

Research Article



Diversity and Selection of Cotton Germplasm Based on Morpho-Agronomic Character Performance

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ABSTRACT

Cotton is a plant that produces natural fibers of high economic value. Developing a new superior variety of cotton is expected to support fiber demand in Indonesia. The purpose of this study was to determine the diversity of morpho-agronomic characters and obtain secondary characters and cotton accessions that support the development of a new superior cotton variety. The genetic material used was 266 cotton accessions from the ISFCRI germplasm collection. Primary data of the 16 quantitative morphological characters were observed and analyzed using descriptive data, correlation analysis, path analysis, scatterplot, and grouping. The fiber length character has a very low diversity with a value of 0.01, and the plant height character has the highest variance, which is 637.42. The lint percentage with a variance value of 9.37 is influenced by duplicate epistasis genes and controlled by many genes. The fiber's fineness character has a positive and very significant correlation value to the character of the lint percentage. It has fairly high direct and total effect values of 0.156 and 0.134, respectively. Four accession groups can potentially support developing a new superior variety; accession CEA N 268, CEDIX, or L21999-10-71 can be used as the parent of new superior cotton varieties.

1. Introduction

Cotton is one of the producers of natural fibers, which has an important role in the textile industry and affects a country's economic, agricultural, and industrial sectors. This plant is classified as an annual plant. It is one of the main commodities in the textile and fabric industry, contributing to the industrialization sector as well as economic and social transformation in many countries (Kedisso *et al.* 2023). In other countries, 90 varieties of upland cotton, 3 of Egyptian cotton, and 4 varieties cultivated in Mozambique and Brazil (Franca *et al.* 2022). Indonesia is the 6th

largest cotton-importing country globally and the 9th cotton consumer (USDA 2022). According to data on agriculture in Indonesia, the cotton development area in 2019 decreased from 1620 ha to 1281 ha in 2021 (Dirjenbun 2021). That data shows that cotton production has decreased from 280 tons to 191 tons.

Biotic and abiotic influences dominate the factors causing the decline in cotton production. Several environmental factors that can reduce cotton production are rainfall that does not occur during planting time, high-temperature winter from the beginning to the flowering stage, late cotton harvesting, which also results in a decrease in the harvest area, the emergence of cotton leaf curl disease, soil systems, air use constraints, pest, and insect outbreaks (Soomro *et al.* 2020). Efforts to increase cotton production in Indonesia include

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optimizing existing land and preparing new areas specifically for cotton cultivation, using cultivation and fiber processing technology, and improving adequate storage and transportation facilities. Additionally, developing cotton varieties that are more resistant to pests and diseases and have high productivity can further enhance production (Razaq *et al.* 2018).

Problems in fulfilling cotton fiber demands can be solved by developing high-yielding varieties with production potential. The development of superior varieties is obtained through the selection stage on germplasm or by selecting segregated populations with a large genetic diversity. Estimating genetic diversity can be done through quantitative analysis (biometrics), which estimates the genetic model and the number of controlling genes and decomposes genetic diversity into additive, dominant, and epistatic variations. The plant breeding method is crucial for generating useful genetic variations and identifying desired superior traits. Conventional breeding results can significantly enhance cotton fiber production and fiber quality (Shahzad *et al.* 2022).

Cotton genotype selection can be done directly or indirectly. Direct selection is made based on the character of the yield components. Meanwhile, indirect selection requires other agronomic characteristics positively correlated with yield characteristics. In this study, the selection is only based on yield component characteristics. The effectiveness of selection decreases because the effect of environmental factors is greater than genetic factors (Yamin 2014). Several other morphological or agronomic characteristics can also be observed, especially those related to the yield value. Furthermore, the value of the correlation coefficient was calculated to determine the degree of closeness relationship between the yield and other characteristics observed.

The interrelationships between various characters determine the selection characteristics in genetic improvement to increase yields (Erande *et al.* 2014). A linear relationship between observational characters is also used in correlation analysis. Estimating correlation coefficient values mostly shows the relationships between different characters but does not provide information about cause and effect. Therefore, it is necessary to carry out a path analysis to identify the selection index (Pujer *et al.* 2014). Path analysis of precise information on direct and indirect effects is to determine the most influential characteristics to be used as selection criteria in breeding programs (Reddy *et al.* 2015). In addition, the complex

relationship between observed characters can be explained with path analysis (Seesang *et al.* 2013).

The research related to plant breeding requires character selection to produce secondary characters and individual selection in determining hybridization parents who have distant kinship relationships through grouping based on two variables using a scatter plot. A scatter plot can support collecting large plot data through ranking, filtering, and searching functionalities shown in the four plot areas (Shao *et al.* 2016). Grouping individual samples can use an approach by measuring the distance between a pair of samples assigned to the same cluster.

The research results on the correlation of agronomic characteristics, such as plant height and boll weight, with technology characteristics, such as fiber length and fiber strength, indicate that fiber strength, uniformity, and elongation influence fiber length and can be used as direct and indirect selection criteria for cotton genotypes (Farias *et al.* 2016). Boll weight showed a close relationship with the yield at the genotype and phenotype levels. Path analysis shows that all characteristics directly and positively affect the results except the bolls per plant (Farooq *et al.* 2015). The main panicle grain weight trait shows a positive and highly significant correlation coefficient with yield component traits, making it a suitable selection criterion for producing high-yielding wheat (Yamin 2014). Meanwhile, the research results by RS *et al.* (2022) showed that the characteristic of 50% flowering has the best indirect influence on dry wood weight, so that this character can be used as a secondary selection character. This research aims to identify the diversity of morpho-agronomic characters, obtain secondary selection characters, and find potential cotton accessions to support the development of new superior cotton varieties.

2. Materials and Methods

2.1. Study Area and Plant Materials

The study was conducted at the Agricultural Instruments Standardization Agency for Sweetener and Fiber Crops was established in July 2022; it is a continuity from the Indonesian Sweetener and Fiber Crops Research Institute (ISFCRI) in Malang, East Java, conducted from September 2021 to April 2022. The research location is at 7.907439 South Latitude and 112°37'27" East Longitude, Altitude 515 meters above sea level, climate type D (moderate), soil type Gleysol/Inceptisol, and rainfall 1,500 mm/year. The

genetic materials used were 266 cotton accessions from the Indonesian Instruments Standardization Testing Center for Sweetener and Fiber Crops germplasm collection.

2.2. Culture Practice and Measurement of Plant Variables

Primary data were collected from 266 cotton accessions that were previously planted in the field. Each cotton accession was planted in a single row 10 meters long with a spacing of 150 cm × 25 cm. Observations were conducted by collecting quantitative data from 10 randomly selected sample plants per accession.

The Quantitative data of 16 morphological characters observed in the generative growth phase were X1 = plant height, X2 = monopodial branches number, X3 = date to the first square, X4 = sympodial branches number, X5 = date to the first flower opening, X6 = date to the first boll opening, X7 = the number of bolls per plant, X8 = boll weight, X9 = seed index, X10 = fiber length, X11 = fiber fineness, X12 = fiber strength, X13 = fiber elongation, X14 = fiber uniformity, X15 = insect susceptibility (*Amrasca biguttula*) and Y = content of lint (UPOV 2001).

The observation of accession susceptibility to the insect *A. biguttula* was conducted by evaluating leaf damage scores due to pest attacks at 60 and 90 days, based on (Indrayani *et al.* 2009) method. The scoring criteria are as follows: a score of 0.1-1.0 indicates no signs of attack (resistant), a score of 1.1-2.0 indicates yellowing and slight curling of leaf edges (moderately resistant), a score of 2.1-3.0 indicates yellowing and curling of leaves (moderately susceptible), and a score of 3.1-4.0 indicates leaves turning reddish-brown and then falling off (susceptible).

2.3. Data Analysis

The observed data were analyzed for correlation, path analysis, scatter plot, and grouping (30 cotton accessions from selected 10% based on the character of the lint percentage and fiber fineness). Data was grouped using cluster analysis based on the most significant score from Principal Component Analysis (PCA) to obtain main components that can retain some of the information. For cluster analysis, the Average linkage method and the previously obtained Euclidean distance matrix can be used (Busanello *et al.* 2020). Correlation analysis, path analysis, scatterplot, skewness, gene action, kurtosis value, and graphic form of cotton accession distribution

and cluster analysis were performed using Minitab software version 14.

Skewness is a statistic used to describe the distribution of data, a measure of symmetry, or, more precisely, whether it is skewed to the left, right, or symmetrical, and it can be used to show the action of genes that control a character. In comparison, kurtosis is a measure of the peakedness of distribution and is used to describe whether the distribution of data tends to be flat or sharp (Mishra *et al.* 2019).

According to Roy's assessment (2001), if the skewness is 0, then the character is controlled by additive gene action; the skewness < 0 indicates additive gene action with duplicate epistasis, while skewness > 0 indicates additive gene action with complementary epistasis. Meanwhile, based on the kurtosis criterion, if kurtosis is negative, the shape of the distribution graph is platykurtic, and many genes control the character; in positive kurtosis, the graph is leptokurtic indicating that few genes control the character.

Correlation analysis between variables was carried out by calculating the value of a simple linear correlation coefficient based on the formula:

$$r = \frac{\sum X_1 X_2}{\sqrt{(\sum X_1^2)(\sum X_2^2)}} \quad (\text{Singh and Chaudhary 1978})$$

r : correlation coefficient. X_1 and X_2 are the means of the observed variables. Path analysis was carried out based on the following simultaneous equation:

$$\begin{bmatrix} r_{11} & r_{12} & \dots & r_{1p} \\ r_{21} & r_{22} & \dots & r_{2p} \\ \vdots & \vdots & \ddots & \vdots \\ r_{p1} & r_{p2} & \dots & r_{pp} \end{bmatrix} \begin{bmatrix} C_1 \\ C_2 \\ \vdots \\ C_p \end{bmatrix} = \begin{bmatrix} r_{1y} \\ r_{2y} \\ \vdots \\ r_{py} \end{bmatrix}$$

$R_x \qquad \underline{C} \qquad R_y$

R_x is the inter-independent variable correlation matrix in the multiple regression model with p independent variables, a matrix with elements r_{xixj} ($i, j = 1, 2, \dots, p$). C is the path coefficient vector, which shows the direct effect of each standardized independent variable. Z_i on the dependent variable (standard regression coefficient value). R_y is the correlation coefficient vector between the independent variables X_i . $i = 1, 2, \dots, p$; and the independent variable y . The matrix equation can be determined by the path coefficient vector C as follows:

$$C = r_x^{-1} r_y$$

$$\begin{bmatrix} r_{11} & r_{12} & \dots & r_{1p} \\ r_{21} & r_{22} & \dots & r_{2p} \\ \cdot & \cdot & \dots & \cdot \\ \cdot & \cdot & \dots & \cdot \\ r_{p1} & r_{p2} & \dots & r_{pp} \end{bmatrix} \begin{bmatrix} C_1 \\ C_2 \\ \cdot \\ \cdot \\ C_p \end{bmatrix} = \begin{bmatrix} r_{1y} \\ r_{2y} \\ \cdot \\ \cdot \\ r_{py} \end{bmatrix} \quad \text{(Singh and Chaudhary 1978)}$$

$R_x \quad C \quad R_y$

r_x^{-1} is the inverse matrix of R_x , and R_y is a vector correlation coefficient between the independent variable X and the dependent variable Y. The direct effect of standardized independent variables on the dependent variable Y is measured by the coefficient C_i . The indirect effect of the independent variable Z_i on the dependent variable Y through the independent variable Z_j (in the model) is measured by $C_j \cdot r_{ij}$.

The path analysis model explains the error effect. The effect that a model cannot explain is calculated as the effect of the error or residual, which is measured by the formula:

$$h(\text{error}) = \sqrt{1 - C^{-1}} \quad \text{(Singh and Chaudhary 1978)}$$

3. Results

3.1. Genetic Diversity Estimation of Germplasm Cotton Accessions

The observation morpho-agronomic result of 266 cotton germplasm showed the variance value from the lowest to the highest (Table 1). In probability theory

and statistics, the variance or the variety of a random variable (or probability distribution) measures how far a set of numbers is spread out. The zero variance indicates that all values are equal. Table 1 shows low coefficient variance (0.01-9.37) on the monopodial branches number, sympodial branches number, boll weight, seed index, fiber length, fiber fineness, elongation, insect susceptibility, and content of lint. The medium variance (15.34-18.32) was shown by the number of bolls per plant and fiber strength; meanwhile, the variance that is classified as large (37.55-637.42) is shown by the characteristics of plant height, date to the first square opening, date to the first flower opening, date to the first boll opening, and fiber uniformity.

3.2. Gene Action Estimation of Morpho-Agronomic Character

Estimation of gene action controlling the agronomic character of cotton germplasm in segregated populations was obtained using descriptive statistics based on the skewness and kurtosis values. Table 2 shows the characteristics of plant height, monopodial branches, and date to the first square opening, sympodial branches, date to the first flower opening, date to the first boll opening, number of bolls per plant, seed index, fiber length, fiber fineness, fiber strength, and fiber elongation have skewness values > 0 . The characteristics of boll weight, fiber uniformity, insect susceptibility, and content of lint have skewness values < 0 . These three characters

Table 1. Descriptive value of the morpho-agronomic character of 266 cotton accessions

Production characteristic	Interval	Mean	Standard deviation	Median	Variance
Vegetative phase					
Plant height	51.00-233.00	110.00	25.25	108.00	637.42
Monopodial branches	1.00-6.00	3.05	0.84	3.00	0.71
Generative phase					
Date to the first square	35.00-135.00	38.58	6.66	38.00	44.40
Sympodial branches	7.00-25.00	14.73	2.86	14.00	8.17
Date to the first flower opening	49.00-160.00	58.91	7.02	58.00	49.21
Date to the first boll opening	102.00-182.00	113.23	6.13	115.00	37.55
Yield					
Number of bolls per plant	1.00-28.00	8.58	3.92	8.00	15.34
Boll weight	1.40-7.70	5.16	0.98	5.10	0.96
Seed index	4.64-13.70	9.77	1.35	9.74	1.84
Fiber length	0.95-1.47	1.20	0.07	1.19	0.01
Fiber fineness	3.10-7.90	4.37	0.58	4.40	0.34
Fiber strength	18.20-52.60	30.83	4.28	30.60	18.32
Fiber elongation	4.20-8.50	6.07	0.51	6.00	0.26
Fiber uniformity	44.40-89.70	84.05	7.12	85.40	50.74
Insect susceptibility	0.90-4.00	2.65	0.68	2.60	0.46
Content of lint	24.00-41.10	34.04	3.06	34.15	9.37

Table 2. Skewness, gene action, kurtosis value, and graphic form of cotton accession distribution

Character	Skewness	Gene action	Kurtosis	Description
Plant height	0.63	Additive + Complementary epistasis	1.42	Controlled by many genes
Monopodial branches	0.14	Additive + Complementary epistasis	0.37	Controlled by many genes
Date to the first square	12.15	Additive + Complementary epistasis	169.75	Controlled by a few genes
Sympodial branches	0.70	Additive + Complementary epistasis	1.09	Controlled by many genes
Date to the first flower opening	11.57	Additive + Complementary epistasis	164.34	Controlled by a few genes
Date to the first boll opening	6.23	Additive + Complementary epistasis	64.81	Controlled by a few genes
Number of bolls per plant	1.21	Additive + Complementary epistasis	3.14	Controlled by a few genes
Boll weight	-0.23	Additive + Duplicate epistasis	0.71	Controlled by many genes
Seed index	0.10	Additive + Complementary epistasis	0.50	Controlled by many genes
Fiber length	0.20	Additive + Complementary epistasis	1.45	Controlled by many genes
Fiber fineness	0.77	Additive + Complementary epistasis	4.29	Controlled by a few genes
Fiber strength	0.47	Additive + Complementary epistasis	2.99	Controlled by many genes
Fiber elongation	0.53	Additive + Complementary epistasis	2.92	Controlled by many genes
Fiber uniformity	-4.98	Additive + Duplicate epistasis	24.95	Controlled by a few genes
Insect susceptibility	-0.08	Additive + Duplicate epistasis	-0.45	Controlled by many genes
Content of lint	-0.15	Additive + Duplicate epistasis	-0.11	Controlled by many genes

(boll weight, insect susceptibility, and content of lint) have a near-normal distribution and are controlled by many genes with additive gene action and duplicate epistasis.

3.3. Agronomic Character Correlation to the Content of Lint

Plant height character (Table 3) was significantly and positively correlated with several growth characteristics, including sympodial branches and age at first flower opening. Apart from that, plant height also had a significant and positive correlation with the characteristics of yield components, including fiber strength and fiber percentage, indicating that an increase in plant height can also increase the ability of plants to produce photosynthate, which is translocated to plants that need it to impact plant growth and production directly. The number of bolls per plant characteristic was significantly and positively correlated with the characters of plant height ($r = 0.482$), monopodial branches ($r = 0.149$), sympodial branches ($r = 0.484$), fiber fineness ($r = 0.144$), and fiber strength ($r = 0.130$). However, the number of bolls per plant character was negatively correlated with the attacking character of insect susceptibility ($r = -0.439$). The boll weight character was significantly and positively correlated with the seed index character ($r = 0.436$), meaning that each increase in the boll weight caused an increase in the seed index character.

The fiber length character was significantly and positively correlated with the plant height characters ($r = 0.175$), date to the first boll opening ($r = 0.195$), fiber strength ($r = 0.518$), fiber uniformity ($r = 0.307$),

and insect susceptibility ($r = 0.122$). However, the fiber length was significantly and negatively correlated with the fiber fineness character ($r = 0.149$), suggesting that increasing the fiber length decreased the level of cotton fiber fineness. Fiber fineness character was significantly and positively correlated with the plant height ($r = 0.194$), sympodial branches ($r = 0.111$), number of bolls per plant ($r = 0.144$), and content of lint ($r = 0.210$). However, it was negatively correlated to the seed index ($r = -0.149$), fiber length ($r = -0.141$), fiber strength ($r = -0.238$), and insect susceptibility ($r = -0.237$).

The insect susceptibility character was significantly and negatively correlated with plant height ($r = -0.344$), monopodial branches ($r = -0.184$), sympodial branches ($r = -0.416$), number of bolls per plant ($r = -0.439$), fiber fineness ($r = -0.237$); however, it was significantly and positively correlated with the fiber length character ($r = 0.122$). In this characteristic, a negative correlation means that the higher insect attacks, especially cotton leafhopper (*Amrasca biguttula*), can inhibit plant growth and production and reduce the quality of cotton fiber. This can suggest that there are cotton accessories that are morphologically intolerant to leafhopper insect attacks.

3.4. Path Analysis Content of Lint on Cotton Germplasm Accessions

The characters that are positively and significantly correlated with the content of the lint character in correlation analysis are described in Table 4. The path analysis shows that six agronomic characters have the highest direct and total influence, namely plant height

Table 3. Matrix of correlation analysis on morpho-agronomic characters of cotton germplasm

Characteristics	Plant height	Monopodial branches	Date to the first square	Sympodial branches	Date to the first flower opening	Date to the first boll opening	Number of bolls per plant	Boll weight	Seed index	Fiber length	Fiber fineness	Fiber strength	Fiber elongation	Fiber uniformity	Insect susceptibility
Monopodial branches	0.290**														
Date to the first square	0.115ns	-0.022ns													
Sympodial branches	0.533**	0.111ns	-0.129*												
Date to the first flower opening	0.248**	0.072ns	0.929**	-0.043ns											
Date to the first boll opening	0.198**	0.053ns	0.807**	-0.074ns	0.793**										
Number of bolls per plant	0.482**	0.149**	0.022ns	0.484**	0.112ns	-0.043ns									
Boll weight	-0.075ns	0.049ns	-0.063ns	-0.190**	-0.089ns	-0.133*	-0.093ns								
Seed index	-0.180**	-0.062ns	-0.040ns	-0.084ns	-0.098ns	-0.025ns	-0.046ns	-0.436**							
Fiber length	0.175**	0.084ns	0.095ns	0.040ns	0.117ns	0.195**	-0.000ns	-0.006ns	0.052ns						
Fiber fineness	0.194**	0.063ns	0.043ns	0.111**	0.087ns	0.112ns	0.144*	-0.092ns	-0.149*	-0.141*					
Fiber strength	0.268**	0.120ns	0.114ns	0.257**	0.144*	0.176**	0.130*	-0.135*	-0.000ns	0.518**	-0.238**				
Fiber elongation	0.001ns	0.126*	0.023ns	0.112ns	0.051ns	-0.003ns	0.073ns	-0.035ns	-0.049ns	-0.070ns	0.021ns	0.183**			
Fiber uniformity	0.026ns	0.141*	0.048ns	0.038ns	0.066ns	0.094ns	-0.039ns	-0.042ns	-0.007ns	0.307**	0.056ns	0.469**	0.233**		
Insect susceptibility	-0.344**	-0.184**	-0.016ns	-0.416**	-0.086ns	0.021ns	-0.439**	-0.059ns	0.006ns	0.122*	-0.237**	0.014ns	-0.090ns	0.087ns	
Content of lint	0.144*	0.079ns	-0.022ns	-0.031ns	0.020ns	0.011ns	0.115ns	-0.003ns	-0.258**	0.006ns	0.210**	-0.144*	-0.079ns	-0.058ns	0.036ns

Table 4. Matrix of path analysis on lint percentage of cotton germplasm

Characteristics	Direct influence	Plant height	Monopodial branches	Date to the first square	Sympodial branches	Date to the first flower opening	Date to the first boll opening	Number of bolls per plant	Boll weight	Seed index	Fiber length	Fiber fineness	Fiber strength	Fiber elongation	Fiber uniformity	Insect susceptibility	Total influence
Plant Height	0.099		0.012	-0.017	-0.052	0.012	0.014	0.088	-0.010	0.050	0.016	0.030	-0.041	0.000	-0.001	-0.053	0.147
Monopodial branches	0.041	0.029		0.003	-0.011	0.003	0.014	0.027	0.006	0.017	0.007	0.010	-0.018	-0.005	-0.003	-0.028	0.093
Date to the first square	-0.152	0.011	-0.001		0.013	0.012	0.014	0.088	-0.010	0.050	0.016	0.030	-0.041	0.000	-0.001	-0.053	-0.024
Sympodial branches	-0.097	0.053	0.005	0.020		0.003	0.014	0.027	0.006	0.017	0.007	0.010	-0.018	-0.005	-0.003	-0.028	0.011
Date to the first flower opening	0.048	0.025	0.003	-0.141	0.004		0.014	0.088	-0.010	0.050	0.016	0.030	-0.041	0.000	-0.001	-0.053	0.032
Date to the first boll opening	0.073	0.020	0.005	-0.123	0.007	0.012		0.027	0.006	0.017	0.007	0.010	-0.018	-0.005	-0.003	-0.028	0.007
Number of bolls per plant	0.183	0.048	0.006	-0.003	-0.047	0.003	0.014		-0.010	0.050	0.016	0.030	-0.041	0.000	-0.001	-0.053	0.195
Boll weight	0.131	-0.007	0.002	0.010	0.018	0.012	0.014	0.088		0.017	0.007	0.010	-0.018	-0.005	-0.003	-0.028	0.247
Seed index	-0.275	-0.018	-0.003	0.006	0.008	0.003	0.014	0.027	-0.010		0.016	0.030	-0.041	0.000	-0.001	-0.053	-0.295
Fiber length	0.089	0.017	0.003	-0.014	-0.004	0.012	0.014	0.088	0.006	0.050		0.010	-0.018	-0.005	-0.003	-0.028	0.217
Fiber fineness	0.156	0.019	0.003	-0.007	-0.011	0.003	0.014	0.027	-0.010	0.017	0.016		-0.041	0.000	-0.001	-0.053	0.134
Fiber strength	-0.153	0.027	0.005	-0.017	-0.025	0.012	0.014	0.088	0.006	0.050	0.007	0.030		-0.005	-0.003	-0.028	0.008
Fiber elongation	-0.042	0.000	0.005	-0.003	-0.011	0.003	0.014	0.027	-0.010	0.017	0.016	0.010	-0.041	0.000	-0.001	-0.053	-0.068
Fiber uniformity	-0.022	0.003	0.006	-0.007	-0.025	0.012	0.014	0.088	0.006	0.050	0.007	0.030	-0.018	0.000	-0.001	-0.053	0.115
Insect susceptibility	0.154	-0.034	-0.008	0.002	vw	0.003	0.014	0.027	-0.010	0.017	0.016	0.010	-0.041	-0.005	-0.001	-0.028	0.135

characters (0.099 and 0.147), number of bolls per plant (0.183 and 0.195), and boll weight (0.131 and 0.247), fiber length (0.089 and 0.217), fiber fineness (0.156 and 0.134), and insect susceptibility (0.154 and 0.135) compared with other agronomic characters.

The effect of residual in path analysis can determine how much the independent variable can explain the dependent variable (Figure 1). Figure 1 produces a residual influence of 0.92, which shows that the characteristics of plant height, number of bolls per plant, bolls weight, fiber length, fiber fineness, and insect susceptibility (*Amrasca biguttula*) attack can explain 0.07 of their influence on the content of lint, or it can be stated that 92% of the fiber percentage is influenced by other characters, which cannot be explained by the results of the cross-print analysis that has been carried out.

3.5. Selection of Cotton Accession Grouping Based on the Content of Lint and Fineness Characters

The grouping of 266 accessions to cotton germplasm was based on two agronomic characters obtained from correlation and path analysis. The genetic material used consisted of 266 cotton accessions. After obtaining the selection characteristics, the best cotton accessions were selected based on these characteristics using a selection percentage of 10%, resulting in 30 selected cotton accessions (Figure 2). If the cutting were carried out at an Euclidean distance of 50%, there would be 4 varying accession groups. The first group consists of 5 accessions with characteristics, namely the content of lint (39.5-41.1%) and fiber fineness (3.7-4.4 mic). The second group comprised 11 cotton accessions with a content of lint (37.6-38.9%) and fiber fineness (3.8-4.6 mic). The third group consists of 3 cotton accessions with a range of content of lint (40.3-40.9%) and fiber fineness (5.0-5.6 mic).

Meanwhile, the fourth group consisted of 11 cotton accessions with a range content of lint character of 37.8-39.7 and fiber fineness character of 4.1-5.3 mic. The result of the grouping of cotton accessions can be used as an indicator of the parents, namely the accessions from the third group. This is because the accessions in the third group have far more genetic similarities than the other groups. If crosses are carried out between lines that have close genetic similarities, it will cause inbreeding depression, which will result in genetic decline in the plant.

3.6. Selection of 30 Cotton Germplasm Accessions to Get Potential Cotton Accessions

The selection of cotton germplasm accessions on two characteristics: content of lint and fiber fineness. Accessions are separated into four quadrants. The boundary line between quadrants is based on the 90% confidence interval value presented in Figure 3. It shows that the first quadrant (I) is the quadrant for accessions with a low content of lint and high fiber fineness. The second quadrant (II) is the quadrant for accessions with a low content of lint and fiber fineness.

The third quadrant (III) is the quadrant for accessions with a high content of lint and fineness. The fourth quadrant (IV) is the quadrant for accessions with a high content of lint but low fiber fineness. Based on the quadrant separation, several potential accessions from the 30 selected cotton germplasm accessions can be used as the parent materials for the development of new varieties, namely CEAN 268, CEDIX, or L21999-10-71. The analysis results prove that these accessions have the best content of lint and fiber fineness compared to other accessions.

4. Discussion

4.1. Genetic Diversity Estimation of Germplasm Cotton Accessions

In the cotton germplasm population, the criteria for the coefficient variance (CV) are divided into 3; namely, the coefficient variance is large if the CV value is at least 20%, CV is moderate if it is 10%-20%, and CV of low if it is a maximum of 10% (Wardiana 2018). The plant height character showed the highest CV value compared to other agronomic characters. The spread of phenotype data is quite large, with a value interval of 51-233 cm and a mean of 110 cm. The large distribution of the data indicates that the population is heterogeneous (Table 1). In addition, a character with a large CV value also indicates that the influence of genetics and heterogeneous environmental conditions causes the amount of phenotypic diversity (Misfalah *et al.* 2017).

Meanwhile, the low coefficient of variance indicates that the distribution of genetic variance in the cotton population is homogeneous. Daryanto *et al.* (2020) and Farhah *et al.* (2022) stated that the value of the suitability coefficient indicates that the level of homogeneity of the variety is met. The lower the CV value, the higher the level of data homogeneity, and the higher the CV, the higher the level of data heterogeneity, and it has an impact

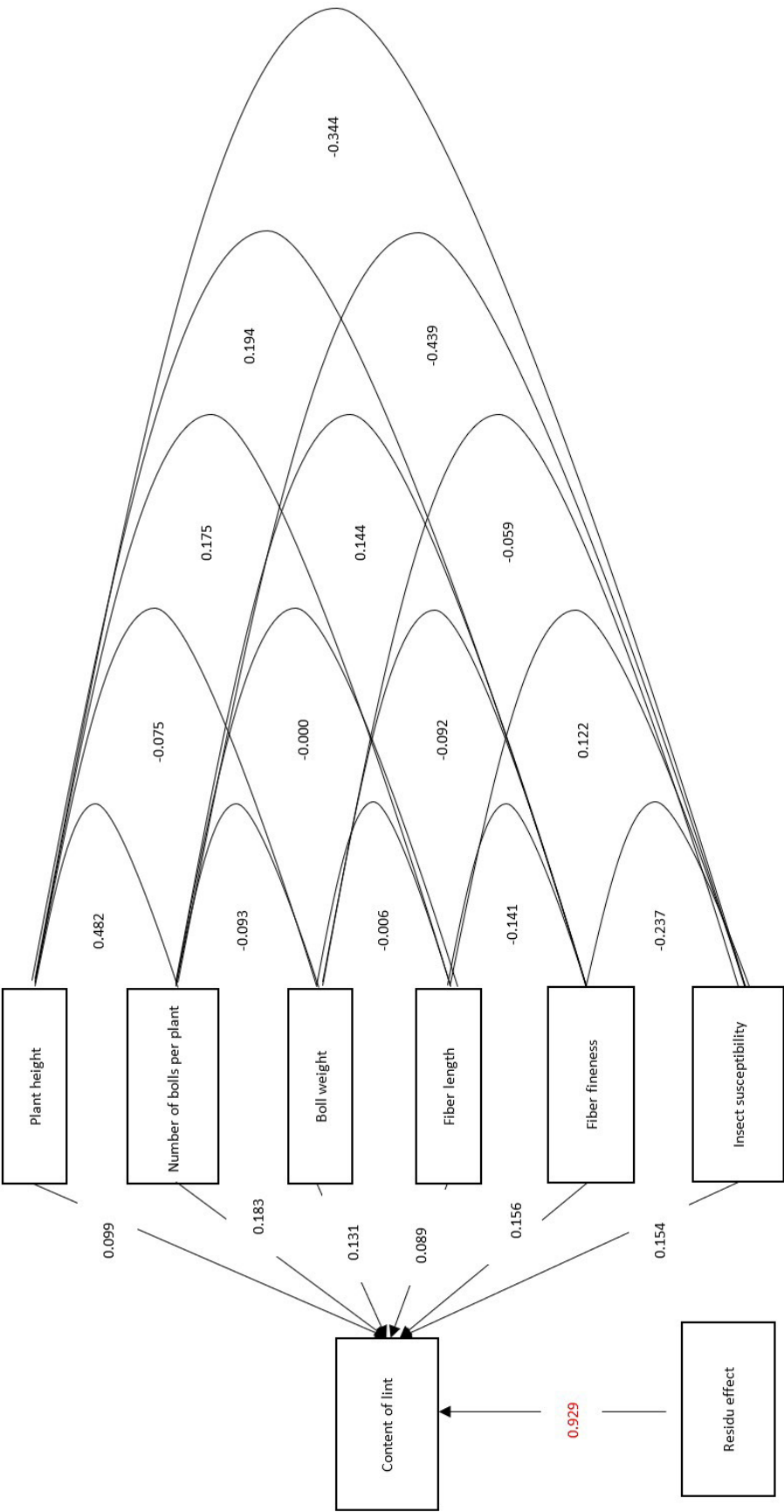


Figure 1. Path analysis content of lint character on cotton germplasm accessions

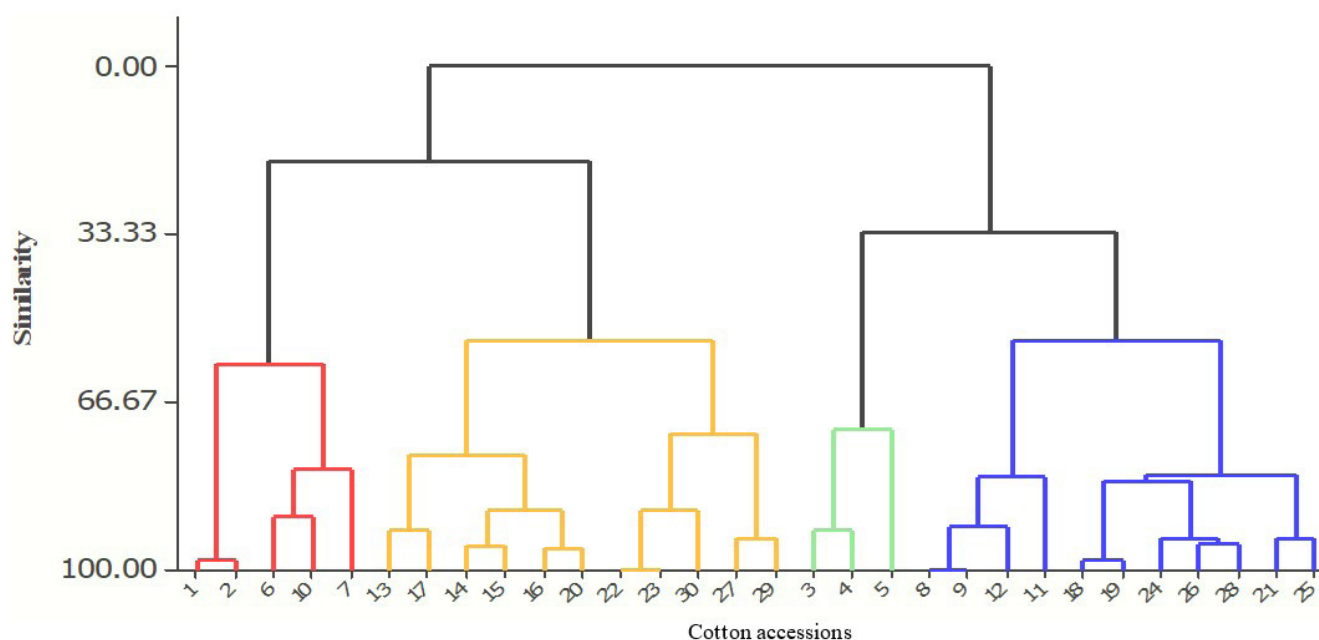


Figure 2. Dendrogram of cotton accessions based on the content of lint and fineness characters. (Group 1: CEA N 81986, PI.437736, PD 4416, PI.433735; dan REBA 1887), (Group 2: PI.433737, MC NAIR 235, A/38 L-124-9, ISA 205 B, STONEVILLE 21985, CIAMIS 1/1, BJA SM-68, CEA G 21986, HL 1, STONEVILLE 2 B, DELTAPINE 30), (Group 3: CEDIX, L21999-10-71, CEA N 268), (Group 4: IRMA (95-97), NURSE F, CEA S 119, ISA 205 A, SICOT 3, MONADO, CEA K 696, T-120-79, MORI 5/1/1, SIOKRA, ALAR (1985)17

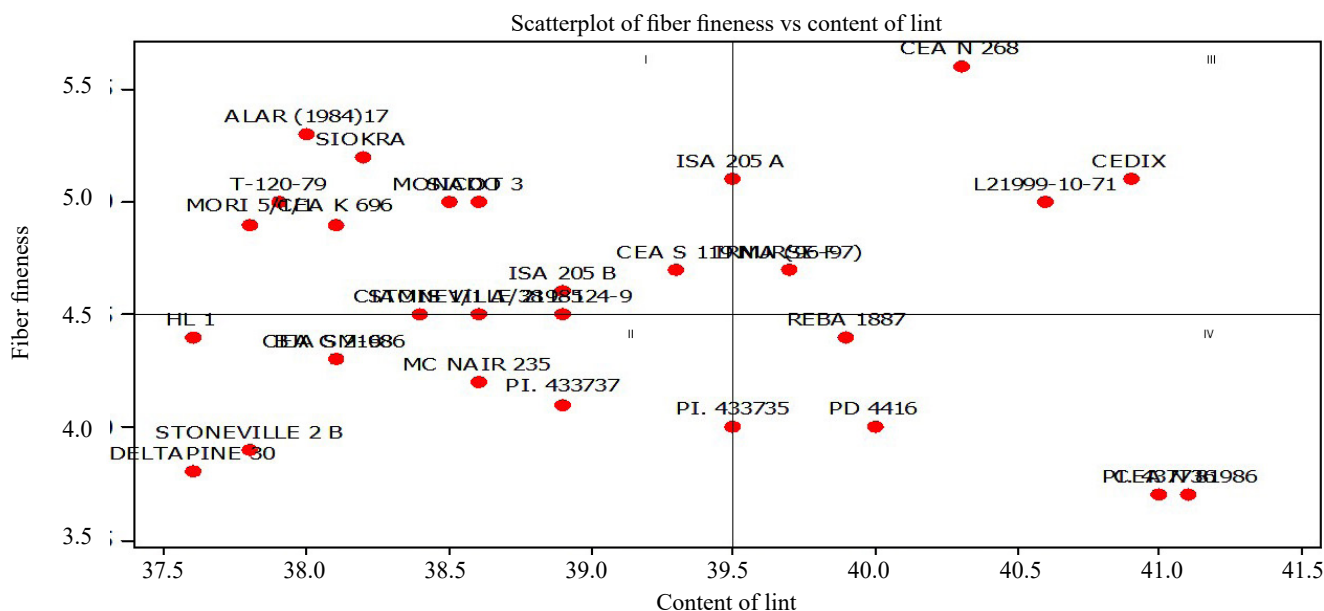


Figure 3. Distribution of 30 accessions of cotton germplasm for the content of lint and fiber fineness character

on the distribution of data from each variety (Yamin and Qadri 2023; Yamin *et al.* 2024).

4.2. Gene Action Estimation of Morpho-Agronomic Character

Information regarding differences in the performance of the cotton germplasm population (Table 1) revealed

a relatively low information material to obtain selection characters through direct or indirect selection. Thus, it is necessary to analyze the estimation of gene action presented in Table 2 by skewness and kurtosis value to predict gene action from plant agronomic character with descriptive statistics. The wheat research reported that the estimation of gene action as the base for selection can be

estimated through the skewness value for gene action and kurtosis value for the controlling gene (Yamin 2014). A skewness value < 0 indicates that the shoot growth of the sugarcane parameter is controlled by additive gene action with duplicate epistasis. Meanwhile, skewness value > 0 is produced by the sugarcane parameter's leaf area, which means that it is controlled by additive gene action with complementary epistasis. Positive kurtosis indicates that many genes control the plant height character, and negative kurtosis indicates that a few genes control the greenness parameter of sugarcane leaves (Hastini *et al.* 2021; Yamin *et al.* 2024). Morphological character and yield components of cotton germplasm are quantitative characteristics controlled by many minor (polygenic) genes, which means that each gene has a small and additive effect on the phenotypic character (Sackton and Hartl 2016). The genes that control these quantitative characteristics work together so that genetically, they have a greater influence than environmental influences (Acquaah 2020). Some characters show positive and large skewness values, which could explain that the action of genes controlling these characters is complementary; the gain is slower with mild selection, and the gain is faster with intensive selection (Sumathi *et al.* 2018). At the same time, the rest have a nearly normal distribution that genes can influence. Furthermore, the dominance of x dominance interaction was larger than the additive x or additive x dominance effects. In addition, duplicate epistasis played a greater effect than the complementary epistasis effect (Pavan and Gangaprasad 2022). Due to the magnitude of dominant effects and unpredictable interactions, the type of duplicate epistasis gene action is relatively difficult for breeders to perform selection (Mistry *et al.* 2016). The high influence of non-allelic interactions causes high levels of non-additive gene action, reflected in the heritability values of the observed characters. So, the action of the epistasis gene was found in all the observed characters, with more influencing in duplicate epistasis than complementary epistasis (Mawasid *et al.* 2019). The results of Xie *et al.* (2023) show that most genetic combinations in alleles that are specifically non-additive will physically interact and result in additional epistasis and dominance.

4.3. Agronomic Character Correlation to the Content of Lint

Correlation analysis estimation is aimed at studying the pattern of relationships between agronomy characters and yields when selecting the main characters, which are always the goal of improvement in every plant breeding

program. In general, correlation analysis can only describe the correlation between the observed characters without seeing the extent to which the characters give value to other characters. Thus, there is a path analysis that can describe the extent to which each character gives value to the main character, either directly or indirectly. The linear correlation coefficient information on the extent to which two variables have a very close association (Senthilnathan 2019). Plant height was significantly and positively correlated with the character of growth and yield of cotton; the correlation between source and sink is crucial for normal growth, yield, and cotton quality. The assimilated transport and distribution affected crop yield. Cotton photosynthesis has been correlated with yield at the genetic level and can be configured to improve synthesized assimilates to be partitioned for sink (fiber) growth (Khan *et al.* 2020). In cotton plants, there is a greater biomass accumulation in vegetative organs due to their indeterminate nature; that will affect production as it allows for leaf and fruit shedding (Qin *et al.* 2023).

The number of bolls per plant character was significantly and positively correlated with the growth component but was negatively correlated with the attacking character of insect susceptibility. This shows that the decrease in the number of insects causes an increase in the number of cotton fruits per plant and vice versa. Thus, it is concluded that insect infestation affects the production and quality of cotton fiber. Because the insect absorbs a lot of cell sap relative to its mass, a certain part of this liquid is excreted as a sticky, sugary substance without being digested (Yusupova and Irisova 2023). When such substances are released in large quantities, they stick cotton fibers together and reduce the quality of the fiber. The negative correlation values exist when increasing values of one variable are associated with a decrease in the values of the other (Aggarwal and Ranganathan 2016). The boll weight character was significantly and positively correlated with the seed index character; the photosynthate obtained was translocated for the boll formation, fiber, and cotton seed. The increase in the sink and source activity causes an increase in effective plant yield growth and harvest index. The improvement in photosynthesis of the main-stem leaf contributed to enhanced boll density, while the improvement in photosynthesis of the sympodial leaf promoted greater boll weight (Wang *et al.* 2023).

On the contrary, higher insect susceptibility can inhibit the growth, production, and quality of cotton fiber. There is a suspected cotton accession that morphologically has a leaf hair number that is tolerant to leafhopper insects. Leafhoppers' infection was quantified by counting the

number of nymphs/adults on the underside of the fully opened leaves on one-third of the upper part of the plant. The severity of the leafhoppers' infection was recorded based on the level of leaf damage severity measured using a scoring system, which was developed from the description of *A. biguttula* infestation symptoms in cotton (Madhu *et al.* 2024; Sunarto *et al.* 2024). In addition to the leaf hair number, the influence of natural enemies such as spiders has a positive correlation to the population of leaf hoppers (*A. biguttula*) in an early phase. Still, among all the sucking pests, leaf hopper is the most destructive insect pest and causes damage to the crop (Nemade *et al.* 2015).

Generally, strength, fineness, and elongation influence the character of cotton fibers. Direct influence is the influence of a character on another character directly without going through the intermediary of other characters. At the same time, indirect influence is the character's influence on other characters through the intermediary of other characters. The agronomic character with the highest direct effect indicates that the character can be used as a secondary selection character. However, if the direct effect of the character is negative, it is due to the relationship of other characters that indirectly affects the result through the content of lint. In linseed plants, it was found that seed yield per plant, branches per plant, and plant height showed a positive association and had a positive direct effect on seed yield (Dabalo *et al.* 2020). The character with the highest correlation value and direct influence in path analysis can be used as indirect selection criteria to improve leaf yield in African spider plants under stress and non-stressed conditions (Zakaria *et al.* 2017; Houdegbe *et al.* 2022).

4.4. Path Analysis Lint Percentage on Cotton Germplasm Accessions

Path analysis has been used to present causal relationships between prediction and response variables through a path chart. The research on rice shows that path correlation analysis indicated a direct positive effect of the three traits (panicle number, full grain number, and 1000 grain weight), which were associated with a positive correlation with the grain yield per plant (Saleh *et al.* 2020). The correlation and path analysis results revealed the relationship pattern of 16 morpho-agronomic characters to the content of the lint character. The pattern of the relationship between these characters can be considered selection criteria.

The characters that can be used to determine the selection criteria have several considerations, namely positively and significantly correlated with the target character (the content of lint), and have a high direct and indirect (total) value. The result of the correlation analysis showed that the plant height and fiber fineness characters were positively and significantly correlated with the content of the lint character. A positive correlation indicates that increasing the value of a variable will increase the value of other variables, such as plant height, and have a highly positive correlation with branches, clusters, weight, and lobes in tomato fruits (Kousar *et al.* 2021). Based on the results of the path analysis in plant height, number of bolls per plant, boll weight, fiber length, fiber fineness, and insect susceptibility characters have a high direct and indirect influence on the content of lint character. Some of these characteristics are directly related to the fiber production process. The potential content of lint, which is higher and heavier, usually has a thick fiber layer. Genetically, it is known that there are genes that can significantly synthesize cell walls to regulate fiber elongation and produce higher-quality fibers (Delhom *et al.* 2022).

The combined correlation and path analysis result on 16 characters observed only fiber fineness character affected the lint content. Therefore, the fiber fineness character can be used as an indirect selection character in 266 accessions of cotton germplasm. The correlations of cause and effect are important tools to assist breeders in defining priority traits for the selection, allowing them to choose which genetic changes should be made to increase the yield (Silva *et al.* 2016). Lestari *et al.* (2022) stated that the residual value was 0.63, which means that cross-analysis could not explain the total variation in the number of pods per plant by 63% but could only explain 37% of the total diversity. The study revealed the residual value was 0.93.

4.5 Selection of cotton accession grouping based on the content of lint and fineness characters

Germplasm management through grouping and characterization is useful for determining the diversity of certain genes and facilitating the introgression of certain genes into superior varieties, understanding evolutionary relationships between accessions, and increasing efficiency in germplasm management. Germplasm grouping can be done by conducting principal component analysis (PCA) to obtain the main components that retain some of the

information contained in the original data. The results of the quadrant division obtained several accessions that had been selected from 30 germplasms that could be used as parent materials in the development variety program, namely CEA N 268, CEDIX, or L21999-10-71, which have the best content of lint and fiber fineness characters. A scatter plot was made based on the loadings of each genotype in principal components one and two, namely PCA1 and PCA2. In addition, different clusters were formed in the scatter plot based on the similarity of evaluating diverse soybean genotypes for seed longevity. Three distribution clusters of 70 kenaf accessions were obtained based on dry core and fiber weight characters; furthermore, in quadrant III, 2 accessions potentially produce the best dry fiber and can be considered a parent source in developing a new superior variety of kenaf (RS *et al.* 2022).

In conclusion, the result showed that the fiber length character had a very low variance with a value of 0.01%. On the contrary, the plant height character has the highest variance at 637.42. However, both characters were influenced by additive gene action and controlled by a few genes based on skewness and kurtosis values. The content of lint has a variance value of 9.37%, and it is influenced by duplicate epistasis genes and is controlled by many genes. The fiber fineness character has a positive and very significant correlation value to the character of the lint percentage ($r = 0.210$). These characters also have a fairly high direct influence and total effect value of 0.156 and 0.134, respectively. In the grouping of 30 cotton accessions based on principal component analysis with a selection intensity of 10% for the content of lint and fiber fineness character, one accession group (the third group) has the potential to be used as a source for the parent group. Three cotton accessions could be used as one of the parents, namely CEA N 268, CEDIX, or L21999-10-71. The potential accessions and selection characteristics obtained will contribute to developing future superior cotton varieties.

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