Genetic, phenotypic, and agronomic diversity of arrowroot (*Maranta arundinacea*) across agroclimatic zones for environmental adaptation and conservation

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[2]Department of Agrotechnology, Faculty of Agriculture, Universitas Jember. Jl. Kalimantan 37, Jember 68121, East Java, Indonesia
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Abstract. *Hariyono K, Noviana L, Ubaidillah M, Kurnianto AS, Handoko RNS. 2024. Genetic, phenotypic, and agronomic diversity of arrowroot (*Maranta arundinacea*) across agroclimatic zones for environmental adaptation and conservation. Biodiversitas 25: 3431-3441.* Understanding the genetic and phenotypic diversity of crops like arrowroot is essential for developing sustainable agricultural practices and ensuring food security. This study, therefore, is of significant importance as it provides insights into how environmental factors influence trait variability, which can inform targeted conservation and agronomic strategies in the context of climate change. This study investigated the genetic, phenotypic, and agronomic diversity of arrowroot (*Maranta arundinacea* L.) across three agroclimatic zones in Jember District, East Java Province, Indonesia, with a focus on the impact of environmental factors on these traits. Sampling was conducted in Wuluhan, Sumbersari, and Arjasa subdistricts, covering 20 locations. Genetic analysis revealed that Arjasa presented the highest genetic diversity, with a median Genetics_Mean of 0.75 and an interquartile range (IQR) of 0.7-0.83, whereas Sumbersari and Wuluhan had median Genetics_Mean values of 0.8 and 0.8, respectively. Phenotypic diversity was also highest in Arjasa, with a Phenotypic_Mean of 0.83 (IQR: 0.78-0.86), whereas it was 0.8 in Sumbersari and 0.82 in Wuluhan. The quantitative traits presented less variability, with median Quantitative_Mean values of 0.22 in Arjasa, 0.19 in Sumbersari, and 0.21 in Wuluhan. The conservation priority index (CPI) was calculated, identifying Antirogo as the location with the highest CPI value of 0.440, indicating a high priority for conservation efforts. In contrast, Kebonsari had the lowest CPI value of 0.235. Principal component analysis (PCA) further highlighted the significant variability in genetic and phenotypic traits across sub-districts, particularly in Arjasa, where unique environmental conditions likely contribute to this diversity. The results emphasize the importance of targeted conservation strategies and agronomic practices to optimize the genetic and phenotypic potential of arrowroot, particularly in the face of environmental variability and global climate change.

Keywords: Agroclimatic variation, conservation strategies, environmental adaptation genetic diversity, phenotypic traits

INTRODUCTION

Arrowroot (*Maranta arundinacea* L.) is a significant tuber crop, particularly valued for its high amylose content, which can reach up to 35% of its dry weight. This makes it a vital food source in tropical regions where it is commonly grown and consumed (Tarique et al. 2021; Malki et al. 2023a). Beyond its role as a staple food, arrowroot starch is widely used in both the food and pharmaceutical industries. It is especially sought after in the production of gluten-free products and as a natural thickening agent, catering to the rising global demand for healthier, allergen-free food alternatives (Firoskhan and Muthuswamy 2021; Fidianingsih et al. 2022; Malki et al. 2023a). These attributes not only enhance the nutritional value of arrowroot but also increase its economic importance, supporting efforts toward food diversification and sustainability (Deswina and Priadi 2020).

However, the cultivation of arrowroot, like other tuber crops, faces significant threats from climate change and land-use conversion. The increasing frequency of extreme weather events-such as rising temperatures, prolonged droughts, and erratic rainfall-poses major risks to crop production. These climatic changes can disrupt photosynthesis, impair tuber development, and reduce both yield and quality (Taylor et al. 2018; Jacques et al. 2020). Additionally, extreme weather can accelerate tuber decay in the soil, further decreasing productivity. To address these issues, sustainable farming practices and innovative land management techniques have the potential to mitigate some of these negative effects.

In addition to climate challenges, the conversion of agricultural land for non-agricultural purposes exacerbates the problem. The ongoing trend of converting arable land to urban and industrial uses reduces the availability of fertile land, threatening food security and leading to environmental degradation (Guillaume et al. 2016; Ross et al. 2017). This loss of productive farmland makes it increasingly difficult to meet the food demands of a

growing global population, further highlighting the need for sustainable agricultural practices.

Against this backdrop, this study in Jember District, East Java, Indonesia, provides critical insights into sustainable arrowroot cultivation. Jember's diverse agroclimatic conditions support a range of horticultural crops, including tobacco, okra, and edamame. These crops thrive due to the region's high soil fertility and favorable environmental conditions. However, Jember is also facing the dual challenges of declining soil fertility and the conversion of agricultural land, which threaten the long-term sustainability of its agricultural sector (Rondhi et al. 2018; BPS Jember 2024).

The research focuses on exploring genetic and phenotypic variations in arrowroot in response to Jember's environmental conditions. Understanding these variations is crucial for developing climate-resilient arrowroot varieties that can withstand adverse conditions while maintaining productivity. The findings could serve as a model for developing resilient crops in other regions facing similar environmental challenges. Resilient varieties can play a crucial role in ensuring food security in the face of climate change by maintaining yields even in less-than-ideal conditions. Furthermore, this research could contribute to conservation strategies aimed at preserving crop genetic resources, which are vital for sustaining agricultural productivity.

Genetic studies are essential for identifying and preserving crop traits like drought tolerance, disease resistance, and high yield potential, which are increasingly important in the context of climate change (Koshariya 2022). By analyzing the genetic diversity of arrowroot, this study offers valuable insights into selecting and conserving varieties that are more resilient to environmental stressors. These insights can inform breeding programs that aim to develop climate-resilient crops, ensuring the stability of food production systems in the long term.

This research provides practical solutions to pressing issues related to climate change and food security by focusing on developing climate-resilient crops and promoting sustainable agricultural practices. The integration of local agricultural research, such as the findings from Jember, with global food security efforts is crucial. It ensures that crops like arrowroot can continue to provide nutritional and economic benefits in an increasingly unstable climate. This study highlights the importance of adopting a global perspective in agricultural practices to safeguard future food supplies while addressing the immediate challenges posed by climate change and land-use conversion.

MATERIALS AND METHODS

Sampling sites

This study analyzed the genetic, phenotypic, and quantitative variations in arrowroot tubers across three subdistricts, i.e. Wuluhan, Sumbersari, and Arjasa in Jember District, East Java Province, Indonesia, sampling a total of 20 sites within these areas (Figure 1, Table 1). Data collection was carried out through field sampling at each of the 20 locations, with each location representing one accession of arrowroot. In total, 20 accessions were collected, and each accession consisted of one representative sample per location. Therefore, the study analyzed 20 samples, one from each site, ensuring a broad representation of the genetic, phenotypic, and agronomic variations of arrowroot across the three subdistricts.
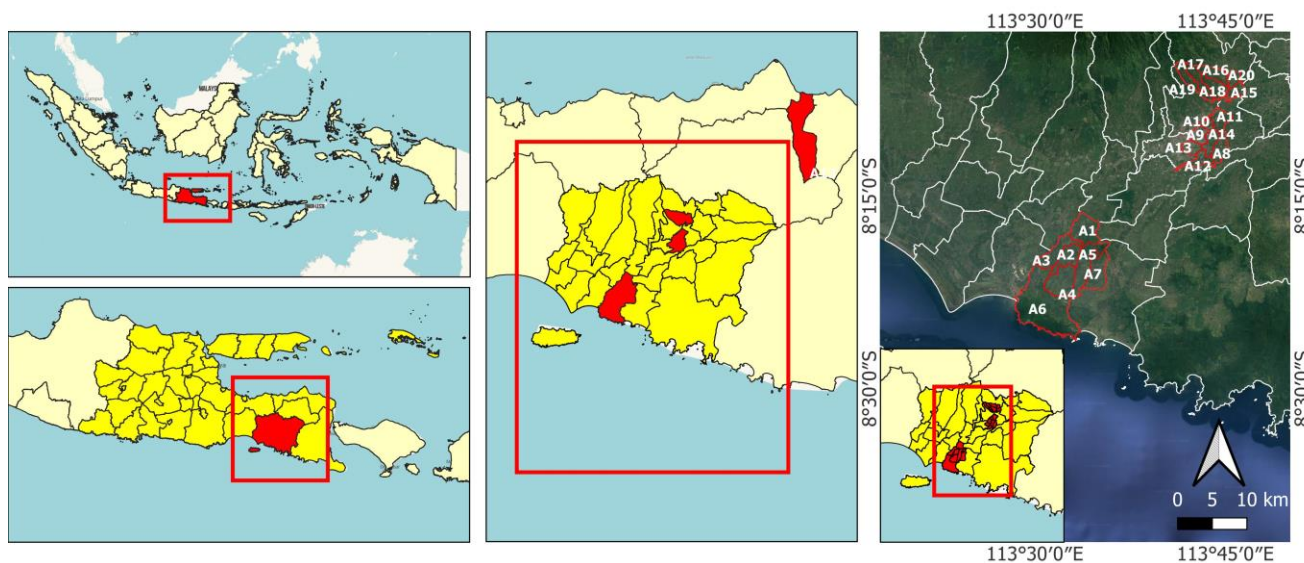
Figure 1. A map showing the research location in Jember, East Java, Indonesia

Table 1. List of samples used in the research in Jember District, Indonesia

Number of accessions	Sub-district	Location	Coordinate
A1	Wuluhan	Glundengan	-8.1711889, 113.3443869
A2		Dukuh Dempok	-8.1855331, 113.338.759
A3		Taman Sari	-8.1853422, 113.3243563
A4		Ampel	-8.2136212, 113.3154684
A5		Tanjung Rejo	-8.1958627, 113.3351869
A6		Lojejer	-8.2139411, 113.3031277
A7		Kesilir	-8.2024071, 113.3451642
A8	Sumbersari	Wirolegi	-8.2027360, 113.7417028
A9		Sumbersari	-8.1734203, 113.7149619
A10		Tegal Gede	-8.1558902, 113.7233267
A11		Antirogo	-8.1520360, 113.7514017
A12		Kranjingan	-8.2105708, 113.6962538
A13		Kebonsari	-8.1827504, 113.7095214
A14		Karangrejo	-8.1910525, 113.7296896
A15	Arjasa	Biting	-8.1135769, 113.7658917
A16		Kamal	-8.0892458, 113.7424348
A17		Darsono	-8.1097830, 113.7249669
A18		Arjasa	-8.1169794, 113.7389231
A19		Kemuning Lor	-8.0840243, 113.6892647
A20		Candijati	-8.1035345, 113.7606231

Genetic analysis

The twenty selected samples were then analyzed genetically and phenotypically. Genetic analysis follows the RAPD method. DNA was extracted from young arrowroot leaves via the CTAB buffer method, as modified in this study based on the SDS protocol. DNA extraction was conducted to obtain gene templates, which were then analyzed via polymerase chain reaction (PCR). In the PCR process, 20 primers were used with a mixture of materials per sample consisting of 5 μL of Taq DNA Polymerase, 1 μL of forward primer, 1 μL of reverse primer, 1 μL of DNA template, and 2 μL of nuclease-free water. The RAPD primers used in this study, such as OPA-01 (5'-CAGGCCCTTC-3'), OPA-02 (5'-TGCCGAGCTG-3'), and OPB-01 (5'-GTTTCGCTCC-3'), are widely applied in genetic diversity analyses across various plant species (Table 2).

These primers have been shown to effectively amplify polymorphic DNA bands, which are crucial for assessing genetic variability. Similar studies have utilized these primers to analyze genetic diversity in crops such as rice, bamboo, and *Flacourtia rukam* Zoll. & Moritzi (Alam et al. 2014; Desai et al. 2015; Zulfahmi et al. 2023). The PCR program was set with a predenaturation phase for 2 minutes at 95°C, followed by 30 cycles, including denaturation for 30 seconds at 95°C, annealing for 30 seconds at 55°C, and extension for 1 minute at 72°C, with final extension for 5 minutes at 72°C. DNA amplification was carried out via electrophoresis with 1% agarose media and 5 μL of EtBr DNA stain. The amplification results were then observed via a UV transilluminator to detect the presence of arrowroot plant DNA.

Phenotypic traits analysis

Phenotypic traits were classified into two categories: qualitative parameters, which included the morphological characteristics of the roots, stems, leaves, and tubers, such as the root system, root color, stem growth direction, stem color, and tuber shape and color; and quantitative parameters, which included measurements of the number of nodes, tuber circumference, tuber length, tuber weight, stem height, leaf length, and leaf width, recorded manually for each accession in each sub-district. We then scaled and coded genotype, phenotype (qualitative), and quantitative parameters for statistical analysis. This involved assigning a score of 0 was used to indicate the absence or mismatch of a characteristic. In contrast, a score of 1 was used to indicate the presence or match of a characteristic according to the defined criteria.

Table 2. List of primers and their sequences used in this study

Primer	Sequence (5'-3')
OPA-01	5'-CAGGCCCTTC-3'
OPA-02	5'-TGCCGAGCTG-3'
OPA-03	5'-AGTCAGCCAC-3'
OPA-04	5'-AATCGGGCTG-3'
OPA-05	5'-AGGGGTCTTG-3'
OPA-08	5'-GTGACGTAGG-3'
OPA-11	5'-CAATCGCCGT-3'
OPA-17	5'-GACCGCTTGT-3'
OPA-18	5'-AGGTGACCGT-3'
OPA-06	5'-GGTCCCTGAC-3'
OPA-10	5'-GTGATCGCAG-3'
OPB-01	5'-GTTTCGCTCC-3'
OPB-08	5'-GTCCACACGG-3'
OPAJ-04	5'-GAATGCGACC-3'
OPAJ-14	5'-ACCGATGCTG-3'
OPAJ-17	5'-ACCCCTATG-3'
OPAJ-18	5'-GGCTAGGTGG-3'
OPAJ-19	5'-ACAGTGCCCT-3'
OPAJ-20	5'-ACACGTGGTC-3'
OPAK-14	5'-CTGTCATGCC-3'

Soil nutrient and environmental data collection

Soil nutrient measurements were conducted by collecting soil samples from each research location at a depth of 0-30 cm, which is the active rooting zone for arrowroot. Soil samples were randomly taken from three points within each research plot and then mixed into a single composite sample per locality. These soil samples were then analyzed in the laboratory to determine the nitrogen (N), phosphorus (P), and potassium (K) contents via the Kjeldahl method for nitrogen, the Bray I method for phosphorus, and ammonium acetate extraction for potassium. Additionally, soil pH was measured via a pH meter (Soil Tester Manual Takemura DM 15). Rainfall data were obtained from monthly records (BPS Jember 2024). Air temperature and relative humidity were measured via an automatic hygrometer thermometer equipped with temperature and humidity sensors (UT330). This device was placed in a shaded area at a height of approximately 2 m above the ground to ensure the accuracy of the data collected. Temperature and humidity data were collected continuously during the research period, with hourly recordings to provide a detailed overview of the climate variations in each sub-district. Land management practices, such as fertilizer use, irrigation methods, crop rotation, and pest control, were recorded through interviews with local farmers and field surveys. These data were systematically collected to enable correlational analysis between environmental conditions, management practices, and the genetic and phenotypic variation of arrowroot in different sub-districts. All the data were then scaled to be used as supporting data for explaining the phenomenon of arrowroot diversity observed in Jember.

Data analysis

The collected data were analyzed via various statistical methods to evaluate the distributions of genetic, phenotypic, and quantitative traits across the three sub-districts. Statistical analysis was performed via R and Python software, with the libraries ggplot2, pandas, and Matplotlib used for data visualization in the form of boxplots, heatmaps, and PCA. Boxplot analysis was conducted to illustrate the distribution of these traits within each sub-district, allowing for visual comparison of median values and identification of outliers.

To evaluate the overall variability and identify the main components contributing to the differences observed between sub-districts, principal component analysis (PCA) was employed via the sci-kit-learn library in Python. This PCA helps reduce the dimensionality of the data while retaining significant variability, thus facilitating visual interpretation in two main dimensions. Additionally, heatmap analysis was used to visualize the average values of the three parameters across the sub-districts, utilizing the seaborn and p-heatmap libraries in R, providing a clearer understanding of the relative genetic and phenotypic diversity. This study also conducted a comparative analysis of the environmental factors in each sub-district, such as the soil type, pH, and climatic conditions. These factors were analyzed via PCA to determine their potential influence on the genetic and phenotypic traits of arrowroot.

Additionally, the conservation priority index (CPI) is used to provide an overview of the impact of diverse factors on future conservation potential projections. The calculation of the CPI involves the measurement and evaluation of the genetic, phenotypic, and quantitative diversity of arrowroot in various localities, which is modified from the traditional conservation priority index (Rana et al. 2020). Each locality is analyzed based on the level of genetic and phenotypic diversity, which is then correlated with environmental factors and land management practices.

$$CPI = \frac{G + P + Q}{3}$$

Where:
G : the genetic diversity score,
P : the phenotypic diversity score
Q : the quantitative trait score

Each component (G, P, and Q) is assessed on a scale, typically from 0 to 1, where G reflects the genetic variation within the population of arrowroot in a particular locality. This is determined by analyzing the presence of unique genetic alleles and overall genetic diversity; P indicates phenotypic diversity, which is based on the observed variation in morphological traits such as root color, stem height, and tuber shape within the population. Q measures the variation in quantitative traits, such as tuber weight, tuber length, and leaf size. Localities with high genetic and phenotypic diversity, as well as supportive environmental conditions, receive higher CPI values, indicating a more urgent conservation priority. These data are then plotted as bar graphs to compare conservation priorities.

RESULTS AND DISCUSSION

Genetic, phenotypic, and quantitative variation across sub-districts

The boxplot shows the distribution of the mean values of three parameters, namely, Genetics_Mean, Phenotypic_Mean, and Quantitative_Mean, in three different sub-districts: Wuluhan, Sumbersari, and Arjasa (Figure 2). In the Wuluhan sub-district, the median Genetics_Mean is approximately 0.80, with an IQR between 0.75 and 0.85, accompanied by several outliers below 0.70 and above 0.85 (Table 3). The Phenotypic_Mean in this sub-district has a median of approximately 0.82, with an IQR between 0.78 and 0.84 and a few outliers of approximately 0.20 and 0.80. The Quantitative_Mean in Wuluhan has median of approximately 0.21, with a very small IQR of approximately 0.20, and some outliers below 0.20.

In the Sumbersari sub-district, the median Genetics_Mean is approximately 0.80, with an IQR between 0.78 and 0.82, with some outliers below 0.70 and at approximately 0.90. Phenotypic_Mean has a median of approximately 0.80 with an IQR between 0.77 and 0.84, and a few outliers below 0.70 and at approximately 0.90. The Quantitative_Mean shows a median of approximately 0.19, with a very small IQR of approximately 0.19, and some outliers below 0.20. In the Arjasa sub-district, the median Genetics_Mean is approximately 0.68, with an IQR between 0.65 and 0.83, with some outliers below 0.60 and at approximately 0.90. The Phenotypic_Mean in Arjasa has median of approximately

0.83, with an IQR between 0.78 and 0.86, and a few outliers below 0.70 and approximately 0.90. The Quantitative_Mean shows a median of approximately 0.22, with a very small IQR of approximately 0.20, and some outliers below 0.20.

The Arjasa sub-district exhibited the highest genetic diversity; a wider interquartile range indicates this and the presence of numerous outliers on both sides of the median. Although the average genetic value is slightly lower compared to Wuluhan and Sumbersari, the variability within Arjasa is the most significant because it shows a wider range of genetic expressions. Although the Quantitative_Mean has a small interquartile range across all sub-districts, Wuluhan shows slightly more variability than Sumbersari and Arjasa. However, overall, the variation remains low compared with the other two parameters. Genetics provides the foundational framework for the development of tuber size, shape, and color (Malki et al. 2023b). A phenotype is the result of a complex interaction between genetics and the environment. For example, arrowroot tubers with the genetic potential to grow large may not reach that potential if they are planted in an unfavorable environment (De Leon et al. 2016).

The greatest potential for arrowroot growth is found in the Wuluhan sub-district, based on the highest average genetic value (0.75), which indicates that the tubers in this sub-district have the greatest genetic potential for large growth. However, in the Arjasa sub-district, the phenotypic potential is the highest (0.83), suggesting that despite having lower genetic potential, the environmental conditions in Arjasa may better support the full expression of the phenotype. While Arjasa exhibits greater genetic diversity, no single sub-district demonstrates significantly higher phenotypic potential than others. This indicates that with favorable environmental management, tubers in any sub-district could express similar phenotypic outcomes. Human activities, such as farming practices and land management, interact with environmental factors to influence arrowroot diversity. Future research should explore the interaction of these factors to fully understand how they contribute to genetic and phenotypic variability.

Although the genetic potential for large growth is more prominent in Wuluhan, the similar phenotypic and quantitative potential across the three sub-districts indicates that the expression of physical and quantitative characteristics of arrowroot tubers can be optimized with good environmental management in all regions (Xu 2016; Li et al. 2018). Additionally, by preserving arrowroot's genetic and phenotypic richness, we can ensure that this crop retains its ability to adapt to and thrive in various environmental conditions. A heatmap also represents the variation results. The heatmap shows the average values of the genetic, phenotypic, and quantitative parameters for each sub-district analyzed (Figure 3).

Table 3. Comparison of genetic, phenotypic, and quantitative means across three sub-districts, i.e. Wuluhan, Sumbersari, and Arjasa in Jember District, East Java Province, Indonesia

| Sub-district	Genetics_Mean (Median, IQR)	Phenotypic_Mean (Median, IQR)	Quantitative_Mean (Median, IQR)
Wuluhan	0.80 (0.75-0.85)	0.82 (0.78-0.84)	0.21
Sumbersari	0.80 (0.78-0.82)	0.80 (0.77-0.84)	0.19
Arjasa	0.68 (0.65-0.83)	0.83 (0.78-0.86)	0.22
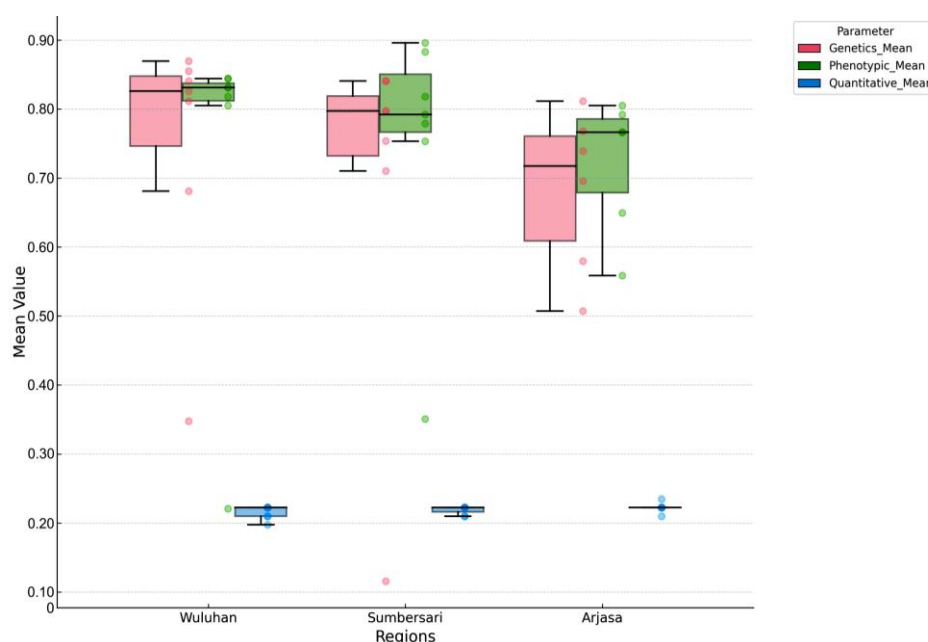
Figure 2. Boxplot of genetic, phenotypic, and quantitative values of arrowroot tubers in three research locations, i.e. Wuluhan, Sumbersari, and Arjasa in Jember District, East Java Province, Indonesia

Wuluhan has the highest average genetic value (0.75), followed by Sumbersari (0.73) and Arjasa (0.71). The heatmap shows that Wuluhan has the highest average genetic value (0.75), followed by Sumbersari (0.73) and Arjasa (0.71). Although Wuluhan has the highest average, the boxplot shows that the distribution of genetic diversity is wider in Arjasa, indicating a wider distribution of genetic traits even though the average value is lower. The three sub-districts also have the same average quantitative value (0.07), indicating that the quantitative diversity is very low in these sub-districts. This heatmap confirms that Wuluhan has the highest genetic potential, while the average phenotypic and quantitative values are the same across all three sub-districts. This suggests that although there are differences in genetic diversity, phenotypic expression, and quantitative variation are relatively consistent across sub-districts, indicating that environmental factors might have a more uniform influence on these traits or that the genetic potential for phenotypic expression is similar despite variations in genetic diversity. This may indicate that environmental factors or agricultural practices are similar in all three sub-districts (Gratani 2014; Großkinsky et al. 2015; Liu et al. 2017). The PCA graph shows the distribution of samples from the three sub-districts (Arjasa, Sumbersari, and Wuluhan) based on PCA component 1 (27.54%) and PCA component 2 (12.32%, see Figure 4).

The Arjasa sub-district (red) appears to be more spread out toward the positive components of PC 1 and PC 2, indicating that samples from Arjasa have greater variation in these two components; this confirms the findings analyzed with the boxplot (Figure 2). The Sumbersari sub-district (green) tends to be centered, showing a narrower distribution around the mean values for both PCA components. Finally, the Wuluhan sub-district (blue) has a more concentrated distribution on the negative side of PC 1, indicating that samples from this sub-district have less variation than those from Arjasa.

There is an overlap between the Sumbersari and Wuluhan sub-districts, suggesting that some samples from these two sub-districts share similarities in principal component 1 and principal component 2 (Figure 4). However, Sumbersari also has a unique sample that does not overlap with other sub-districts, indicating that at least one sample from Sumbersari exhibits distinct characteristics. This uniqueness may be due to specific genetic or environmental factors in the Sumbersari sub-district that differentiate this sample from others. Arjasa, similarly, has some samples that do not overlap with other sub-districts, indicating that samples from Arjasa have more unique characteristics in these two components.
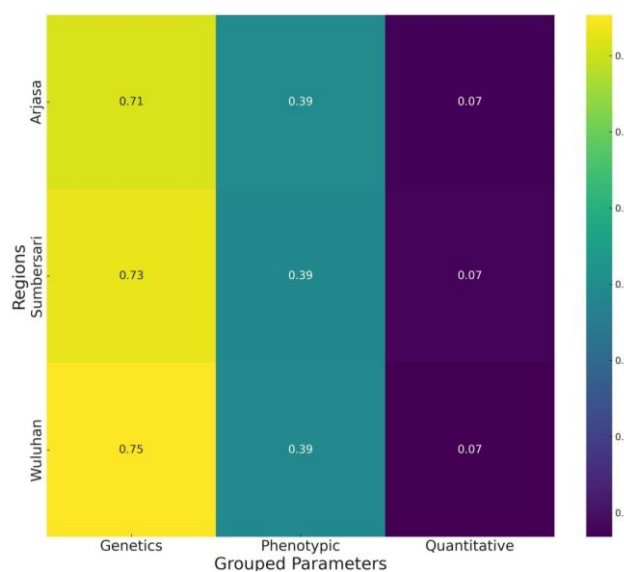
Figure 3. Genetic, phenotypic, and quantitative variation of arrowroot tubers in Wuluhan, Sumbersari, and Arjasa sub-districts, Jember District, East Java Province, Indonesia
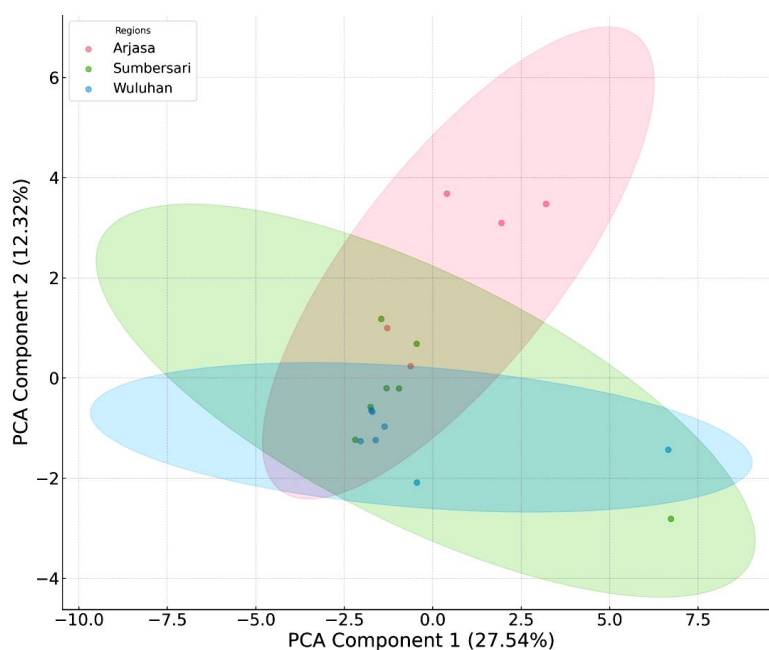
Figure 4. PCA Distribution of the arrowroot samples in Wuluhan, Sumbersari, and Arjasa sub-districts, Jember District, Indonesia

PCA analysis highlights genetic and phenotypic diversity across arrowroot samples

The PCA results clearly revealed that the samples from Arjasa presented the highest genetic and phenotypic diversity, which was consistent with the previous boxplot results. The broader distribution of the Arjasa samples in the PCA graph indicates this. The samples from Sumbersari and Wuluhan are more closely related to each other than those from Arjasa, as indicated by the significant overlap between these two sub-districts in the PCA graph. This finding aligns with the boxplot results showing lower diversity in the Sumbersari and Wuluhan sub-districts. The low quantitative variation across all sub-districts (as seen in the boxplot) is also reflected in the PCA graph, where there are no significant differences along PC 2, which might represent smaller quantitative variations.

Arjasa may have a greater genetic variation than is not found in Sumbersari and Wuluhan; this could include differences in genetic alleles or the presence of unique genes not found in other sub-districts (Shintu et al. 2016). The environmental conditions in Arjasa are quite different and include unique soil types, climates, and agricultural practices. These factors can lead to significant differences in phenotypic expression and plant adaptation (Figure 4) (Martre et al. 2015; Xu 2016). Plants in Arjasa have adapted to very specific environmental conditions, resulting in genotypes and phenotypes with unique variants. Differences in agricultural practices, such as fertilization methods, pesticide use, or crop rotation, can affect genetic and phenotypic variation over the long term, both consistently and specifically (Sadras and Denison 2016).

The PCA (principal component analysis) graph with bubble plots shows the distribution of samples from various locations on the basis of two main components: PCA component 1 (26.48%) and PCA component 2 (11.29%, Figure 5). The bubble size represents the data density or the number of samples from each location. Kebonsari is located far in the upper right of the graph, indicating that samples from this location have very different characteristics than those from other locations in terms of the two main components analyzed. Glundengan is also located far from the lower right, indicating significant differences in PCA Component 1 compared with the other locations. Arjasa and Kemuning Lor are positioned on the right side of the graph, indicating that these two locations have some unique characteristics but are still within a closer range than Kebonsari and Glundengan.

Most locations, such as Tanjung Rejo, Ampel, Bukit Sari, Sukoharjo, and others, are clustered in the lower left part of the graph, with significant overlap. This indicates that samples from these locations have similar characteristics in terms of the two main components. Other locations, such as Taman Sari and Arjasa, slightly overlap with other locations, suggesting that while they share some similarities, these locations also exhibit distinct characteristics. The bubble size in the graph reflects the data density or the number of samples from each location. Larger bubbles, such as in Kebonsari, indicate a higher number of samples, while smaller bubbles, such as in Glundengan, indicate fewer samples. The high data density in several locations in the lower left part of the graph indicates that these locations not only share similar characteristics but also contain a larger number of samples.

When looking at the overall sub-districts Sumbersari, Wuluhan, and Arjasa it becomes clear that the overlap between Sumbersari and Wuluhan in both principal components 1 and 2 demonstrates that these sub-districts share similar characteristics. However, the more unique characteristics of Arjasa are evident from its broader distribution and the distinct placement of its locations, as reflected in the PCA graph. This is consistent with the genetic and phenotypic diversity previously highlighted in the boxplot results. Additionally, the variation in bubble size across the 20 sites within these sub-districts further supports the idea that locations like Kebonsari have a higher sample density, contributing to their distinct placement in the PCA, while others, like Glundengan, stand out with fewer samples, emphasizing their unique characteristics within the broader sub-districtal patterns.

Kebonsari has distinct environmental conditions, such as unique soil types, climates, and humidities (Figure 5). These factors could lead to significant genetic and phenotypic differences as the plants have adapted to the unique environmental conditions in that area. However, this diversity may also be influenced by extreme environmental differences, including areas with specific topographies (Forsman 2015; Chevin and Hoffmann 2017). The agricultural methods used in Kebonsari and Glundengan also differ, such as fertilizer application, irrigation techniques, and specific pest control practices, which ultimately support their unique phenotypes (Xu 2016; Wang et al. 2021).
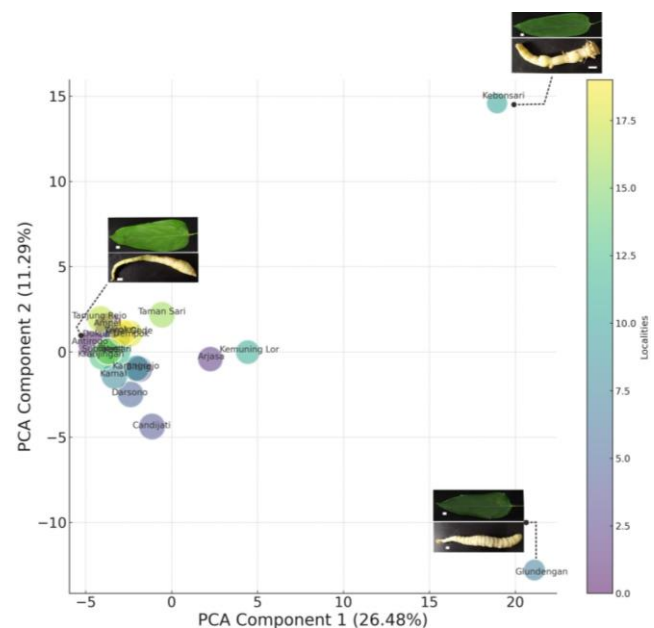
Figure 5. PCA graph representing scoring density in testing sites

## Conservation priorities and genetic diversity in arrowroot populations

Given the high degree of genetic variation and uniqueness, it is essential to protect and conserve arrowroot populations in Kebonsari. This can be achieved through gene banks or conservation gardens. The high genetic diversity in Kebonsari can be used in breeding programs to develop arrowroot varieties that are more resistant to various environmental conditions, especially in response to ongoing climate change (Kaushal et al. 2016; Korres et al. 2016; Raza et al. 2019). Although the sample size is small, it is important to protect the unique genes from Glundengan. Conservation programs focused on the collection and preservation of genetic material are crucial for the sustainability of these genes. The unique genes from both areas could be integrated into breeding programs to increase genetic diversity and plant adaptation (Huang and Han 2014).

Antirogo has the highest CPI value, approximately 0.440, indicating the highest conservation priority among the other locations (Figure 6). These findings suggest that Antirogo has significant genetic, phenotypic, and quantitative diversity, making it a key area for arrowroot conservation efforts in Jember. With a fairly high population density, Antirogo faces significant environmental threats, such as land-use conversion and pollution, making conservation efforts urgent (BPS Jember 2024). The localities of Lojejer and Ampel have CPI values of approximately 0.435 each, indicating that these areas also require significant conservation attention. In contrast, Kebonsari has the lowest CPI value, approximately 0.235. To increase its conservation priority, conservation efforts in Kebonsari can focus on increasing genetic and phenotypic diversity. Overall, these results highlight the importance of conservation in Antirogo as the top priority, followed by Lojejer, Ampel, and other locations.

Locations such as Wirolegi, Tegal Gede, Tanjung Rejo, Taman Sari, and Sumbersari have higher CPI values, approximately equal to or greater than 0.4. Arrowroot stems in these locations appear yellow, indicating that they have a higher conservation priority than other locations. Locations such as Kemuning Lor, Kebonsari, Karangrejo, Kamal, Glundengan, Dukuh Dempok, and Darsono have medium CPI values, approximately 0.3-0.4. The arrowroot stems there range from green to blue, indicating a moderate conservation priority. Locations such as Ampel, Antirogo, Arjasa, Biting, and Candijati have lower CPI values, approximately 0.3 or less. The arrowroot stems tended to be more purple in those locations, indicating a lower conservation priority.

Table 3 and Figure 4 reveal that the Arjasa sub-district exhibits the highest genetic diversity, as reflected by its broader distribution of genetic traits. This suggests that Arjasa's environmental conditions support greater variability, making it a crucial area for understanding the genetic landscape of arrowroot. However, despite Arjasa's rich genetic diversity, the focus of conservation efforts shifts to other sub-districts, as highlighted in Figure 5. Sub-districts such as Antirogo, Lojejer, Ampel, Wirolegi, Tegal Gede, Tanjung Rejo, Taman Sari, and Sumbersari-though not part of Arjasa-are identified as having higher Conservation Priority Index (CPI) values. This indicates that these areas face more immediate environmental or anthropogenic threats, making them critical targets for conservation. While Arjasa remains a valuable genetic reservoir, these sub-districts with higher CPI values require urgent attention to safeguard their existing genetic and phenotypic potential. By focusing conservation efforts on these vulnerable areas, we can help maintain the genetic integrity of arrowroot populations across various regions. The genetic diversity observed in sub-districts like Arjasa suggests the need for targeted conservation strategies, including both in-situ and ex-situ conservation efforts. These approaches, combined with controlled cross-breeding, can enhance crop resilience to environmental stresses and climate variability.
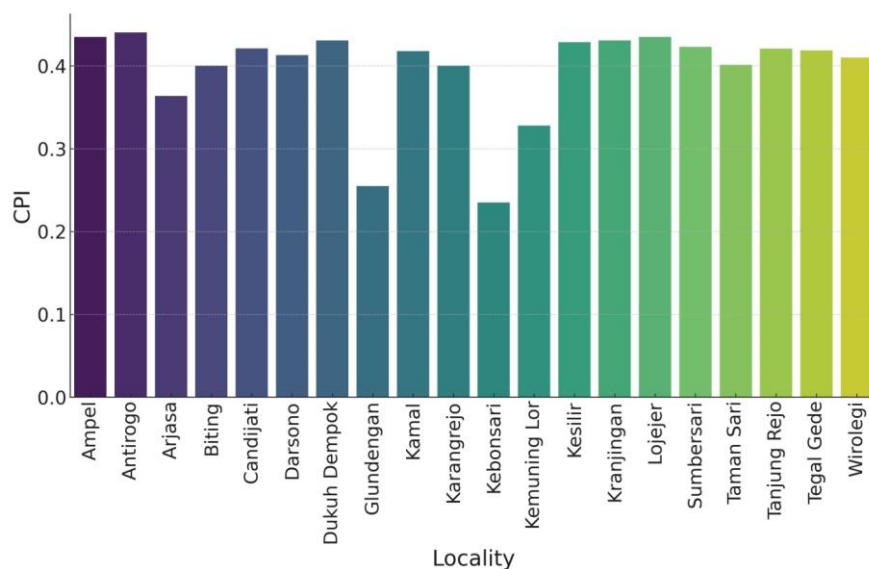
**Figure 6.** Bar graph of CPI analysis results in sampling site locations

Although there is no significant evidence of a correlation between physical environmental factors and the characteristics of arrowroot, a qualitative analysis based on PCA indicates that significant variability in the genotypic, phenotypic, and quantitative data of arrowroot in the three study sub-districts (Arjasa, Sumbersari, and Wuluhan) is influenced by environmental factors such as soil pH, nitrogen, phosphorus, and potassium content, as well as climatic conditions such as rainfall and temperature (Figure 7). Compared with the Sumbersari and Wuluhan sub-districts, the Arjasa sub-district presented greater genetic and phenotypic diversity, as evidenced by the broader distribution of the main PCA components. On the other hand, quantitative variation tends to be lower and more uniform across all sub-districts, reflecting the more limited influence of genetic and environmental factors on tuber size and shape.

These findings suggest that the Arjasa sub-district has more diverse genetic and phenotypic potential, possibly due to more varied or unique environmental conditions, such as differences in soil type and more diverse agricultural practices. In contrast, the Sumbersari and Wuluhan sub-districts tend to have more uniform environmental characteristics, resulting in more limited genetic and phenotypic variability. These data support the importance of maintenance strategies tailored to specific environmental conditions to optimize the genetic and phenotypic potential of arrowroot. In the long term, preserving genetic diversity in sub-districts such as Arjasa can provide benefits for plant breeding and conservation programs aimed at enhancing the resilience and adaptation of plants to various environmental conditions (Bueren et al. 2018; Roberts and Mattoo 2018).

Although Table 3 and Figure 4 show that Arjasa has the highest genetic diversity, Figure 5 emphasizes the importance of conservation in sub-districts outside Arjasa, such as Antirogo, Lojejer, Ampel, Wirolegi, Tegal Gede, Tanjung Rejo, Taman Sari, and Sumbersari, due to their higher Conservation Priority Index (CPI) values. While Arjasa holds significant genetic potential, these sub-districts face more immediate environmental or anthropogenic threats, making them critical targets for conservation efforts. Therefore, to maintain the high genetic diversity in Arjasa and increase it in Sumbersari and Wuluhan, distinct conservation strategies are required. In Arjasa, where genetic diversity is highest, preserving this diversity is crucial through the promotion of sustainable agricultural practices, such as crop rotation and organic fertilization, while also establishing in-situ conservation areas to protect native genotypes in their natural environment. Engaging local farmers through community-based programs can further enhance these efforts and ensure the long-term preservation of genetic resources. For Sumbersari and Wuluhan, where genetic diversity is lower, introducing new genetic material from sub-districts like Arjasa through controlled cross-breeding and agroforestry systems can help boost genetic diversity.
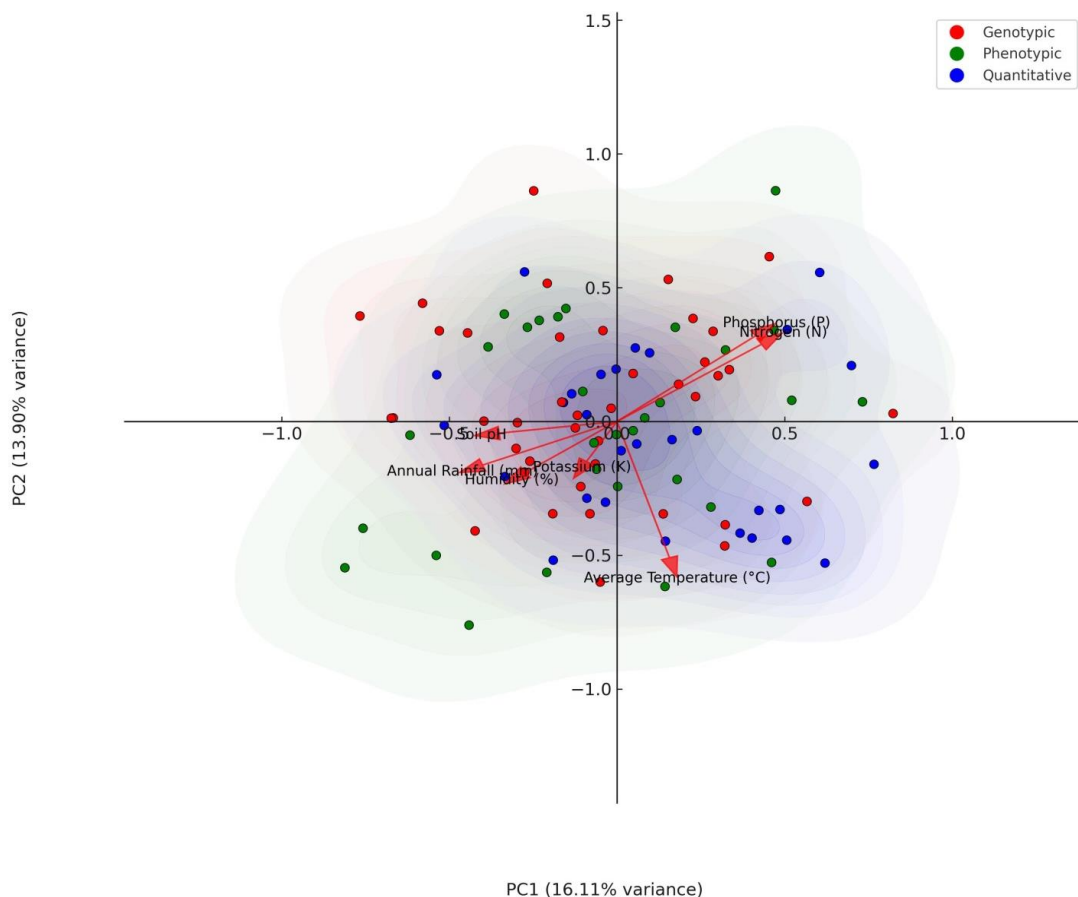
Figure 7. PCA graph showing the influence of physical environmental factors and management on the three main parameters

Ex-situ conservation efforts, such as seed banks and breeding programs, can also play a key role in developing more resilient varieties that can be reintroduced into these sub-districts. These combined strategies aim to strengthen the adaptability and resilience of arrowroot crops across various environmental conditions while addressing the immediate conservation needs of vulnerable sub-districts outside Arjasa. While the CPI provides valuable insights into conservation needs, it may overlook other crucial factors such as genetic diversity and local farming practices. Future studies should incorporate these additional metrics to create a more holistic conservation framework.

In conclusion, the genetic and phenotypic diversity of arrowroot (*Maranta arundinacea* L.) varied considerably across three agroclimatic zones in Jember, Indonesia, with Arjasa having the highest levels based on medians for Genetics_Mean=0.75 (IQR: 0.7-0.83) and Phenotypic_Mean=0.83 (IQR: 0.78-0.86). Kebonsari has the lowest conservation priority index (CPI) value of 0.235, indicating that it was a moderate priority for conservation effort compared with Antirogo, with the highest CPI value of 0.440, indicating high priority for management efforts in this area. Our study highlights the necessity of conservation programs for ensuring both high allelic and phenotypic diversity in arrowroot, especially in the face of future environmental changes with increasing global climate change. The researcher suggests conservation efforts focused on establishing in-situ conservation areas and ex-situ methods like seed banks to preserve genetic and phenotypic diversity in arrowroot. Additionally, promoting sustainable agricultural practices and introducing new genetic material through controlled breeding can enhance crop resilience against environmental changes and climate challenges. In addition to the fact that these findings increase local food security, they can lead us to develop a broader spectrum of varieties that are more suitable for different environments.

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