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Varietal Characterization and Diversity Analysis of Blue Ternate (Clitoria sp.)

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Abstract

Clitoria crops, commonly known as blue ternate and blue peas, have many colors ranging from blue, violet and white, and some varieties have double-layered petals while others are single-layered. This study was conducted to evaluate varietal differences of five species of Clitoria, and was carried out in Randomized Complete Block Design. This was conducted at USM, Kabacan, Cotabato from July 2019 to June 2020. Results of the study revealed significant differences in terms of the nutritive values of the Clitoria flowers and the morphological traits of the crop. In terms of the flower yield, highest yielding variety was the Clitoria with double-layered petals. More pod and seed production was generally attained by white-colored Clitoria variety which was single-layered. In terms of the diversity analysis of the five varieties, using all available plant traits, the five Clitoria varieties were grouped into three clusters with cophenetic value of 0.76. Results of the study implied varietal differences on the five species of Clitoria evaluated. Significant differences on the flower, seed and pod yield were highly noted between Clitoria with double- and single-layered petals. A double-layered variety is recommended for the purpose of flower production, while a single-layered variety, most especially the white-colored Clitoria is more preferred for pod and seed production.

Keywords: blue ternate; butterfly pea; Clitoria.

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1. Introduction

1.1 Background of the Study

Clitoria ternatea L. (Family Fabaceae; sub-family Papilionaceae) is popularly known as blue ternate. The most common English names of this crop are butterfly pea, Darwin pea, blue pea while Pukingan or Puki-Reyna are the local Tagalog names. Clitoria covers 60 species in which majority are found in the tropical belt, but there are still few species existing in temperate areas. Clitoria is a vigorous, herbaceous, perennial, leguminous vine or creeper which is known to have multiple uses. It is used as a legume for building soil fertility, as cover crop, green manure, for fodder/hay, phyto-remediation of coal mines, ornamental plants, medicinal uses and for culinary purposes. Flower extracts can also be used as edible dye and are also used to color foods. In the Philippines, young pods and tender fruits are used as a vegetable [1]. Some are using the colorful petals in garnishing and decorating salads, ice cream and soups. The extracts of the flowers are added in hot water to prepare various desserts, jams and similar products [2, 3]. Blue ternate flowers are commonly used as colorant to variety of foods and beverages such as "Blue Tea and Nam Dok Anchan" in Thailand and "Blue Nasi Kerabu" in Malaysia [3]. For its medicinal values, almost all parts of Clitoria have medicinal properties and are useful in curing different diseases [4]. There are available published reports on the medicinal benefits of the crop which include analgesic, antipyretic and anti-inflammatory [5]. Clitoria possesses a significant high number of benefits against various diseases and ailments in the human body [6] and emphasized the biological impacts of the crop on anti-oxidant, anti-diabetic, anti-helminthic, hepatoprotective, anti-asthmatics and neuropharmacological activities.

1.2 Statement of the Problem

The most common varieties or species of C. ternatea in the Philippines are the blue-colored flowers, with either single- or double-layered petals. Violet- and white-colored Clitoria are also existing but in limited areas only. Characterization of these different varieties is not yet well-explored and majority of the studies and researches published with regards to blue ternate focused primarily on the nutritive benefits of the crop on various health aspects, particularly on its pharmaceutical properties. However, the characterization and the production aspects of different Clitoria varieties were very limited to almost nil most especially when it comes to the yield and productivity of the flower, pods and seeds. By characterizing the germplasm, information on the accession traits could be provided which may result in maximum utilization of the germplasm collection to the end users.

1.3 Score and Delimitation

Five different varieties of Clitoria (colors blue, violet and white), which are either single- and double-layered petals, were evaluated in the study. The study covered the morphological characterization of all parts of blue ternate plant, including the leaves, lateral branches, roots, flowers, pods and seeds, as well as the susceptibility of the varieties to fungal infection. The nutritional components of the flowers and seeds of the crop were also analyzed. The varietal characterization study was limited only to Clitoria varieties which were available during the time of conduct. Only the flowers and seeds of the crop were subjected to mineral/nutrient analysis. The

field studies were limited only up to the time when the plant samples attained the physiological maturity.

1.4 Significance of the Study

This study explored the varietal characteristics of Clitoria in terms of the morphological traits, growth parameters, productivity and yield, crop's response to fungal diseases, nutritional and proximate components of the flowers and seeds, respectively, and the crop's diversity analysis. By knowing these important information, propagation and production of the crop can be enhanced. The varietal decision is one of the key components in establishing a profit potential before a single seed is ever planted. Major general components of profit potential include the capacity to produce good yield, nutrition quality, response to management, resistance to pests and diseases, in addition to consistency and product acceptance of the consumers and market potential. Comparing the yield productivity and nutritional components of different varieties of Clitoria are also necessary in considering which among the existing varieties have more impact on the increase in production as well as in exploring the benefits of the crop in terms of many aspects, most especially when it comes to the health and nutrition benefits and pharmaceutical potentials of the crop. Diversity in plant genetic resources provides opportunities for plant breeders and geneticist to breed and develop new and improved cultivars with desirable characteristics which are both farmers-preferred traits (yield potential, ability to produce good and quality seeds, etc.) and breeders-preferred traits (pest and disease resistance, etc.).

1.5 Time and Place of the Study

This diversity analysis and varietal characterization of Clitoria was conducted at the University of Southern Mindanao, Kabacan, Cotabato, Philippines from July 2019 to June 2020.

2. Materials and Methods

The following materials were used in this study: Clitoria seeds, trellis, planting guide, measuring devices (digital weighing scale and refractometer, meter stick, chemicals and laboratory equipment for nutrient analysis and fungal isolation and other materials which were deemed necessary.

2.1 Experimental Design

The study was carried out in a Randomized Complete Block Design (RCBD) with five (5) treatments and four (4) replications. The following varieties (Figure 1) of blue ternate were evaluated in the study:

T1 - Blue double-layered petal (North Cotabato)

T2 - Blue double-layered petal (Davao City)

T3 - White single-layered petal (Davao City)

T4 - Violet single-layered petal (North Cotabato)

T5 - Blue single-layered petal (South Cotabato)

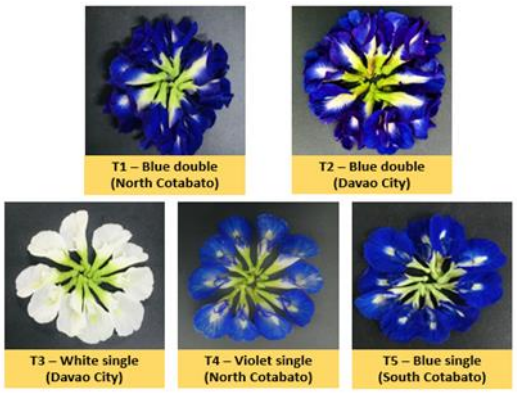

Figure 1: Different varieties of Clitoria: T1 - Blue-double layered variety (NC), T2 – Blue double (DC), T3 – White single (DC), T4 – Violet single (NC) and T5 – Blue single (SC).

2.2 Cultural Management Practices

Preparation of the Experimental Area. The experimental area was thoroughly cleaned by slashing the weeds and removing unnecessary materials in the area. Some trees were also cut in order to ensure that the plant samples were exposed to uniform sunlight. Collection and Sowing of Blue Ternate Seeds. To ensure purity of the variety, all the seeds used in the study came from specific known locations only. Seeds of good quality and condition with uniform size, shape and maturity were used as test samples. One hundred (100) seeds were sown per plot at approximately 12 mm deep. Planting distance was 1 x 1 m between rows and between hills. The plot to plot distance was 2 m. Thinning of seedlings was done at 21 days after sowing (DAS). Only 70 seedlings were left per plot: 10 seedlings were used for the shoot data parameters, 20 for root data parameters, and 40 as buffer plants.Flower and Pod Tagging. The newly emerged buds were tagged using a colored waterproof tags to regularly monitor the developmental stage of the flowers and pods. Proper labelling was done to identify the flower bud and pod samples, date of tagging and sampling numbers. A total of 80 flower buds were tagged per plot: 30 buds to monitor stages of the flower development, 20 tagged full-bloomed flowers for the flower size measurement, and 30 buds in addition to the first 50 buds to check the percentage of flowers which turned into pods. Thirty (30) mature pods were also tagged specifically for pod size measurement and for monitoring the developmental stage and maturity up to senescence of the pods. Harvesting of Flowers and Pods. Flowers and pods were hand-harvested using a pair of scissors. The harvests were placed inside a plastic bag, labelled properly and brought immediately to laboratory. To avoid mixing of harvests, various plant parts were assigned to a particular tray, and checking of proper tags and labels were ensured.

2.3 Plant Materials for Mineral/Nutrient Analysis

Clitoria flowers (2 kgs/variety) and seeds (1 kg of Blue-double) were subjected to nutritional analysis. Nitrogen analysis was done through Kjeldahl method which determined the nitrogen contained in organic substances and inorganic compounds ammonia and ammonium. Phosphorus content was run using Ultraviolet-Visible (UV-VIS) spectrophotometry which measured the intensity of light after passing through a sample, and compared it

to the intensity of light before it passes through the sample. For potassium, calcium, sodium and magnesium contents, evaluation was done by Atomic Absorption Spectroscopy which quantified the absorption of ground state atoms in the gaseous state. The ash content was measured by Gravimetric method.

2.4 Data Analysis

Growth and yield data parameters were analyzed using one-way Analysis of Variance. If significant difference was detected between treatments, a post hoc test, Tukey's Range Test was employed. The statistical analyses were carried out using the STAR software version.

3. Results

3.1 Morphological Characteristics of Different Clitoria Varieties

Table 1 presents the germination percentage at 21 days after sowing (DAS), number of days to 50% germination, plant height, leaf and leaflet count at 42 DAS, root count and root length at 60 DAS and the measurement of the flowers, pods and seeds at maturity stage as affected by varietal differences of Clitoria. Highest percentage germination (74.60%) was attained by Blue-single (SC) which was statistically comparable with Blue-double (NC) with 71.15%. The 50% germination of all entries was observed at 7 to 8 DAS. Plant height of all varieties at 42 DAS except for the Blue-single (SC) was statistically comparable ranging from 77.00 to 89.50 cm. Stages of seedling growth up to 31 DAS were shown in Figure 2A. No significant difference was observed on the leaf and leaflet count at 42 DAS, measurement of the mature leaves and the length of the roots at 60 DAS. Highest number of lateral roots was recorded from Blue-single (SC) and was statistically comparable with Violet-single (NC) with 72.00 and 60.25 roots, respectively (Figure 2B). With regards to the size of the flowers, pods and seeds (Figure 3), highly significant differences were observed on the length measurement. Longest flower length (5.65 cm) was attained by Violet-single (NC), longest pod (10.00 cm) by White-single (DC) and longest seed (7.80 cm) by Blue-double (NC). Both the length and width of blue flowers were longer and wider than white flower variety [7], with 5.3 x 3.8 cm for the blue variety (428.80 mg) and 4.5 x 3.3 cm for white variety (268.80 mg). Moreover, the pods of Clitoria were noted to contain 8 to 11 seeds. The weight of the flowers, pods and seeds, number of pods and seeds in one kg and the number of days from sowing to first bud emergence, flower opening and flower bloom, percentage of flowers which turned into pods and number of seeds per pod were shown in Table 2. In terms of the weight, significant differences were noted on the flowers wherein the weight of the double-layered flowers generally resulted in heaviest weight (573.00 to 588.00 mg) than the single-layered varieties (265.00 to 303.00 mg). The pod and seed weight were not statistically different from each other. With regards to the development and maturation of the flowers (Figure 4), there were significant differences on the number of days from the sowing date to the emergence of the first flower bud up to the senescence. For the percentage of flowers turned into pods, highest results (65.00 to 69.15%) were recorded from White- and Violet-single varieties, and the double-layered varieties generally obtained the lowest results. Pods of the blue variety was observed to have heavier pod (1,600.00 mg) compared to white variety (1,430.00 mg) [7].

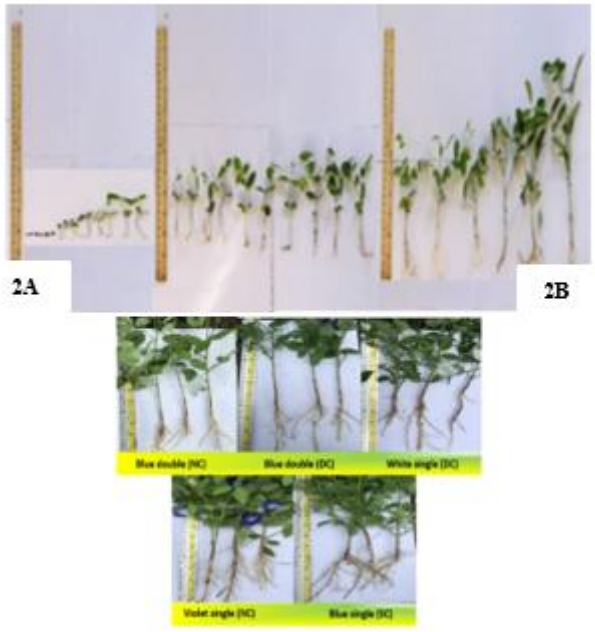

Figure 2A: Stages of seedling growth of Clitoria sp. from day 1 to 31.

Figure 2B: Roots of Clitoria at 60 DAS.

Table 1: Percentage (%) germination at 21 DAS, days to 50% germination, plant height (cm), leaves and leaflet count at 42 DAS, lateral root count and root length (cm) at 60 DAS, measurement of the flowers, pods and seeds (cm) of different Clitoria varieties.University of Southern Mindanao, Kabacan, Cotabato, Philippines. 2020.

Varieties	PG (%) 21 DAS[1/]	DT50G (no. of days)[ns]	PH (cm) 42 DAS[2/]	Leaf Count 42 DAS[ns]	Leaflets Count 42 DAS[ns]	Leaflet Length (cm)[ns]	Leaflet Width (cm)[ns]	LR Count 60 DAS[2/]	Root Length 60 DAS[ns]	FL (cm)[1/]	FW (cm)[ns]	PL (cm)[1/]	PW (cm)[ns]	SL (cm)[1/]	SW (cm)[1/]
Blue-double (NC)	71.15^{ab}	7.00	86.88^{a}	17.05	63.85	5.37	3.35	34.62^{c}	13.77	5.12^{ab}	3.80	8.50^{c}	0.81	7.80^{a}	4.50^{a}
Blue-double (DC)	71.91^{b}	7.25	89.50^{a}	17.10	65.95	5.42	3.35	31.75^{c}	15.95	4.62^{b}	3.30	8.38^{c}	0.82	7.60^{b}	4.50^{a}
White-single (DC)	57.25^{c}	8.00	77.00^{ab}	21.35	80.50	5.52	3.25	47.00^{b}	15.25	5.50^{a}	3.92	10.00^{a}	0.83	6.60^{e}	4.50^{a}
Violet-single (NC)	67.10^{b}	7.75	82.15^{a}	21.05	74.80	5.20	3.17	60.25^{ab}	14.15	5.65^{a}	3.42	8.57^{c}	0.83	7.20^{c}	4.40^{b}
Blue-single (SC)	74.60^{a}	7.75	69.15^{b}	27.05	72.95	5.35	3.43	72.00^{a}	14.62	4.70^{b}	3.60	9.20^{b}	0.82	6.80^{d}	4.30^{c}
P-value	0.000	0.468	0.033	0.052	0.670	0.341	0.484	0.0130	0.823	0.043	0.057	0.000	0.937	0.000	0.000
CV (%)	4.890	11.150	10.330	22.050	24.410	3.940	6.160	31.010	19.310	9.700	8.100	3.400	4.210	1.240	0.830

1- Means in the same column with common letter superscripts are not significantly different at 1% level (Tukey's Test).

2- Means in the same column with common letter superscripts are not significantly different at 5% level (Tukey's Test).

ns – not significant

Legend:PG – Percentage germination; DT50G – Days to 50% germination; PH – Plant height; LR – Lateral

Root; FL – Flower length; FW – Flower width; PL – Pod length; PW – Pod width; SL – Seed length; SW – Seed width; North Cotabato (NC), Davao City (DC), South Cotabato (SC).

Table 2: Flower weight (mg), number of days from sowing to first bud emergence, flower bud emergence to flower opening, flower bud emergence to full bloom, percentage (%) of flowers turned into pods, pod weight (mg), pod count in one kilogram, number of seeds per pod, seed weight (mg) and number of seeds in one kilogram of different Clitoria varieties. University of Southern Mindanao, Kabacan, Cotabato, Philippines. 2020.

Varieties	FWt (mg)[1/]	DSFBE (ND)[1/]	FBEFO (ND)[1/]	FBEFB (ND)[1/]	FTP (%)[1/]	PWt (mg)[ns]	PCOK (count)[ns]	NSPP (count)[1/]	SWt (mg)[ns]	SCOK (count)[1/]
Blue-double (NC)	588.00^{a}	40.50^{cd}	12.50^{a}	14.25^{c}	30.82^{c}	615.00	1,630.52	6.50^{b}	64.50	15,933.33^{b}
Blue-double (DC)	573.00^{b}	42.50^{bc}	12.25^{a}	14.75^{bc}	30.00^{c}	595.00	1,691.75	6.75^{b}	63.30	16,350.00^{b}
White-single (DC)	265.00^{e}	46.75^{a}	9.25^{b}	12.00^{d}	65.00^{a}	660.00	1,519.25	9.45^{a}	47.50	21,383.33^{a}
Violet-single (NC)	286.00^{d}	45.00^{ab}	11.00^{a}	15.50^{ab}	69.15^{a}	557.50	1,810.30	8.50^{a}	55.10	18,200.00^{ab}
Blue-single (SC)	303.00^{c}	37.75^{d}	12.00^{a}	15.75^{a}	46.67^{b}	638.50	1,573.50	9.00^{a}	52.40	19,166.67^{ab}
P-value	0.000	0.000	0.007	0.000	0.000	0.069	0.062	0.000	0.076	0.0360
CV (%)	2.330	4.450	9.510	3.740	13.730	8.730	8.690	8.660	15.370	12.740

1- Means in the same column with common letter superscripts are not significantly different at 1% level (Tukey's Test). ns – not significant

Legend: FWt – Flower weight; ND – Number of days; DSFBE – Days from Sowing to First Bud Emergence; FBEFO - Flower bud emergence to flower opening; FBEFB – Flower bud emergence to full bloom; FTP – Flowers turned into pods; PWt – Pod weight; PCOK – Pod count in one kg; NSPP – Number of seeds per pod; SWt – Seed weight; SCOK – Seed count in one kg.

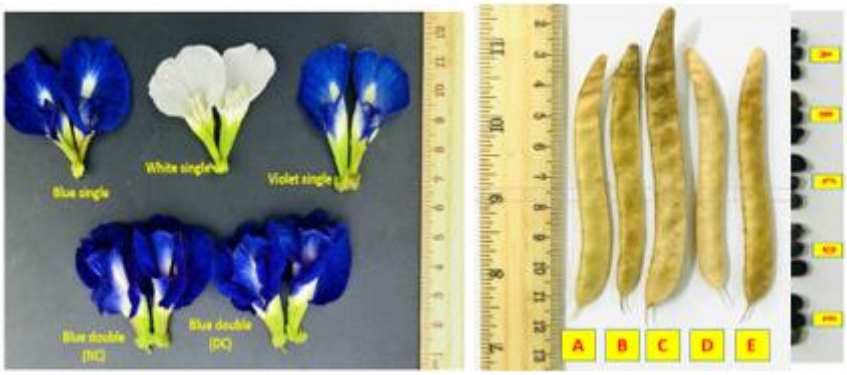

Figure 3: Flowers (A), pods (B) and seeds (C) of different Clitoria varieties: A – Blue double (NC), B – Blue double (DC), C – White single (DC), D – Violet single (NC) and E – Blue single (SC)

3.2 Developmental Stages of Clitoria Buds to Flowers and Pods

The developmental stages and maturity of buds into flowers, flower senescence to pod formation up to pod senescence were shown in Figure 5. Formation of the buds up to full-bloom of the flowers took 17 days and

flower senescence had approximately 5 to 7 days prior to the visibility of the pod production. Regular monitoring of the pod size was done. The maximum pod size was already attained at 45 days after bud emergence or 31 days after the pod visibility. After attaining the maximum size, only changes in the color and appearance of the pods were observed. As shown in Figure 6, the pods of five varieties of Clitoria revealed differences on their maturity and development.

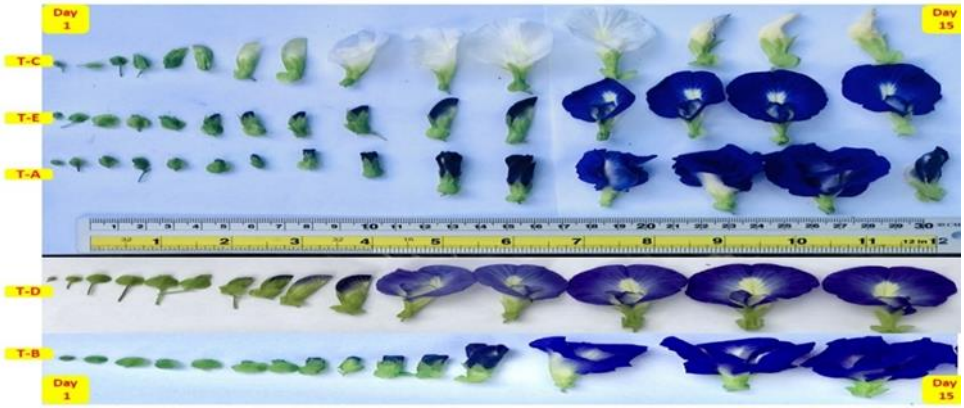

Figure 4: Developmental stages of flowers of different Clitoria varieties from the formation of the visible buds at Day1 up to the status of the flowers at Day15: T-A – Blue double (NC), T-B – Blue double (DC), T-C – White single (DC), T-D – Violet single (NC) and T-E – Blue single (SC).

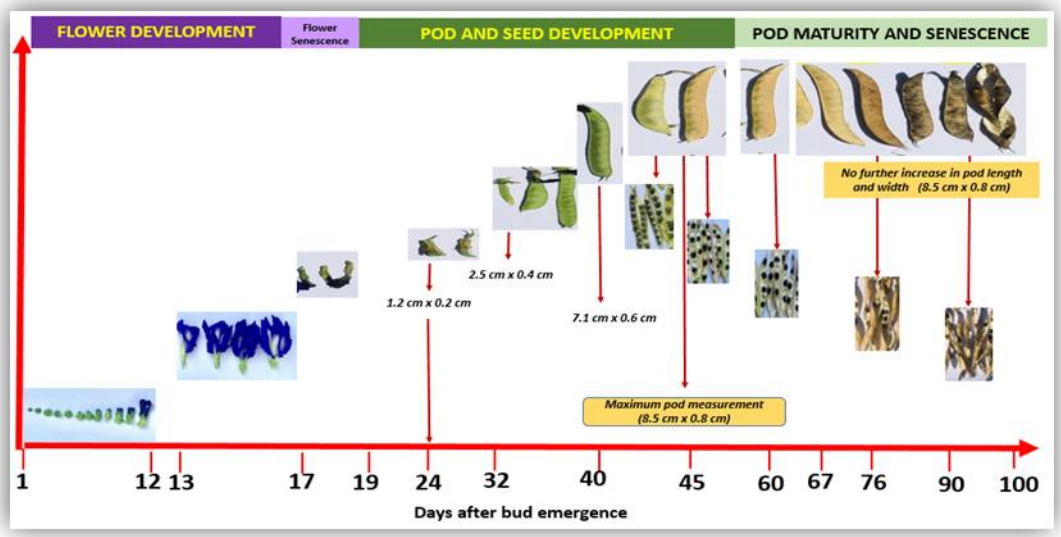

Figure 5: Flower and pod development of Blue-double layered variety of Clitoria.

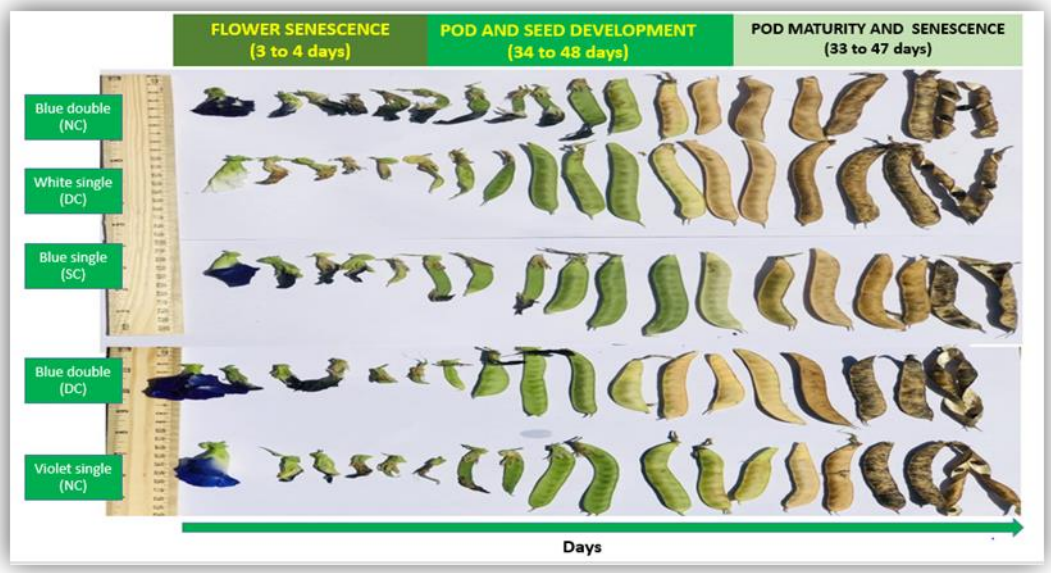

Figure 6: Stages of pod development from flower drying stage up to pod maturity and senescence.

Just like other legumes, the common bean plants initiate many reproductive structures that can be carried to full seed maturity [8]. The reproductive legume development typically occurs over a period of 40 to 65 days starting from the appearance of the first reproductive bud to harvest maturity [9]. After anthesis, there are reproductive structures at different stages of development at any given moment. There is considerable genetic variation for the time pods require to reach maturity which can vary within a plant on source-sink limitations, biotic and abiotic stresses [10].

3.3 Flower, Pod and Seed Yield of Clitoria sp

The yield of different Clitoria varieties in Table 3 was based on the performance of the flowers, pods and seeds which are all critical components of the productivity of blue ternate plants. In this study, the term 'FloPoSe' was coined from the words flowers, pods and seeds of Clitoria and FloPoSe data refer to the summation of the flowers, pods and seed yield of Clitoria plants. With regards to flower production at 240 DAS, highest flower yield of 10.98 t/ha was noted from Blue-double (DC) variety and was statistically comparable with Blue-double (NC) with 10.02 t/ha. Lowest production was generally observed in the three single-layered varieties with statistically comparable yield ranging from 1.16 to 1.88 t/ha. In terms of the production of pods, White-single (DC) variety had the highest yield of 3.23 t/ha. This was followed by the Violet and Blue-single Clitoria having a statistically comparable result, while the lowest number of pods produced was observed in the two Double-layered Clitoria. Highest seed yield (1.53 t/ha) was obtained by White-single (DC) and was statistically higher compared to the other single varieties and most especially to the varieties with double-layered petals. There is variation in seed yields between Clitoria accessions [11]. One accession (CP49664) which contained double petal arrangement is a potential ornamental, but has poor seeding ability in the field. Moreover, the 'FloPoSe' yield obtained highly significant differences among treatment means. The very high count of flowers throughout the duration of the study as observed in the two double-layered varieties greatly contributed to their

highest FloPoSe yield of 12.35 t/ha (DC) and 11.19 (NC). In the case of the single-layered varieties, low yield was greatly affected by the limited flower count even though higher pod and seed count were observed on these entries. Blue-single variety (SC) obtained the lowest FloPoSe yield of 4.31 t/ha which was mainly due to variety's low flower and pod count up to 240 DAS, and light seed weight (52.4 mg/seed). In terms of number of fully-bloomed flowers from 150 to 240 DAS, highly significant differences were recorded as illustrated in Figure 7. The drop in the number of flowers at 150 DAS was due to the development from flowers to pods as also shown in Figure 8 wherein there was also a consistent increase on the pod count per plant most especially in the single-layered varieties than the double-layered. It can be noted that throughout the duration of the study, the White-single variety attained the highest number of pods per plant and was statistically higher than the other two single varieties (Blue- and violet-single) with a statistically comparable pod count. Lowest pod count was observed in double-layered Clitoria. Double petal arrangement of Clitoria has poor seeding ability in the field [12].

Table 3: Cumulative yield (t/ha) of the flowers, pods and seeds of different Clitoria varieties from 60 and 240 DAS. University of Southern Mindanao, Kabacan, Cotabato, Philippines. 2020.

Varieties	Flower Yield (t/ha)[1/]							Pod Yield (t/ha)[1/]		Seed Yield (t/ha)[1/]		FloPoSe (t/ha)[1/]
	60 DAS	90 DAS	120 DAS	150 DAS	180 DAS	210 DAS	240 DAS	180 DAS	240 DAS	180 DAS	240 DAS	
Blue-double (NC)	0.07^{a}	0.45^{a}	1.04^{a}	1.40^{a}	3.44^{a}	6.51^{a}	**10.02^{a}**	0.24^{c}	**0.70^{c}**	0.16^{c}	**0.48^{c}**	**11.19^{a}**
Blue-double (DC)	0.05^{b}	0.41^{a}	0.99^{a}	1.36^{a}	3.64^{a}	7.30^{a}	**10.98^{a}**	0.24^{c}	**0.84^{c}**	0.15^{c}	**0.53^{c}**	**12.35^{a}**
White-single (DC)	0.02^{c}	0.11^{b}	0.32^{b}	0.37^{b}	0.54^{b}	0.80^{b}	**1.16^{b}**	0.94^{a}	**3.23^{a}**	0.45^{a}	**1.53^{a}**	**5.91^{b}**
Violet-single (NC)	0.03^{c}	0.17^{b}	0.41^{b}	0.49^{b}	0.88^{b}	1.38^{b}	**1.88^{b}**	0.54^{b}	**1.85^{b}**	0.30^{b}	**1.02^{b}**	**4.75^{bc}**
Blue-single (SC)	0.02^{c}	0.15^{b}	0.37^{b}	0.48^{b}	0.77^{b}	1.18^{b}	**1.62^{b}**	0.46^{bc}	**1.76^{b}**	0.24^{bc}	**0.92^{b}**	**4.31^{c}**
P-value	0.000	0.001	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.001	0.000	0.000
CV (%)	31.26	38.85	22.07	21.62	15.77	21.72	13.90	30.85	23.65	28.96	22.40	10.54

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Legend: FloPoSe - Flower Yield + Pod Yield + Seed Yield

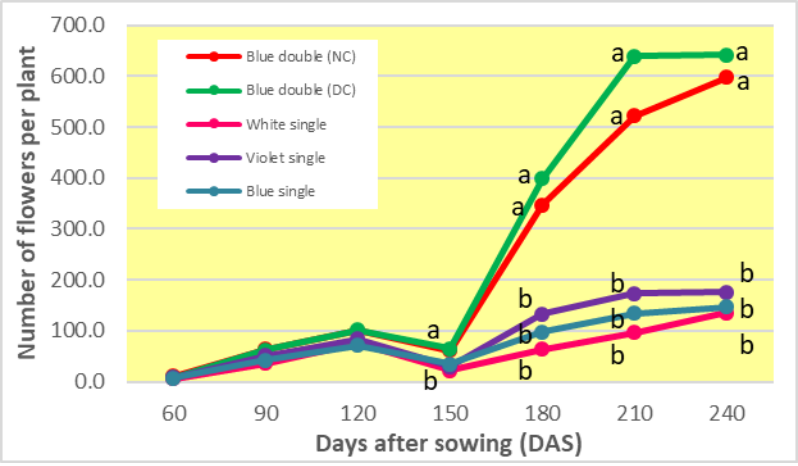

Figure 7: Number of flowers per plant of different Clitoria varieties at 60 to 240 DAS.

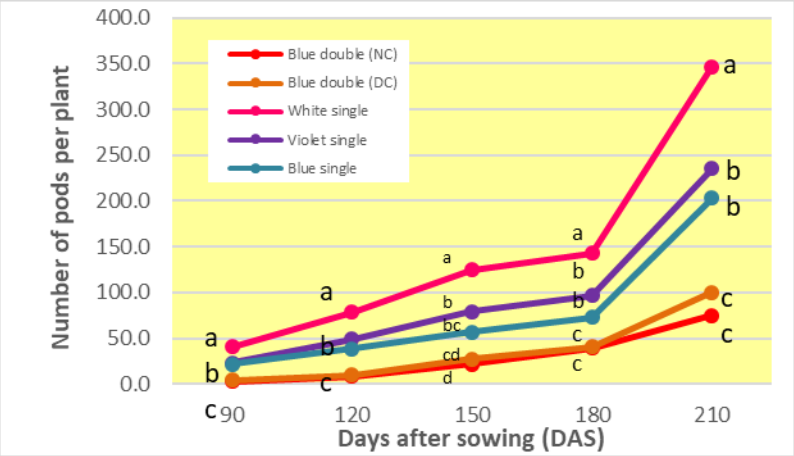

Figure 8: Number of pods per plant of different Clitoria varieties at 60 to 210 DAS.

Yield result in this study implies that the number of flowers per plant (NFP), number of pods per plant (NPP) and number of seeds per pod (NSPP) were the main determinants of FloPoSe yield among all examined variables. Hence, best variety of crop that can compensate for losses of seed yield due to buds, flowers and young pods abscission may contribute to stable seed yield under stressful conditions. An increase in pod count per plant generally results in increasing yield [13]. The upper limits on seed size and seed count per pod are confined genetically; however, these two components can still fluctuate enough to produce sizable yield increases. Pod count per plant significantly affects seed yield [14].

3.4 Other Qualitative Descriptions of Clitoria Plant

Qualitative traits, usually encoded by one or sometimes few numbers of genes, usually do not change in response to the environment. These include features on the plant that is either present or not present, depending on whether the gene responsible for that trait is functional (present) or non-functional (absent). Other qualitative attributes and descriptive characteristics of Clitoria are presented in Table 4.

Table 4: Qualitative characteristics of different plant organs of Clitoria. University of Southern Mindanao, Kabacan, Cotabato. 2020.

Plant Organs	Characteristics	Remarks
Leaves	Arrangement: Opposite	Two leaves arising from the same level (at the same node) at the opposite sides of the stem.
	Venation: Pinnate	Leaflets are borne in pairs along the rachis without a single terminal leaflet
	Type: Odd pinnately compound	Leaflets occurring on each side of a common axis; there is a terminal leaflet and an odd number of leaflet
	Leaflets' shape:	
	Ovate	Egg-shaped leaves in which broader end of the leaf found at the nearest petiole.
	Elliptic	Middle portion of the leaf is the broadest part and the two ends are equally narrow.
Flowers	Color: Blue violet and white	
	Shape: Pea, butterfly	Having the corolla-shaped like a butterfly.
	Type: single- or double-layered petals (paired or solitary)	
	Papilionaceous	Flowers contain both male and female reproductive components
	Zygomorphic	
	Corolla with five free petals	
	Calyx with five sepals	
Stems	Sub-erect pubescent	Twining and fine
Pods	Fruit type: Legume	Flattened with thick margins; compressed; dehiscent when dry; sparsely pubescent or glabrous when mature.
	Color: Green, brown	
	Shape: Linear to oblong	
Seeds	Color: Brown/black/olive	Shiny and dark; flattened; often mottled
	Shape: Oblong	
Roots	Thick and horizontal	Extensive deep root system

3.5 Fungal Disease Occurrence

Fungal disease symptoms were noted starting at 18 WAS, with 3.60 to 8.02% disease infection (Table 5). Symptoms observed were tiny yellow flecks and brown spots surrounded with yellow halo on the leaves. As the disease progressed, extensive death tissue (blight) and enlargement of the lesions and yellowing of the leaves, rotten roots with white mycelial growth were observed (Figure 9). Increased disease infection was noted at 22 to 26 WAS which resulted in wilting, defoliation and death of the whole plant. Highest infection (19.67%) was recorded in Blue-single (SC) and was statistically comparable to other varieties except White-single (DC). The result of pathogenicity tests revealed that the isolated fungi were pathogenic. Fusarium sp. was isolated from the infected leaves while Rhizoctonia sp. was from the roots of the diseased plants. The cultural and morphological characteristics of the two isolates were shown in Table 6. Fusarium had septate, hyaline and boat-shaped conidia, and smooth, white and septate conidiophores; while Rhizoctonia had no conidia and conidiophores (Figure 10). Fungal pathogens causing leaf diseases in blue ternate were Cercospora, Colletotrichum, Oidium and Rhizoctonia [15]. Severe damages of blue ternate caused by Rhizoctonia solani which destroyed majority of the Clitoria accessions under study [16].

Table 5: Percentage (%) infection of fungal disease of different Clitoria varieties at 18, 22 and 26 WAS. University of Southern Mindanao, Kabacan, Cotabato. 2020.

Varieties	Percentage Infection (%)		
	18 WAS[ns]	22 WAS[1/]	26 WAS[1/]
Blue-double (NC)	7.15	10.70[ab]	13.40[ab]
Blue-double (DC)	7.12	13.40[a]	16.07[a]
White-single (DC)	3.60	5.38[b]	8.05[b]
Violet-single (NC)	8.02	15.17[a]	17.88[a]
Blue-single (SC)	5.35	11.60[a]	19.67[a]
P-value	0.283	0.024	0.047
CV (%)	47.400	32.170	33.120

1/a-aaMeansainatheasameacolumnawithacommonaletterasuperscriptsaareanotasignificantly differentaata5%alevel (Tukey's Test).

ns - not significant

Figure 9: Occurrence of disease symptoms at 26 WAS: defoliation of the infected plants (A), wilting and death of the entire infected plant (B); and manifestation of the disease symptoms on the leaves (C), stems and roots of inoculated Clitoria seedlings (D).

Table 6: Cultural and morphological characteristics of fungal isolates from infected Clitoria plants. University of Southern Mindanao, Kabacan, Cotabato, Philippines. 2020.

Fungal Isolates	Cultural Characteristics						Morphological Characteristics	
	Shape	Color	Texture	Density	Zonation	CD	Conidia	Conidiophores
Fusarium sp.	Circular	White	Cottony	Medium	Absent	42.25 mm	Septate, hyaline boat-shaped	Smooth, white, septate
Rhizoctonia sp.	Circular	White	Cottony	Medium	Absent	42.50 mm	Absent	Absent

Legend: CD = Colony diameter at 7 days after incubation

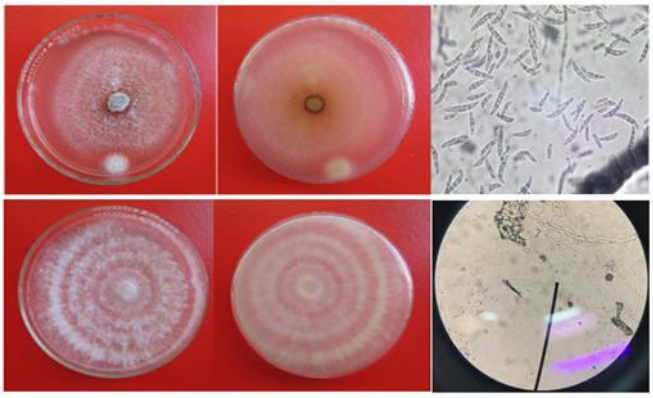

Figure 10: Pure cultures and microscopic photos of Fusarium sp. (A) and Rhizoctonia sp. (B)

3.6 Nutrient Assessment of Clitoria Flowers and Seeds

Mineral contents of the flowers of Clitoria coming from three varieties, specifically the Blue-colored varieties which were single- and double-layered and the White-single and proximate analysis of the seeds of Blue-double variety were presented in Table 7. Results of the nutrient analysis showed that the flowers of Clitoria are rich in Nitrogen, Potassium and Calcium. Flowers of Blue-single variety had numerically higher amount of N, P, K, Ca, Mg, ash content and dry matter. Sodium content was highest in Blue-double, highest pH in White single, and all varieties had TSS of 4.0 In proximate analysis of Clitoria seeds, the parameters determined were carbohydrates (36.69%), total sugar (4.92%), crude fat (12.26%), crude protein (40.59%) and sodium content (76.29 µg/g). The findings indicate that varietal differences of Clitoria vary also on the content or amount of nutrients they contain. Mineral nutrition is necessary to maintain good health [17]. Minerals like calcium, phosphorus, potassium and sodium are essential to a healthy diet, while some minerals such as mercury, lead, cadmium and aluminium are toxic. There were previous research studies which performed mineral and proximate composition analysis of Clitoria.

Table 7: Mineral contents of flowers and proximate analysis of seeds of Clitoria sp. University of Southern Mindanao, Kabacan, Cotabato. 2020.

Parameters	*Clitoria* flowers			*Clitoria* seeds	
	Blue double		*White single*		*Blue single*
Nitrogen (%)	3.70	3.20	4.10	Carbohydrates, kcal/100g	419.00
Phosphorus (%)	0.30	0.29	0.48	Carbohydrates (%)	36.69
Potassium (%)	2.03	1.97	2.08	Total Sugars (%)	4.92
Calcium (%)	1.60	1.90	2.00	Crude Fat (%)	12.26
Magnesium (%)	0.25	0.24	0.29	Crude Protein (%)	40.59
Sodium (%)	0.15	0.14	0.12	Sodium (µg/g)	76.29
Ash content	7.40	4.20	8.30		
Dry matter (%)	91.35	91.97	92.36		
pH	4.18	4.34	4.32		
TSS (°Brix)	4.00	4.00	4.00		

3.7 Diversity Analysis

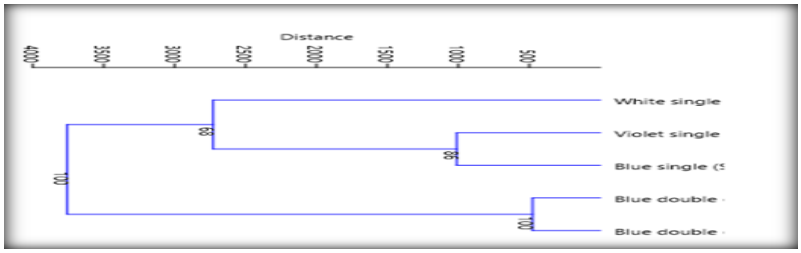

Figure 11: UPGMA dendrogram showing similarities among Clitoria varieties.

Cophen Coor: 0.76 Clustering method : Unweighted Pair Group Method with Arithmetic Mean (UPGMA) Dissimilarity index: Euclidean distance

All morphological traits and nutritive/mineral compositions were used to estimate phenotypic diversity. The dendrogram for phylogenetic relationships, according to UPGMA, showed that the five Clitoria varieties were clustered into three groups (Figure 11). The first cluster included the White single variety. The second cluster was composed of the two single layered varieties (Violet- and Blue-colored). The two double-layered varieties (Blue double NC and DC) were grouped on the third cluster. The cophenetic correlation of the dendrogram to the respective genetic distances was 0.76. The genomic sequences of four Clitoria genotypes (C. ternatea white, C. ternatea blue, C. ternatea double petalloid, and C. biflora) were not clustered together even though the origin was the same for all the genotypes [18]. In addition, the cluster analysis on the genetic similarity between the flower color types, i. e. blue and white in each population showed that all the accessions were separated according to their geographical locations rather than morphological character [19].

4. Summary, Conclusion and Recommendation

The results of the study can be summarized as follows:

1. Variations in morphological characteristics of different varieties of Clitoria were observed, particularly in terms of seed germination, plant height, production, measurement and weight of flowers and pods, and mineral/nutrient compositions of the flowers.

2. Varietal differences did not significantly influence some agronomic parameters such as leaf and root count, leaf measurement, root length, measurement and weight of individual seed.

3. Differences in the developmental stages from bud emergence to maturity stage and senescence of the flowers and pods of different Clitoria varieties were noted.

4. Clitoria variety with double-layered petals produced more flowers per plant, heavier weight of individual flower, resulting in higher flower yield (10.50 t/ha) compared to varieties with single-layered petals (1.55 t/ha).

5. Clitoria variety with single-layered petals produced more pods per plant, longer pod length (9.26 cm), and more seeds per pod (9.00 seeds) than double-layered petals (with 8.44 cm pod length, 6.65 seeds per pod), resulting in higher pod yield (2.28 t/ha) and seed yield (1.16 t/ha) than double-layered varieties (0.77 t/ha pod and 0.51 t/ha seed yields).

6. Among the Clitoria varieties, White variety significantly obtained the most number of pods per plant and seeds per pod, longest and heaviest pod which resulted in the highest pod and seed yield.

7. In terms of nutritional contents of the flowers of Blue-single, Blue-double and White-single varieties, the flowers of Blue-single variety had numerically higher amount of N, P, K, Ca, Mg, ash content and dry matter.

8. Pathogenicity test was conducted and two fungal pathogens were isolated: Fusarium sp. and Rhizoctonia sp.

9. The diversity analysis based on the available morphological traits and nutritional composition revealed that the five Clitoria varieties were grouped into three clusters: a) Blue-double varieties (from North Cotabato and Davao City), b) Blue- and violet-single varieties, and c) White-single variety.

Based on the above findings, it can be concluded that varietal differences in Clitoria also varied with their morphological characteristics and nutritive values. Differences in the structure of flowers also resulted in differences on their ability to produce or develop pods. Some varieties were more productive in terms of flowers, some were good in terms of pods and seeds. Based on the results of the study, it can be recommended that the choice of Clitoria variety should be based on the intended purpose of the plant. For the purpose of flower production, a double-layered variety is recommended; but for pod and seed production, single-layered varieties are more preferred.

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