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RESEARCH ARTICLE

Evaluation of the fresh butterfly pea (*Clitoria ternatea* L.) accessions for the flavonoid content from the dried flowers

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Abstract

Butterfly pea flowers (*Clitoria ternatea* L.) are vines frequently used as ornamental plants and health supplements. Butterfly pea flowers have a very interesting colour variation, namely blue, which is the most commonly used variation, and white. Butterfly pea flowers are thought to contain phenolic compounds, including phenols, flavonoids and tannins that are useful as antidotes to free radicals or antioxidants that can stabilise electrons and provide hydrogen to the body. The colour of the flowers can change from blue to pale purple as they dry. This study investigated the colour diversity and colour differences of fresh flowers of 44 accessions of blue and white single and double petals. The selected accessions were Cte-61482-01 with double petals and Cte-55891-01, Cte-55891-02, Cte-64293-01 and Cte-56362-01 with single petals, which consistently produced the highest flower weight with bright colour intensity and high flavonoid content.

Keywords: Butterfly pea flowers, antioxidants, health, weight, color

Introduction

Butterfly pea flowers (*Clitoria ternatea*) are vines that are usually used as ornamental plants as well as health ingredients for the body. Butterfly pea flowers have a very interesting color variation, namely blue, which is the most widely used variation, and white (Suarna dan Wijaya, 2021). Butterfly pea flowers are rich in health benefits such as anti-cancer, antioxidant, anti-inflammatory and anti-diabetic (Marpaung *et al*., 2020). This is due to the presence of bioactive compounds in butterfly pea flowers, such as tannins, saponins, triterpenoids, flavonoids, alkaloids, phenols, flavanol glycosides, anthraquinones, anthocyanins, essential oils, proteins, and carbohydrates (Fitrilia *et al*., 2020). The exotic colour of butterfly pea flowers and the many health benefits make butterfly pea flowers widely used in the food industry, such as traditional medicine, fresh vegetables, food colouring and herbal drinks (Weerasinghe *et al*., 2023). Butterfly pea flowers are starting to become widely known for their beautiful and attractive colours, so many people are starting to use butterfly pea flowers on a production scale. There is a growing interest in butterfly pea flowers, but the scale of production does not meet the target market and consumer requirements. In general, consumers are interested in high weight and light colour.

The colour and weight of the flower is very important on an industrial scale because the weight of the flower determines the flavonoid content to produce an attractive colour (Prabawati *et al.*, 2021). The value of diversity is one of the factors in the appearance of fresh flower colour and weight with different characteristics for each accession. Therefore, it is necessary to conduct research on the diversity of fresh flower colour characters by determining the fresh weight and colour of each accession in different regions to provide important information on the superior characteristics of each accession. The higher the fresh weight of butterfly pea flowers, the higher the flavonoid content in the dried flowers. Determining the value of diversity will be a recommendation for the industrial scale to select superior accessions in terms of colour and fresh flower weight.

Material and methods

The research was conducted from December 2023 to April 2024 in the Greenhouse, Faculty of Agriculture, Universitas Brawijaya. The tools and materials used were 30 x 30 poly bags, Iphone camera, 0.000 g analytical balance, motar, pestle and stationery. The materials used were seeds from 44 accessions of single and double petalled blue and white flowers. This study used the observation method to compare the characteristics of fresh flowers and flower colour. When observing flowers from 2 plants, 10 flowers were taken as random samples and then the sensory colour of the fresh flowers was immediately assessed using the Colour Grab application, which can be downloaded from the Play Store. Descriptive statistics were used to analyse the research data, with the results of the observations obtained from the observational data presented descriptively and the results presented in the form of photographs, frequency histograms and descriptions of each accession. The results of descriptive observations were calculated using the value of variance, standard deviation, between and within accession analysis, Z-test, coefficient of variation, manual graphs and box plots.

Results and discussion

Variation of fresh flower weight in butterfly pea flower accessions

The value of diversity in physical characteristics of fresh flower weight can be known from the value of coefficient of variation (CV). Based on the results of the analysis between and within accessions showed that of the 44 collections of butterfly pea flower accessions gave a significant effect on the weight of fresh flowers. The mean fresh flower weight is shown in Table 1. The results for the diversity value tested in the accession showed a coefficient of variation ranging from >1% - 4%, indicating that the low diversity value is due to the butterfly pea plant self-pollinating to produce homozygous offspring. Butterfly pea flower (*C. ternatea*) can produce low diversity because most flowers are self-pollinated and produce offspring that are similar to their parents (Jacob and Kunuku, 2021). The low diversity within accessions shows the uniformity of fresh weight of samples within accessions which are almost the same. The diversity between accessions of all 44 accessions tested results in a coefficient of variation of 45%, indicating that there is high diversity in fresh weight between accessions. The high level of diversity between accessions may be caused by differences in origin or population, which bring unique genetic variation to the butterfly pea plant. Differences in plant origin will have different environmental adaptations, this can form different characters in each plant so the resulting diversity will be more varied. Earlier variation in pea/cowpea was studied by different researchers (Doumbia *et al.*, 2019; Ullah *et al.*, 2019; Siyunda *et al.*, 2023).

Table 1: Mean fresh weight and diversity values of butterfly pea flower accessions

Accessions	Origin	Characteristics	Mean (g)	CV(%)
Cte-Ternate-01	Ternate City	Single Petal Blue	0.3890	0.78
Cte-Ternate-02	Ternate City	Single Petal Blue	0.4140	3.43
Cte-Malaysia	Malaysia	Single Petal Blue	0.4660	1.70
Cte-16421-01	Beji District, Depok	Single Petal Blue	0.4620	0.93
Cte-97719-01	South Ternate City District, Ternate	Single Petal Blue	0.3520	3.50
Cte-55891-01	Gunungkidul Regency	Single Petal Blue	0.5020	1.28
Cte-55891-02	Gunungkidul Regency	Single Petal Blue	0.5140	2.21
Cte-55891-03	Gunungkidul Regency	Double Petal Blue	0.8880	2.35
Cte-62211-02	Lamongan Regency	Single Petal Blue	0.3300	0.77
Cte-62211-03	Lamongan Regency	Single Petal Blue	0.3200	1.10
Cte-62211-04	Lamongan Regency	Single Petal Blue	0.3630	1.32
Cte-62211-05	Lamongan Regency	Single Petal Blue	0.3240	1.24
Cte-62211-06	Lamongan Regency	Single Petal Blue	0.3930	1.17
Cte-64293-01	Kepung District, Kediri Regency	Single Petal Blue	0.5310	2.30
Cte-69281-01	Camplong District, Sampang Regency	Single Petal Blue	0.2290	0.60
Cte-69281-02	Camplong District, Sampang Regency	Single Petal Blue	0.3290	1.10
Cte-69281-03	Camplong District, Sampang Regency	Single Petal Blue	0.2640	1.71
Cte-69281-04	Camplong District, Sampang Regency	Single Petal Blue	0.3320	1.12
Cte-69281-05	Camplong District, Sampang Regency	Single Petal Blue	0.2800	0.35
Cte-69281-06	Camplong District, Sampang Regency	Single Petal Blue	0.3640	1.86
Cte-69281-07	Camplong District, Sampang Regency	Single Petal Blue	0.3380	0.87
Cte-68175-01	Ajung District, Jember Regency	Single Petal Blue	0.3980	2.12
Cte-68175-02	Ajung District, Jember Regency	Single Petal Blue	0.4470	1.08
Cte-68175-03	Ajung District, Jember Regency	Single Petal Blue	0.3780	1.27
Cte-67111-01	Panggungrejo District, Pasuruan City	Single Petal Blue	0.3560	1.40
Cte-45141-01	Harjamukti District, Cirebon City	Single Petal Blue	0.4220	1.50
Cte-61111-01	Gresik District, Gresik Regency	Single Petal Blue	0.3930	0.98
Cte-61482-01	Mojoagung District, Jombang Regency	Double Petal Blue	1.0600	2.82
Cte-56362-01	Leksono District, Wonosobo Regency	Single Petal White	0.4800	0.96
Cte-56362-02	Leksono District, Wonosobo Regency	Single Petal White	0.4743	1.67
Cte-56362-03	Leksono District, Wonosobo Regency	Double Petal Blue	0.9167	2.67
Cte-56362-04	Leksono District, Wonosobo Regency	Double Petal Blue	0.9325	2.91
Cte-68483-01	Banyuwangi Regency	Double Petal White	0.6700	1.19
Cte-68483-02	Banyuwangi Regency	Double Petal Blue	0.9300	3.30
Cte-65392-01	Ngantang District, Malang Regency	Single Petal Blue	0.3100	0.72
Cte-69416-01	Sumenep Regency	Single Petal Blue	0.3230	0.57
Cte-69416-02	Sumenep Regency	Single Petal Blue	0.3030	1.52
Cte-69416-03	Sumenep Regency	Single Petal Blue	0.3330	0.50
Cte-69416-05	Sumenep Regency	Single Petal Blue	0.2860	2.16
Cte-69416-06	Sumenep Regency	Single Petal Blue	0.3190	1.00
Cte-69416-07	Sumenep Regency	Single Petal Blue	0.3220	1.11
Cte-69416-04	Sumenep Regency	Single Petal Blue	0.3010	1.42
Cte-69416-08	Sumenep Regency	Single Petal Blue	0.3560	0.98
Cte-69416-09	Sumenep Regency	Single Petal Blue	0.3430	0.70
			CV(%)	45

Description: CV values on the side are CV within accessions and CV values below are CV between accessions.

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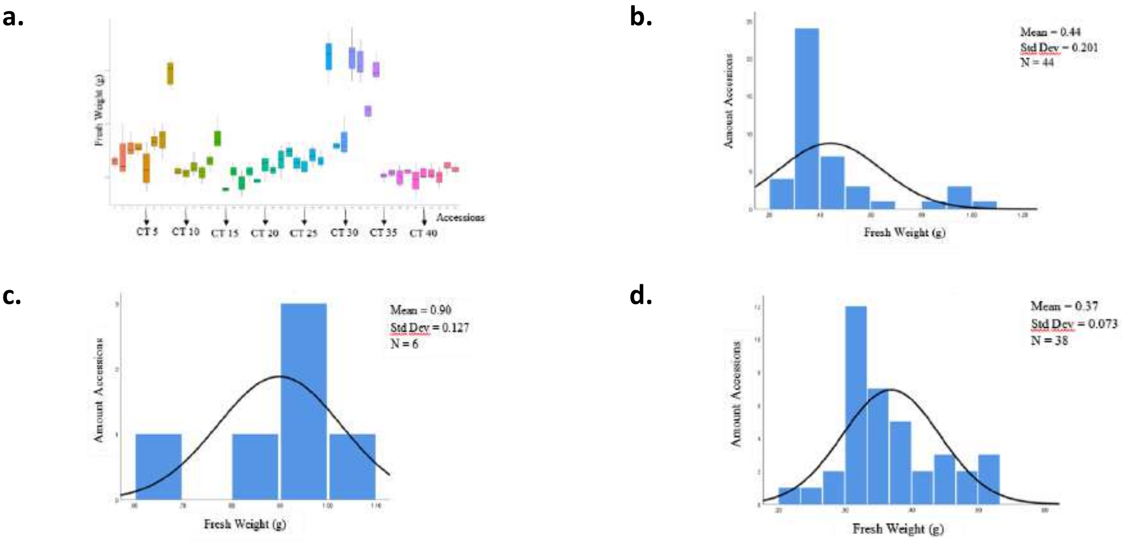
Fig 1: Data distribution of single and double petal fresh weight. a) Box plot distribution of fresh weight flower; b) Histogram and normalised graph of fresh weight of single petal and double petal ; c) Histogram and normalised graph of fresh weight of double petal ; d) histogram and normalised graph of fresh weight of single petal



The results of the experiments carried out have a diversity value of 14.2%, which shows that the percentage of error rate in an experiment is classified as moderate and can be significant in producing fresh flower weight between accessions and within the accession of butterfly flower studied (Ulimaz *et al*., 2020). Diversity assessment is a fundamental step in plant breeding programmes to identify and select superior traits (Demirel *et al*., 2024). Variation with a medium category will have a value of data distribution is not too homogeneous but also not too varied, as seen in Figure 1. Fresh weight is very variable shows that the 44 accessions in the plant flower collection have a very real influence on the value of diversity in fresh weight of flowers. The average fresh weight produced by the characteristics of the double petals is higher than that of the single petal. This is because the double petal characters have more than one petal layer (Zhang *et al*., 2024). The distribution of fresh weight data is shown in Fig 1.

The high variation value between accessions causes a variety of fresh weight data distribution values. There are 2 accessions with symmetrical data distribution, 19 accessions with lower value data distribution, 20 accessions with higher value data distribution and 3 accessions with very narrow range values and only a few higher values. Superior accessions are accessions that pass the value criteria at the 5% level of the z-test, namely Cte-61482-01 from Mojoagung District, Jombang Regency; Cte-55891-01 and Cte-55891-02 from Karangmojo District, Gunungkidul Regency; Cte-64293-01 from Kepung District, Kediri Regency; and Cte-56362-01 from Leksono District, Wonosobo Regency. The selected accessions were superior for their blue and white single petal and blue double petal characteristics. The selected accessions are accessions with high fresh weight and data distribution, which are classified as larger values. Different seeds in each region certainly have different environmental conditions, so that environment

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becomes one of the factors influencing genetic traits in regulating plant growth and development, although genotypes provide the basic potential for growth and development (Kurniati *et al.*, 2023).

Colour variation of fresh flowers in butterfly pea flower accessions

The diversity in the colour of butterfly pea flowers can be seen from the results of L*, a*, b* values in each butterfly pea flower accession. These measurements show the category of colour class in butterfly pea flowers. The L* value indicates brightness levels from 0 (black) to 100 (white), the a* value indicates the intensity of red (+) and green (-), and the b* value indicates the intensity of yellow (+) and blue (-). According to Hossain (2019), The a* value ranges from 0 to 80, indicating red, while values from -80 to 0 represent green. Similarly, the b* value follows this pattern. Detailed color diversity of fresh flowers for each accession is presented in Table 2. The diversity of fresh flower colour intensity shown in Table 2 shows different L*, a* and b* values even though the colour classes are of the same colour. Overall, the accessions provide differences in brightness, whiteness and blueness of butterfly pea flower colour. Blue butterfly pea flowers have higher L*, a* and b* values than white butterfly pea flowers. Blue butterfly pea flowers have lighter colours with red (+a*) and blue (-b*) tones, whereas white butterfly pea flowers have green (-a*) and blue (+b*) tones. The colour of the blue butterfly pea flowers is mostly included in the purple-blue colour and the white butterfly pea flower accession is included in the white colour. The b* value affects the concentration of colour in both blue and white butterfly pea flowers. Increasing the concentration of butterfly pea flowers will result in a higher b* value. According to Loppies *et al.*, (2020), the diversity of anthocyanins is the result of different groups in the basic structure, so they produce different colour results. Accessions that pass the selection are included in the light colour with a large b notation value on the characteristics of single and double petals of blue colour and a larger L value than the characteristics of white single petals. Further details can be seen in Fig. 2. The superior accession in Figure 2. light colour quality is influenced by genetic and environmental factors that contribute to the colour diversity of fresh flowers. Genetic factors relate to the genetic diversity of each accession or variety, while environmental factors include climatic conditions, soil and cultivation techniques used (Yulina *et al.*, 2021). The colour of each selected accession falls into a diverse colour category such as Cte-61482-01 with dark violet, Cte-55891-01 with dark violet, Cte-55891-02 with violet, Cte-64293-01 with blue violet, Cte-56362-01 with white. This colour variation is the quality of the flowers that consumers are interested in. According to Priska *et al.*, (2018), Darker butterfly pea colour intensity tends to be preferred by consumers because it is considered to have better colour quality. Darker colour intensity indicates a higher anthocyanin pigment content, so the resulting product has a brighter and more attractive colour. This indicates that darker colour intensity can also increase consumer acceptance of functional beverage products from butterfly pea flowers. The brighter the colour of fresh flowers in butterfly pea flowers, the colour intensity of butterfly pea flower powder in dry conditions will also remain bright even though there is a slight decrease in dullness in colour. Varying colour intensity can also affect consumer acceptance of food or beverage products that use butterfly pea flowers as a natural colourant. Generally, consumers like consistent and uniform colours in a variety in order to produce quality food or beverage products that they consume (Wang *et al.*, 2024).

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Table 2: Colour intensity of fresh spring flowers

Accessions	Colour Parameters			Color
	L*	a*	b*	
Cte-ternate-01	16.91	48.07	-60.09	*Dark Blue Violet*
Cte-ternate-02	21.43	41.22	-44.31	*Blue Violet*
Cte-malaysia	42.40	46.11	-80.67	*Blue Violet*
Cte-16421-01	21.26	44.95	-52.22	*Dark Violet*
Cte-97719-01	19.29	39.36	-47.01	*Dark Violet*
Cte-55891-01	20.88	44.97	-47.72	*Dark Violet*
Cte-55891-02	29.73	53.6	-58.9	*Violet*
Cte-55891-03	21.61	42.3	-43.9	*Dark Violet*
Cte-62211-02	55.48	12.93	-27.79	*Blue Violet*
Cte-62211-03	35.29	29.87	-49.64	*Blue Violet*
Cte-62211-04	29.97	37.89	-51.83	*Blue Violet*
Cte-62211-05	44.92	50.6	-76.56	*Blue Violet*
Cte-62211-06	29.26	41.34	-57.4	*Blue Violet*
Cte-64293-01	37.08	53.35	-70.35	*Blue Violet*
Cte-69281-01	49.9	34.12	-63.62	*Blue Violet*
Cte-69281-02	43.92	43.38	-66.03	*Blue Violet*
Cte-69281-03	44.93	45.5	-69.06	*Blue Violet*
Cte-69281-04	47.14	56.23	-79.1	*Blue Violet*
Cte-69281-05	43.59	22.26	-37.6	*Blue Violet*
Cte-69281-07	28.21	39	-61.28	*Blue Violet*
Cte-69281-07	48.85	38.68	-66.25	*Blue Violet*
Cte-68175-01	31.94	64.75	-77.26	*Violet*
Cte-68175-02	20.35	46.07	-47.79	*Dark Violet*
Cte-68175-03	22.31	40.16	-51.57	*Dark Blue Violet*
Cte-67111-01	19.63	33.71	-47.9	*Dark Blue Violet*
Cte-45141-01	16.4	33.29	-46.64	*Dark Blue Violet*
Cte-61111-01	31.28	59.35	-77.9	*Blue Violet*
Cte-61482-01	17.55	36.22	-41.43	*Dark Violet*
Cte-56362-01	99.27	-2.53	7.59	*White*
Cte-56362-02	95.26	-1.07	4.83	*White*
Cte-56362-03	21.71	42.32	-49.28	*Dark Violet*
Cte-56362-04	23.35	47.3	-47.89	*Dark Violet*
Cte-68483-01	89.62	2.98	0.2	*White*
Cte-68483-02	18.42	39.61	-40.87	*Dark Violet*
Cte-65392-01	33.01	33.18	-50.73	*Blue Violet*
Cte-69416-01	37.96	34.31	-52.26	*Blue Violet*
Cte-69416-02	55.43	13.92	-84.56	*Blue*
Cte-69416-03	39.7	56.4	-83.33	*Blue Violet*
Cte-69416-05	40.7	45.31	-64.55	*Blue Violet*
Cte-69416-06	47.14	54.07	-77.64	*Blue Violet*
Cte-69416-07	50.43	36.32	-64.41	*Blue Violet*
Cte-69416-04	42.54	33.15	-52.75	*Blue Violet*
Cte-69416-08	45.84	52.88	-74.79	*Blue Violet*
Cte-69416-09	44.6	32.18	-52.59	*Blue Violet*

Description: L*a*b* values are obtained from the results of colour analysis using the Color Grab application

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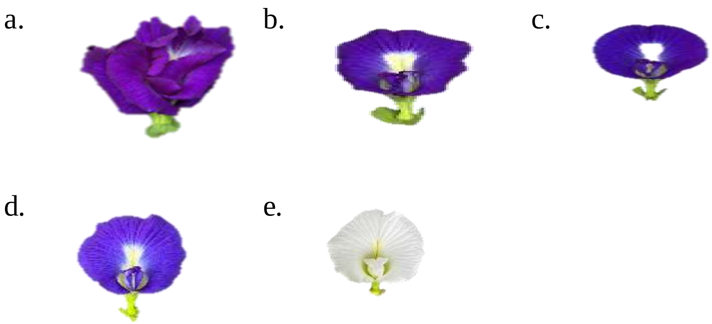
Fig 2: Superior accessions of butterfly pea flower description: a) accession Cte-61482-01; b) accession Cte-55891-01; c) accession Cte-55891-02; d) accession Cte-64293-01; e) accession Cte-56362-01



Hence, in conclusion there is a wide variation between accessions due to differences in the origin of the 44 accessions tested and within-accession uniformity due to the results of self-pollination on 10 flowers in each accession. Accessions that passed the selection with superior categories were Cte-61482-01 from Mojoagung District, Jombang Regency; Cte-55891-01 and Cte-55891-02 from Karangmojo District, Gunungkidul Regency; Cte-64293-01 from Kepung District, Kediri Regency; and Cte-56362-01 from Leksono District, Wonosobo Regency. These accessions have the highest weight according to consumer demand for lighter colour and higher flavonoid content.

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