. It is crucial for assessing its suitability for different cooking methods or food processing industries (e.g., noodle production, instant rice, or baby foods) (22). In practical terms, rice breeders can use these findings to select varieties with desirable pasting and texture properties. At the same time, the food industry can tailor rice-based products based on the identified stability and viscosity profiles, ensuring better product consistency and consumer satisfaction.

Eigenvectors and the percentage contribution of the individual traits to the significant PCs obtained by multiplying the squared eigenvectors with 100 are given in Table 4. Vectors of variable contributions to the amylose content and viscosity profile parameters towards major PCs are shown in Fig. 2. Variable contributions strongly influence BDV and PT towards PC1, with 19.77 % and 19.54 %, respectively. TV and FV were the most contributing traits to PC2, explaining 31.59 % and 31.60 % of variation to PC2, respectively (Table 5). Cos2 values indicate the quality of the variables' representation on the factor map. A high cos2 value shows a good indication of the variables on the PC. BDV, PT and PTem had higher cos2 values with 0.70, 0.69 and 0.68, respectively, for PC1, whereas TV and FV had a cos2 value of 0.82 for PC2.

Discussion

The present study delves mainly into the amylose content and viscosity profile parameters of the 54 rice cultivars. The rice collection displayed a broad range of variance for AAC, PV, TV, FV, BDV, SBV, PT and Ptem (ST 1). Amylose is the most essential component of rice starch that determines the

Table 4. Eigenvalues and Eigenvectors represent the variance explained by each principal component and the contribution of variables to these components

	PC1	PC2	PC3	PC4
Eigenvalues and Variance %				
Eigen Value	3.52	2.60	0.88	0.49
Variance %	43.96	32.46	11.01	6.17
Cumulative Variance %	43.96	76.41	87.42	93.58
Eigenvectors				
AAC	-0.21	-0.23	0.88	0.18
PV	0.38	-0.42	-0.14	0.17
TV	0.14	-0.56	-0.15	-0.36
BDV	0.44	-0.03	-0.08	0.77
FV	-0.21	-0.56	-0.14	-0.02
SBV	-0.41	-0.34	-0.07	0.26
PT	-0.44	-0.03	-0.18	0.37
PTem	-0.44	0.16	-0.35	0.13

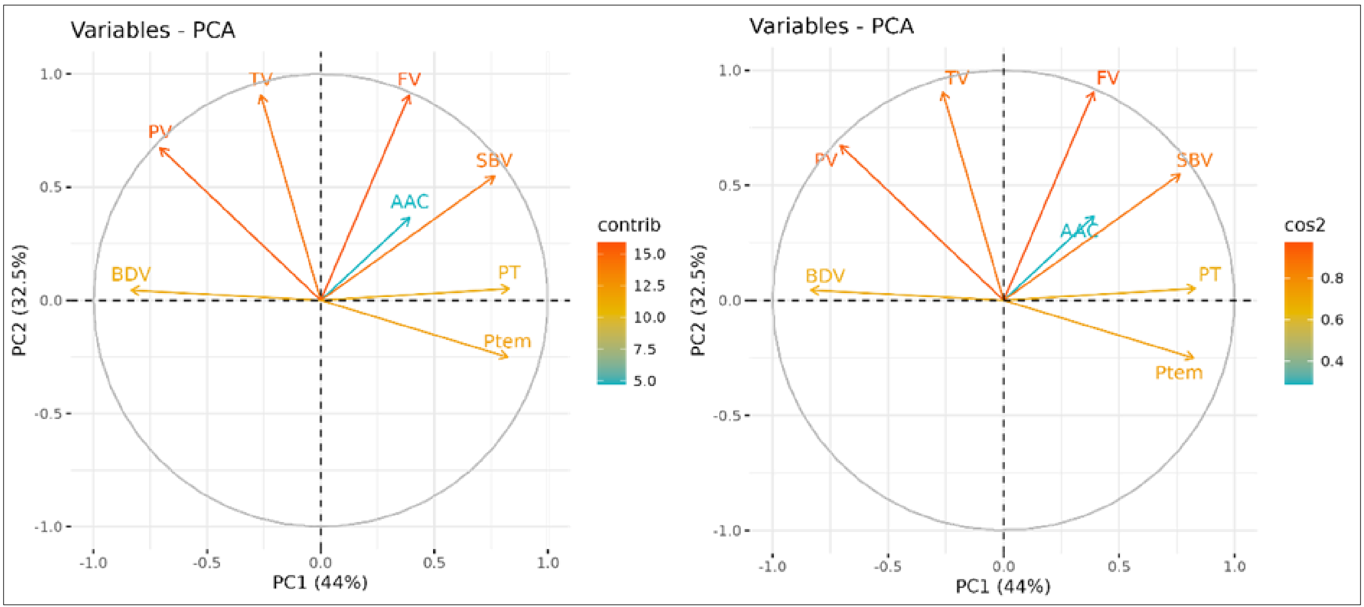

Fig. 2. (a) Vectors of variable contributions towards various traits towards major principal components (PCs). The direction of the variable coordinates shows the direction of their influence on PCs. (b) The first principal component (PC1) vs. second principal component (PC2) score bi-plot shows the means grouped by combined two traits.

Table 5. PCA-trait statistics showing the relationship between principal components and trait contributions, including factor loadings and explained variance

	Correlation between PCs and Traits		Quality of representation of Traits on PCs		Contributions (%) of the Traits to the PCs	
	PC1	**PC2**	**PC1**	**PC2**	**PC1**	**PC2**
AAC	0.39	0.37	0.15	0.13	4.37	5.16
PV	-0.71	0.67	0.50	0.45	14.21	17.47
TV	-0.26	0.91	0.07	0.82	1.97	31.59
BDV	-0.83	0.04	0.70	0.00	19.77	0.08
FV	0.39	0.91	0.15	0.82	4.31	31.60
SBV	0.76	0.55	0.58	0.30	16.60	11.58
PT	0.83	0.05	0.69	0.00	19.54	0.10
PTem	0.82	-0.25	0.68	0.06	19.24	2.42

hardness or softness of cooked rice. Higher amylose content (>25 %) is related to harder-cooked rice and upon cooling, the rice becomes less tender, dry and separate. In contrast, the low amylose varieties are sticky, soft and glossy (23). The present set of rice accessions depict significant variation from low to very high for AAC, which can be exploited to develop rice varieties with better grain quality. The correlation analysis highlights the influence of AAC on rice quality and processing characteristics. Higher AAC correlates moderately with increased FV and SBV, indicating more substantial gel formation and firmer texture upon cooling (24). AAC reflects a negative correlation with BDV, which shows that higher AAC varieties exhibit more excellent stability under heat and shear (25). The weak correlation of AAC with PV and PT significantly impacts gelatinization rate and cooking time (26, 27). PV depicts the maximum viscosity attained during heating at 95° C and its variation is often linked with the swelling capacity of starch as well as the rate at which starch granules dissociate. (28). The accession *Sitwa Dhan*, showing the highest PV, explained the slow granular swelling and disruption compared to *Jhulhat*, which showed low PV where the starches hydrated and swelled rapidly. When sheared and heated, the swollen starch granules were broken up, resulting in TV, the lowest viscosity attained during heating at 95°C. The cultivars *Sitwa Dhan, Bhubana, PB-1401, Ananga* and *Urvashi* showed high TV coupled with high PV which suggests that the starch in these accessions not only swells considerably but also remains highly stable, making it ideal for applications that require both thickening solid powder and paste stability under heat and agitation (29).

BDV and Stability ratio are analyzed to understand the paste stability of rice cultivars. BDV was obtained to show the difference between PV and TV. The stability ratio was the viscosity ratio between TV and PV. Various factors influence the stability ratio, such as amylose and amylopectin content, starch granule properties, processing conditions and cultivar and environment. A high stability ratio indicates strong resistance to shear and minimal breakdown during heating and it is also correlated with strong shear resistance and poor swelling power. A medium stability ratio indicates the balance between shear resistance and acceptable swelling power and a low stability ratio indicates high susceptibility to breakdown under shear or thermal stress. It is linked to increased swelling power and hydration, resulting in less stable pastes. Most of the cultivars used in the current study were found to be in the medium stability ratio category. Breeders can develop targeted rice varieties suited for specific consumer preferences, industrial applications and diverse culinary requirements, enhancing market adaptability (30, 31).

Paste stability describes how starch paste hydrates, swells and resists shearing when heated. High shear resistance, poor hydration and swelling power are often linked to low BDV and high stability ratios (28). *Swarna Sub 1*, showing low BDV and high stability ratio, suggests highly stable starch during heating and after cooling.

FV was measured after the starch molecules were retrograded and reformed in the starch granules and cooled at 50 °C. After heating and cooling, the FV measures the starch's viscous paste capacity. The starch from the accession *Langphou* with the highest FV can be used as a suitable thickening agent for food applications (32). SBV measures the thickening of starch pastes during cooling, indicating starch retrogradation (especially amylose re-association). It gives insight into retrogradation (the re-association of starch molecules), particularly amylose, during the cooling phase. In the present investigation, *Aziz Beoul*, with the highest SBV (4518 RVU), indicates stronger retrogradation and tends to have higher AAC (28.8 %), as it has been reported that amylose is primarily responsible for retrogradation and SBV (33, 34). PTem influences the behaviour of starch during cooking and processing. It impacts gelatinization, texture, processing efficiency and overall quality. The greater pasting temperature indicated the ability of the component to withstand swelling, which might be correlated with the amount of amylose and amylopectin found in the rice varieties (20).

The PCA conducted in the present investigation offered essential insights regarding the relationship between amylose content and viscosity profile parameters and their contribution to the overall phenotypic variability observed in the 54 rice accessions. The first two PCs (PC1 and PC2) accounted for a significant portion of the total variation and the characteristics linked to these components prove more valuable in distinguishing between accessions (17, 36, 37, 38). The scree plot explained that PC1 showed the maximum variation compared to other PCs (Fig. 1).

The factor loadings and eigenvectors revealed that, except for PV, TV and BDV, all the other traits, including amylose content, positively influenced the first PC. This suggests that AAC and most of the viscosity parameters are closely associated, collectively contributing to the primary source of variation among the rice accessions. The second PC was strongly influenced by traits such as TV and FV, indicating that these parameters account for a significant portion of the remaining variability not captured by the first component.

The variable contributions further highlighted the significant impact of BDV and PT on the first PC, collectively explaining nearly 40 % of the variation. In contrast, TV and FV were the primary contributors to the second PC, collectively accounting for over 60 % of the variation. It can be deduced that among the factors considered, BDV, PT, TV and FV contributing maximum to the observed variation can be strategically considered when choosing parental breeding lines. Characters that tend to coexist and converge across several PCs explaining variance should be considered while using these traits in the breeding program (39,40)

Conclusion

To sum up, the study analyzed the degree of genetic variability and diversity for amylose content and viscosity profile parameters, which are essential for rice cooking and textural traits and identified promising donors for enhancing the quality traits and maintaining superior attributes. Stability ratio also has applications in rice breeding and processing by selecting cultivars based on pasting and stability characteristics and breeding for higher stability ratio in regions with high processing requirements. Understanding the stability ratio helps classify rice varieties based on their processing suitability and guides breeding programs for improved rice starch functionality.

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Authors' contributions

NS contributed to writing the original draft, validation, methodology, investigation, formal analysis and data curation. KP was responsible for visualization, supervision and methodology. SK provided validation, supervision, resources and project administration. PB contributed to visualization, validation, resources, project administration and methodology. MN was involved in supervision and resources. KV participated in writing - review & editing, visualization, validation, supervision, software and project administration. RE was responsible for validation, supervision, project administration and investigation. AS contributed to visualization, validation, supervision, project administration and funding acquisition. HB was responsible for writing - review & editing, visualization, validation, supervision, software, project administration, funding acquisition and conceptualization.

Compliance with ethical standards

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

Ethical issues: None

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