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Impact of Maturity Stage on Physicochemical, Phytochemical Characteristics and Antioxidant Activity of Seeds of *Phaseolus Lunatus* (Fabaceae) Three Cultivars Consumed in Ivory Coast

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Abstract To enable growers and provide consumers with *Phaseolus lunatus* (L.) seeds of good nutritional quality, this study was carried out on *Phaseolus lunatus* seeds obtained from pods of white, red and black cultivars harvested at stage 1 (32 days), stage 2 (38 days), stage 3 (45 days) and stage 4 (52 days) after fertilisation. Weight of pods and seeds at harvest decreases from stage 1 to stage 4 of maturity. Major biochemical compounds such as crude protein, carbohydrates and lipids of seeds are highest at stage 4 (52 days) of maturity. Seeds of the black cultivar are richer in protein, with a content varying between 17.51 ± 0.17 and 21.21 ± 0.18 % at stage 4 (52 days) of maturity. Seeds of black cultivar produced at stage 4 (52 days) are rich in vitamins B1, B2, B6 and B9 and antioxidants such as carotenoids and flavonoids. These seeds contain high levels of vitamins B6 (pyridoxine) and B9 (folic acid). The concentrations of these vitamins vary from 1900 to 2000.03 µg/100g of dry matter and from 599.93 ± 0.86 to 600 ± 1.00 µg/100g of dry matter respectively. P. lunatus seeds obtained at stage 4 (52 days) were rich in minerals. The Na/K ratio of white, red and black bean seeds varies from 0.04 to 0.05, i.e. less than 1. As a result, eating these *Phaseolus lunatus* bean seeds would probably reduce high blood pressure. In conclusion, *Phaseolus lunatus* seeds have good nutritional quality, the pods must be harvested at stage 4 (52 days) after fertilisation in order to satisfy consumers.

Keywords: *maturity physiology, pods, nutrients, phaseolus lunatus*

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1. Introduction

Beans represent a considerable source of micronutrients, such as iron, calcium, phosphorus, magnesium and zinc. Beans contain a good level of bioactive compounds. These substances can influence human health, having an impact on various activities at the physiological or cellular level. Beans improve our health, counteract the aging process, prevent chronic diseases, extend life expectancy and support the bodily structure or function [1]. The human body's inability to neutralize free radicals, individuals must resort to foods that are able to neutralize them. Beans provide beneficial levels of antioxidant capacity and substances that are able to prevent or slow down oxidation in easily oxidized materials. Beans protect the body against the action of free radicals by slowing down the aging process and fighting cell degeneration and cell death [2]. Among these beans, the lima bean is an under-utilized seed legume in Ivory Coast [3]. There seeds are commonly consumed in rural country of Ivory Coast [3]. It can either be consumed solely or cooked in combination with cereals such as rice or tuber such as yam [4]. Lima beans can also serve as substitute for expensive soy meal and groundnut meal which constitute the major portion of conventional protein sources used in composite livestock feeds [5]. Lima beans possess good nutritional profile being a good source of minerals, dietary fibers, carbohydrates and proteins but, it is low in fat [6,7]. The seeds contain protein twice as much present in cereals with more balanced profile of essential amino acids including lysine which is lacking in cereals [8]. They are many varieties of lima bean [9] of all these varieties the

immature and mature seeds are the main products. Reports show that both the seeds and leaves can be eaten as pot herb when they are young and tender [10,11]. It is a nutritious food stuff which is cultivated primarily for immature vegetables or mature dry seeds [12]. Seed development is the period between fertilizer and maximum fresh weight accumulation and seed maturation begins at the end of seed developments and continues till harvested [13]. Maximum seed quality may be achieved at the end of seed filling period [14] or slightly after this phase [15]. Knowledge of seed development is essential to successful seed production and crop improvement. A better understanding of optimum harvesting time for *Phaseolus lunatus* (L.) seed contributes to improve quality and quantity of seed produced. This research was carried out to investigate the changes in seed quality of white ; red and black beans rapeseed cultivars at different stages of development and maturity in order to determine the appropriate time for harvest and quality improvement.

2. Material and Methods

2.1. Materials

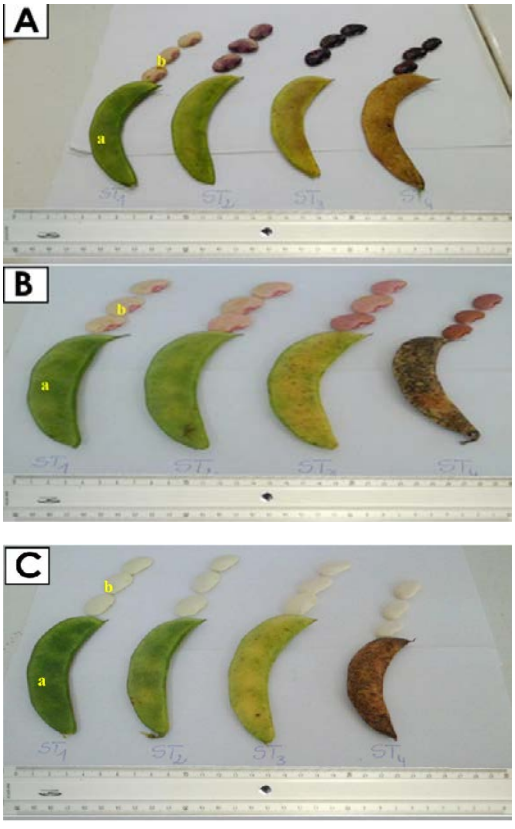

Figure 1. Color evolution of pods (a) and seeds (b) of cultivars (A: black); (B: red) and (C: white) (Tchumou, Biochemistry laboratory at Technical High School of Yopougon, December, 2014). ST1 (green pod), ST2 (more green than yellow pod), ST3 (more yellow than green pod) and ST4 (brown pod).

White, red and black cultivars of lima beans used for this research work, has been cultived from February 2014 to January 2015 at the experimental station of Tomasset (Azaguié, town from 38.7 km to Abidjan). Pods from the three cultivars were harvested at stages (1, 2, 3 and 4) after fertilization [3] (Figure 1).

2.2. Methods

The harvested pods were kept in a food cooler until they reached the laboratory. The seeds were extracted from each pod, washed and oven-dried (Memmert, Germany) at 60 °C for 72 h [16]. The dried powdered samples obtained were stored in polythene bags at 4°C until for analysis.

2.3. Determination of Physical and Biochemical Parameters

2.3.1. Color Determination of Pods and Seeds

The ICL (International Commission for Lighting) L*, a*, and b* color system was used to measure the colors of the pods and seeds of Phaseolus lunatus (L.). The Cie Lab coordinates (L*, a*, b*) were directly read with a spectrophotocolorimeter MS/Y-2500 (Hunter lab, In., Reston, VA, USA), calibrated with a white tile. Color values were recorded as L* (Lightness) – the vertical coordinate runs from L* = 0 (black) through grey to L* = 100 (white); a*(-a, greenness, +a, redness) – the horizontal co-ordinate, that runs from -a* (green) through grey to +a* (red) and b* (-b, blueness, +b, yellowness) – another horizontal co-ordinate, that runs from -b* (blue) through grey to +b* (yellow) [17,18]. The measurements were repeated on four different pods and seeds randomly selected locations at the surface of each sample.

2.3.2. Proximate Analysis of Samples

Moisture, ash, crude protein, crude fat, crude fiber and total sugars were determined respectively by following the standard method [19,20], while Carbohydrate contents were calculated by difference [100- (protein + crude fat + ash + crude fiber)] [21]. In addition to the energy value (EV) was calculated by applying the heat coefficients of [22] according to the following equation: [EV (Kcal/100g) = (4 x Protein %) + (4 x Carbohydrate %) + (9 x Fat %)]. The analyses' values were the basis for three determinations.

2.3.3. Mineral Analysis

Minerals were analyzed by the method reported by [23]. The ash obtained from 1 g of sample was dissolved in 10 % HCl, filtered with filter paper and made up to standard volume with deionised water. Flame photometry method reported by [20] was used to determine sodium and potassium contents of the sample. Calcium, Fe, Mg, Zn and Cu were determined using Atomic Absorption Spectrophotometer (AAS). Phosphorus was estimated colorimetrically (UV-visible spectrophotometer, Model DR 2800/United States)

2.3.4. Vitamin B Determination

All fresh seed of Phaseolus lunatus (L.) was washed and dried weighed 50 mg and cut into small pieces and extracted with 0.1 NHCl on a water bath at a suitable temperature and period. All extracts were filtered through 0.40 micron filter and taken into 100 mL volumetric flashes, and volume was added up for mobile phase. Stock of standard prepared by dissolving 0.01 g of each standard in 100 mL of mobile phase followed by successive

dilutions. HPLC equipped with UV detector and supelco discovery C-18 column (25 cm in lengthand 0.45 internal diameter) was used for analysis. Mobile phase was 50 mL MK2HPO4 and MeOH (70 :30) at 1 mL/min flow rate and 10 µL of each sample/standard was injected and monitored at UV 254 nm by [24].

2.3.5. Anti-Nutritional Factors Estimation

Hydrogen cyanide was analysed by the [23]. Ten (10) g of flour were homogenized in 200 mL of distilled water. The trapped distillate was left to stand for 3 hours and filtered through whatman paper. The filtrate obtained was distilled from 20 mL of sodium hydroxide (0.1 N) and 2 mL of KI (0.02 N). The distillate was titrated with silver nitrate AgNO3 (0.02N) until a yellowish haze appears. The tannin content was estimated spectrophotometrically by the procedure described by [25]. One millilitre of methanolic extract is introduced into a test tube to which 5mL of vanillin reagent were added. The tube was left to stand for 20 min in the dark and the absorbance was read with a spectrophotometer at 500 nm against a blank. The blank was prepared for each test by adding 5 mL of distilled water to the test tubes replacing the vanillin reagent. The amount of tannins in the sample was determined using a standard range established from a tannic acid solution (2 mg / mL) under the same conditions as the test. Phytic acid was determined using the procedure described by [26]. One gram of flour was homogenized in 20 mL of HCl (0.65). The mixture obtained is stirred for 12 hours at room temperature. The mixture was centrifuged at 3000 trs/min for 40 minutes. To 0.5 mL of supernatant, 3 mL of Wade's reagent were added. The blank was prepared for each sample with 0.5 mL of distilled water in the test tubes without Wade's reagent. The tubes were left to stand for 20 min in the dark and the optical density was read with a spectrophotometer at 490 nm against a blank. The amount of phytate was determined using a standard range established from a sodium phytate solution (10 mg / mL) under the same conditions as the test. The oxalate was determined using the method of [27]. Two grams of flour were homogenized in 75 mL of H2SO4 (3M). The mixture was stirred magnetically for 1 hour at room temperature. The whole was filtered through Whatman filter paper. Twenty five milliliters of filtrate were titrated hot with a solution of potassium permanganate (KMnO4, 0.05 M) until the change to persistent pink.

2.3.5. Antioxidant and Phytochemicals

Determination of antioxidant activity in the bean samples was determined using the 1, 1-diphenyl-2-pycrylhydrazyl (DPPH) method, as reported by [28]. The results were expressed as milligrams of Trolox equivalents antioxidant capacity (TEAC g−1) extract. Total phenolic content (TPC) in extracts from the samples was determined by a Folin-Ciocalteu method described by [29], using garlic acid as standard.

2.4. Statistical Analysis

Results for physico-chemical, nutritional and phytochemical parameters within each cultivar, according to harvest stage and interaction (Cultivars × Harvest stage) were processed using Statistica 7.1 software (StatSoft Inc). Two-factor analyses of variance (ANOVA) were used to compare the parameters measured. The completion of the ANOVA and the equality of the variances were first checked using Levene's test. When a significant difference was observed between the modalities of each parameter, the ANOVA was completed by multiple comparisons using the Newman Keul test. Differences were considered significant at the 5% threshold.

3. Results

3.1. Color Changes in Phaseolus lunatus Pods and Seeds

The color of *Phaseolus lunatus* pods and seeds varies according to maturity stage (Table 1 and 2). Pod L* values for white, red and black cultivars increase from stage 1 (32 days) and 3 (45 days), then decrease from stage 3 (45 days). L* values vary respectively from 32.47 ± 0.88 to 45.48 ± 0.56; 45.48 ± 0.56 to 35.12 ± 0.37 and 32.42 ± 1.67 to 38.36 ± 0.46 for the three cultivars. Pod a* values were negative from stage 1 (32 days) to stage 3 (45 days) for both white and black cultivars. The a* values range from 3.92 ± 0.25 to - 10.05 ± 0.45. Pod a* values are negative from stage 1 to stage 4 in the red cultivar (-0.28 ± 0.02 to -10.05 ± 0.45), then become positive at stage 4 (52 days) in the white and black cultivars (+2.31 ± 0.13 to +3.7 ± 0.25). Finally, all b* pod values are positive for all cultivars, increasing significantly ($P< 0.05$) from stage 1 (32 days) to stage 3 (45 81 days), then decreasing at stage 4 (52 days) of maturity. They increase from 18.45 ± 1.55 to 29.26 ± 0.36 and decrease from 29.26 ± 0.36 to 16.72 ± 0.12. All pod luminance (L*) values for the three cultivars ranged from 100 (white) to -100 (black).

Red cultivar pods have a negative (a*) value from stage 1 (32 days) to stage 4 (52 days). Pods of the white and black cultivars have a negative (a*) value from stage 1 to stage 3, then become positive at stage 4. All pods of the three cultivars have a positive (b*) value (yellow). The L* seed values of P. lunatus decrease during ripening in the red and black cultivars (52.13 ± 2.01 to 34.65 ± 0.66 and 52.57 ± 0.38 to 27.50 ± 0.18), while they increase in the white cultivar (51.82 ± 0.17 to 59.29 ± 0.49). The a* seed values of the three cultivars differed significantly ($P < 0.05$) according to maturity stage. Phaseolus lunatus a*-seed values were negative at stage 1 (32 days) of maturity in seeds from all three cultivars (-2.61 ± 0.77 to -9.20 ± 0.21). White cultivar a* seed values are negative after stage 1 (32 days). However, they become positive for the red and black cultivars (+2.85 ± 0.47 to +9.75 ± 1.55). The b*-seed values for the red and white cultivars show no significant variation ($P > 0.05$), while they decrease significantly ($P < 0.05$) for the black cultivar (16.77 ± 0.87 to 6.50 ± 0.30). The (L*) seed value of the white cultivar varies between 0 and 100 (white), while that of the red and black cultivars decreases (black). The (a*) seed value of the white cultivar is negative during ripening (green-colored seed), while that of the red and black cultivars is negative at stage 1 and then becomes positive from stage 2 (38 days) to stage 4 (52 days) (red-colored seed). Seed (b*) values for all three cultivars are positive from stage 1 (32

days) to stage 4 (52 days) of maturity (yellow seed color). Chroma (C*) values for pods are higher than those for seeds, and are positive for all cultivars. Chroma (C*) values for white, red and black cultivars ranged respectively from 22.02 ± 0.38 to 20.8 ± 0.21; from 21.07 ± 0.73 to 17.47 ± 1.62 and from 20.78 ± 1.58 to 20.21 ± 1.33 from stage 1 (32 days) to stage 4 (52 days) of ripening. Seed chroma (C*) values for white and red cultivars showed no significant difference ($P > 0.05$) during ripening, with the exception of those for the black cultivar, which decreased (17.22 ± 0.63 to 8.65± 0.18). Pod hue angle (H*) values for white, red and black cultivars increased significantly ($P < 0.05$) during ripening. On the other hand, hue angle (H*) values for seeds of the white cultivar did not decrease significantly during ripening. Pod hue angle (H*) values for white, red and black cultivars varied respectively from 62.83 ± 0.44 to 83.60 ± 0.20; from 65.81 ± 0.39 to 89.01 ± 0.72 and from 62.53 ± 1.50 to 79.40 ± 1.25 from stage 1 (32 days) to stage 4 (52 days) of ripening. Seed hue angle (H*) values for the white cultivar did not vary significantly ($P > 0.05$) during ripening. Seed hue angle (H*) values for the red and black cultivars decreased, with the exception of the white cultivar, which increased during ripening. Seed hue angle (H*) values for red and black cultivars ranged from 81.80 ± 1.66 to 57.54 ± 1.06 and from 76.87 ± 0.41 to 65.19 ± 1.08 respectively. All chroma (C*) and hue angle (H*) values for pods and seeds of the three cultivars are positive, whatever the maturity stage.

Table 1. Changes pods color of *Phaseolus lunatus* (L.) during maturity

Cultivar	Stages of Maturity (days)	L*gs	a*gs	b*gs	C*gs	H*gs
	32	37.47±0.88^{ef}	-10.01± 0.18^{gh}	19.52±0.47^{fg}	22.02±0.38^{d}	62.83±0.44^{e}
	38	42.32±1.64^{bc}	-8.61 ± 0.29^{e}	24.20±0.55^{bc}	25.68±0.39^{c}	70.35±0.48^{c}
CW	45	45.48 ±0.56^{a}	-3.92 ± 0.25^{f}	29.52 ±0.36^{a}	29.60±0.43a	82.36±0.46^{b}
	52	37.31 0.69^{ef}	+2.31 ± 0.13^{c}	20.66±0.49^{df}	20.8 ± 0.21^{d}	83.60±0.20^{ab}
	32	36.33±0.67^{ef}	-10.05 ± 0.45^{g}	19.30±0.61^{fg}	21.97±0.73^{d}	65.81±0.39^{cde}
	38	40.41±0.26^{cd}	-9.66 ± 0.33^{gh}	22.68±0.61^{cd}	24.66±0.69^{c}	66.93 0.27^{cde}
CR	45	43.20±0.49^{ab}	-5.22 ± 0.25^{b}	25.66 ±0.09^{b}	26.19±0.11^{c}	78.54±0.54^{b}
	52	35.12±0.37^{f}	-0.28 ± 0.02^{d}	16.72 ±0.12^{e}	17.47 1.62^{b}	89.01 ± 0.72^{a}
	32	32.42 ±1.67^{f}	-9.57± 0.46^{h}	18.45±1.55^{eg}	20.78±1.54^{d}	62.51 ± 1.50^{e}
CB	38	36.98±1.78^{ef}	-9.41 ± 0.66^{eh}	19.29±1.29^{fg}	21.42±1.43^{d}	63.99 ±0.52^{de}
	45	38.36±0.46^{de}	-7.43 ± 0.04^{a}	19.76±1.03^{fg}	21.11±0.96^{d}	69.61 ±1.36^{cd}
	52	37.51±0.53^{ef}	+3.7 ± 0.25^{f}	19.86±1.37^{fg}	20.21±1.33^{d}	79.40 ± 1.25^{b}

Mean ± SD, n=3; in the columns, means marked with different letters indicate a significant difference at the threshold of ($P< 0.05$). VW (White Cultivar), CR (Red Cultivar) and CB (Black Cultivar)

Table 2. Evolution of *Phaseolus lunatus* seed color during maturity

Cultivar	**Stages of Maturity (days)**	**L*gr**	**a*gr**	**b*gr**	**C*gr**	**H*gr**
	32	51.82±0.17^{g}	-9.20 ± 0.21^{a}	16.77±0.29^{e}	19.13±0.22^{a}	61.25±0.85^{cd}
	38	52.36±0.47^{g}	-7.26 ± 0.20^{b}	16.17±0.29^{de}	17.73±0.24^{g}	65.82 ±0.40^{d}
CW	45	54.51±0.57^{d}	-7.20 ± 0.43^{b}	16.00±0.47^{de}	17.55±0.10^{g}	65.18 ±0.93^{d}
	52	59.29±0.49^{a}	-6.63 ± 0.41^{b}	15.10±0.28^{ad}	16.68±0.23^{f}	66.33 ±0.98^{d}
	32	52.13±2.01^{g}	-2.61 ± 0.77^{c}	16.10±1.09^{de}	17.03±0.47^{fh}	81.80 ±1.66^{a}
	38	46.55±0.33^{e}	+5.76 ± 0.76^{b}	11.82 ±0.40^{b}	12.97±0.17^{b}	64.13 ±2.63^{b}
CR	45	44.71±0.76^{e}	+9.75 ± 1.55^{a}	12.37 ±0.91^{b}	15.77±0.21^{d}	53.03±1.30^{bc}
	52	34.65±0.66^{c}	+8.92 ± 0.38^{a}	14.03 ±0.33^{a}	16.27±0.40^{df}	57.54±1.06^{cd}
	32	52.57±0.38^{dg}	-3.91 ± 0.32^{c}	16.77 ±0.87^{e}	17.22±0.63^{g}	76.87 ±0.41^{a}
CB	38	37.25 ±1.05^{b}	+3.33 ± 0.44^{c}	6.83 ± 0.63^{c}	7.60 ± 0.34^{e}	66.40±0.95^{cd}
	45	28.78 ±0.76^{f}	+2.85 ± 0.47^{c}	6.50 ± 0.30^{c}	7.01 ± 0.21^{c}	68.95 ±1.37^{d}
	52	27.50 ±0.18^{f}	+3.50 ± 0.41^{c}	7.91 ± 0.28^{c}	8.65 ± 0.18^{e}	65.19 ±1.03^{d}

Mean ± SD, n=3; in the columns, means marked with different letters indicate a significant difference at the threshold of ($P< 0.05$). CW (White Cultivar), CR (Red Cultivar) and CB (Black Cultivar)

3.2. Biochemical Composition of Seeds From Three *Phaseolus lunatus* (L.) Cultivars According to Maturity Stage

A study of the relationship between seed biochemical composition and maturity seeds stage of three lima bean cultivars showed that moisture content, carbohydrate content and energy value decreased significantly ($P < 0.05$) with maturity for all cultivars (Table 3). Moisture content varied between 70.37 ± 0.29 and 29.68 ± 0.75 g/100 g dry matter content in seeds of white, red and black varieties ranged from 69.88 ± 0.17 to 33.30 ± 0.34 g/100 g dry matter and from 72.68 ± 0.16 to 35.36 ± 0.33 g/100 g dry matter. Carbohydrate content in *P. lunatus* seeds decreased significantly ($P < 0.05$) with maturity. It ranged from (71.36 ± 0.34 to 64.16 ± 0.09 g/100 g dry matter) for the white cultivar ; from (73.17 ± 0.25 to 69.63 ± 0.48 g/100 g dry matter) for the red cultivar ; and from (72.88 ± 0.05 to 67.95 ± 0.28 g/100 g dry matter) for the black cultivar. Regardless of stage of maturity, seeds of red varieties contained more carbohydrates than seeds of black and white varieties. The energy values of seeds of all three *P. lunatus* cultivars decreased significantly ($P < 0.05$), from 314.67 ± 0.32 to 300.79 ± 0.59 Kcal/100 g dry matter. The seeds of the red variety have the highest energy value. It fluctuates between 314.54 ± 2.50 and 311.25 ± 0.33 Kcal/100 g dry matter. Seeds of the white variety had the lowest energy values, ranging from 310.66 ± 1.94 to 300.79 ± 0.59 Kcal/100 g dry matter. Regardless of the stage of maturity, the seeds of red varieties have the highest energy value. With the change of maturity stage, the contents protein, lipid and vitamin C in the seeds lima bean increased significantly ($P < 0.05$). The protein content of seeds of all three varieties increased significantly ($P < 0.05$), from 17.22 ± 0.13 to 25.06 ± 0.13 g/100 g dry matter. The protein content of white variety seeds is higher than that of black and red variety seeds. The dry matter content of all three *P. lunatus* seeds ranged from 17.22 ± 0.13 to 25.06 ± 0.13 g/100 g. At maturity stage 4 (52 days), the seeds of the white variety contained more protein than the seeds of the black and red varieties. The protein content is between 19.30 and 25.06 g/100 g dry matter. The lipid content of seeds of white, red and black varieties increased significantly with maturity ($P < 0.05$). They were between 1.01 ± 0.01 and 1.40 ± 0.01 g/100 g dry matter ; 1.05 ± 0.01 to 1.60 ± 0.01 g/100 g and 1.1 ± 0.0 to 2.16 ± 0.08 g/100 g dry matter. At maturity stage 4, red varietie produce seeds with higher lipid content. The vitamin C content of white, red and black seeds increased significantly ($P < 0.05$), from 2.30 ± 0. 40g/100g fresh weight to 8.63 ± 0. 20g /100g fresh weight They were between 2.30 ± 0.40 and 5.81 ± 0.25 g/100 g dry matter ; 3.1 ± 0.11 to 6.23 ± 0.20 g/100 g dry matter and 5.23 ± 0.41 to 8.63 ± 0.20 g/100 g dry matter. The seeds of the black variety contain more vitamin C than the seeds of the white and red varieties. The average ash content of white, red and black cultivars did not increase significantly during ripening ($P > 0.05$). They were between 3.37 ± 0.10 and 4.17 ± 0.00 g/100 g dry matter ; 2.37 ± 0.10 to 4.26 ± 0.05 g/100 g dry matter and 3.42 ± 0.04 to 3.74 ± 0.05 g/100 g dry matter. The fourth stage of ripening produces black seeds rich in vitamin C. There was no significant difference in the crude fiber content of the seeds of the three varieties with different maturity stages ($P > 0.05$). They ranged between 4.89 ± 0.30 and 5.13 ± 0.15 g/100 g dry matter ; dry matter contents from 4.75 ± 0.18 to 5.1 ± 0.26 g/100 g, in white, red and black seeded varieties from 5.20 ± 0.20 to 4.93 ± 0. 11g / 100g. The seeds of the red variety are rich in fiber at all stages of maturity (Table 3).

3.3. Mineral Composition of Seed Flours From Three *Phaseolus lunatus* Cultivars as a Function of Maturity Level

Sodium, potassium and iron levels in the seeds of the three *Phaseolus lunatus* cultivars (white, red and black) increased significantly ($P < 0.05$) with maturity stage (Table 4). They range respectively from 38.68 ± 0.28 to 75.27 ± 0.72 mg/100 g; 724.20 ± 1.05 to 1592.9 ± 6.38 mg/100 g; 6.36 ± 0.17 to 12.56 ±0.28 mg/100 g dry matter. At stage 4 (52 days), seeds from the red cultivar contained more iron than seeds from the white and black cultivars. Iron content ranged from 9.13 ± 0.18 to 12.56 ±0.28 mg/100 g dry matter. Phosphorus, magnesium and calcium levels in the seeds decrease during ripening. They vary respectively from 569.67 ± 0.89 to 230.18 ± 1.13 mg/100 g; 160.14 ± 0.66 to 128.46 ± 0.55 mg/100 g; 669.32 ± 0.76 to 301.03 ± 1.00 mg/100 g dry matter. The most abundant minerals in P. lunatus seeds are potassium, phosphorus, magnesium and calcium. Potassium is the most abundant mineral in the seeds of all three *P. lunatus* cultivars. Seeds of the red cultivar contain high concentrations of potassium, calcium and magnesium. The highest potassium concentration was obtained at stage 4 (52 days) of maturity. The potassium content of the cultivars seeds ranged from 1043.3 ± 1.25 to 1592.9 ± 6.38 mg/100 g dry matter. Average zinc concentrations did not vary significantly during ripening. They ranged from 0.16 ± 0.03 to 1.70 ± 0.01 mg/100 g dry matter. Copper content increased in seeds of the white cultivar as a function of ripening stage. This increase was not significant ($P \geq 0.05$). It varied from 1.87 ± 0.04 to 2.40 ± 0.05 mg/100 g dry matter. Stage 4 (52 days) of maturity produces seeds of the black cultivar rich in sodium, copper and zinc, while at the same stage seeds of the red cultivar are rich in potassium, iron and calcium. The white cultivar produces phosphorus rich seeds at stage 4 of maturity.

Table 3. Biochemical composition of seeds from three *Phaseolus lunatus* (L.) cultivars according to maturity stage

Flours	Stages of Maturity (days)	Moisture (%)	Proteins (%)	Lipids (%)	Glucides (%)	Fibers (%)	Ashs (%)	Value Energy (Kcal/100g)
	ST1(32)	70.37 ± 0.29^{d}	19.30 ± 0.32^{di}	1.01 ± 0.01^{g}	71.36 ± 0.34^{e}	4.89 ± 0.17^{b}	3.37 ± 0.10^{f}	310.71 ± 1.93^{de}
	ST2(38)	65.31 ± 0.61^{b}	20.56 ± 0.15^{ch}	1.22 ± 0.01^{def}	67.15 ± 0.33^{b}	4.90 ± 0.05^{b}	4.53 ± 0.10^{e}	306.42 ± 0.63^{a}

Flours	Stages of Maturity (days)	Moisture (%)	Proteins (%)	Lipids (%) (%)	Glucides (%) (%)	Fibers (%) (%)	Ashs (%) (%)	Value Energy (Kcal/100g)
FCW	ST3(45)	55.36 ± 0.63^{g}	23.69 ± 0.11^{b}	1.34 ± 0.01^{de}	65.16 ± 0.17^{c}	5.01 ± 0.1^{b}	4.58±0.13^{e}	302.55 ± 0.81^{b}
	ST4(52)	29.68 ± 0.75^{c}	25.06 ± 0.13^{a}	1.40 ± 0.01^{d}	64.16 ± 0.09^{c}	5.13 ±0.07^{ba}	4.17 ±0.00^{cde}	300.79 ± 0.59^{b}
FCR	ST1(32)	69.88 ± 0.17^{d}	17.22 ± 0.13^{g}	1.05 ± 0.01^{fg}	73.17 ± 0.25^{d}	4.85 ± 0.15^{b}	2.37 ± 0.01^{b}	314.67 ± 0.32^{c}
	ST2(38)	62.54 ± 0.69^{e}	18.25 ±0.14^{ef}	1.20 ± 0.03^{fg}	72.10 ±0.28^{de}	5.02 ± 0.16^{b}	2.86 ± 0.20^{a}	312.97±0.58^{cd}
	ST3(45)	55.35 ± 0.34^{g}	19.41 ± 0.24^{i}	1.34 ± 0.02^{de}	71.55 ± 0.12^{e}	4.73 ± 0.14^{b}	2.82 ± 0.10^{ab}	312.43±0.33^{cde}
	ST4(52)	33.30 ± 0.34^{f}	20.63 ± 0.20^{h}	1,60 ± 0.01^{c}	69.63 ± 0.48^{a}	4.62 ± 0.04^{b}	4.26 ± 0.05^{ce}	311.25 ±0.33^{de}
FCB	ST1(32)	72.68 ± 0.16^{a}	17.51 ±0.17^{fg}	1.1 ± 0.06^{fg}	72.88 ± 0.05^{d}	5.20 ±0.11^{ab}	3.42 ± 0.04^{f}	312.51±0.10^{cde}
	ST2(38)	61.26 ± 0.48^{e}	18.47 ±0.19^{de}	1.6 ± 0.02^{c}	71.16 ± 0.11^{e}	5.15 ±0.14^{ab}	3.85 ±0.03^{cdf}	311.59 ±0.76^{de}
	ST3(45)	57.35 ± 0.39^{g}	19.70 ±0.17^{ci}	1.87 ±0.06^{b}	69.48 ± 0.14^{a}	5.70 ± 0.17^{a}	3.55 ± 0.07^{f}	310.96 ±0.09^{de}
	ST4(52)	35.36± 0.33^{f}	21.21 ± 0.18^{h}	2.16 ± 0.08^{a}	67.95 ± 0.28^{b}	4.93 ± 0.08^{b}	3.74±0.05c^{df}	310.02 ±0.68^{de}

Mean ± SD, n = 3; in columns, means marked with different letters indicate significant differences at the threshold (P < 0.05). FCW (White Cultivar Flour), FCR (Red Cultivar Flour) and FCB (Black Cultivar Flour).

Table 4. Mineral composition of seed flours from three *Phaseolus lunatus* cultivars as a function of maturity level

Flours	Stages of Maturity	Na	K	P	Mg	Fe	Ca	Na/K	Ca/P
FCW	ST1(32)	38.68 ±0.28^{j}	724.20 ± 1.05^{l}	459.85 ±1.18^{b}	156.05 ±0.36^{f}	6.80 ± 0.15^{fg}	53032± 0.58^{g}	0.05	1.15
	ST2(38)	39.49 ±0.40^{j}	899.16 ± 1.04^{k}	399.30 ±0.75^{f}	150.67 ±0.32^{b}	7.45 ± 0.07^{eg}	480.8 ± 1.48^{h}	0.04	1.2
	ST3(45)	53.79 ± 0.1^{g}	981.43 ± 1.62^{i}	330.69 ±0.30^{h}	147.75 ±0.50^{e}	7.55 ± 0.13^{eg}	401.33±1.23^{d}	0.05	1.21
	ST4(52)	60.80±0.60^{f}	1109.3 ± 0.81^{f}	259.25 ±1.08^{j}	140.60 ±0.53^{g}	10.33±0.17^{bd}	340.5 ± 0.51^{e}	0.05	1.31
FCR	ST1(32)	56.81 ±0.15^{j}	1043.3 ± 1.25^{h}	569.67±0.89a	160.14 ±0.66^{a}	9.13 ± 0.18^{c}	669.32±0.76^{a}	0.05	1.17
	ST2(38)	63.40±0.40^{e}	1359.20±1.07^{d}	430.35 ±0.25^{d}	157.43 ±0.66^{f}	10.78±0.11^{dc}	529.69±0.30^{g}	0.04	1.23
	ST3(45)	69.12±0.12^{c}	1450.50±1.15^{c}	340.46 ±0.80^{g}	146.45 ±0.10^{e}	10.86 ±0.08^{d}	479.33±0.87^{h}	0.04	1.4
	ST4(52)	72.65±0.32^{b}	1592.9 ± 6.38^{a}	250.48 ±0.67^{k}	140.56 ±0.20^{g}	12.56 ±0.28^{a}	359.32 ±0.89^{i}	0.04	1.43
FCB	ST1(32)	46.12±0.87^{h}	909.23 ± 0.75^{j}	470,55 ±0,40^{c}	155.72 ±0.72f	6.36 ± 0.17^{f}	539.50±0.70^{b}	0.04	1.14
	ST2(38)	55.60 ±0.40^{i}	1086.80±1.05^{g}	410,83 ±0,97^{e}	139.63 ±0.55^{g}	6.66 ± 0.24^{fg}	460.18±0.86^{c}	0.04	1.12
	ST3(45)	67.62±0.37^{d}	1209.2 ± 0.64^{e}	320,55 ±0.64^{i}	135.95 ±1.12^{c}	7.80 ± 0.10^{e}	359.47 ±0.74^{i}	0.05	1.12
	ST4(52)	75.27±0.72a	1519.50±1.40^{b}	230.18 ±1.13^{l}	128,46 ±0,55^{d}	9.49 ± 0.27^{bc}	301.03 ±1.00^{f}	0.05	1.3

Mean ± SD, n = 3; in columns, means marked with different letters indicate significant differences at the threshold (P < 0.05). FVW (White Cultivar Flour), FCR (Red Cultivar Flour) and FCB (Black Cultivar Flour)

Table 5. B-vitamin composition of seeds from three *Phaseolus lunatus* cultivars consumed in Ivory Coast (mg/100 g dry matter)

Flourss	Stages of maturity (days)	B1	B2	B3	B6	B9
FCW	ST1(32)	160.15 ±.66^{g}	130.05 ± 2.19^{l}	ND	1471.00 ± 1.47^{f}	500.67 ± 2.42^{b}
	ST2(38)	170.00±1.55^{f}	145.05 ± 0.52^{k}	ND	1500.00 ± 2.48^{h}	500.37 ± 1.10^{b}
	ST3(45)	180.27±0.52^{i}	170.07 ± 0.83^{j}	ND	1500.00 ± 2.54^{h}	500.00 ± 3.24 ^{b}
	ST4(52)	210.80±1.11^{d}	189.90 ± 0.84^{i}	ND	1500.00 ± 2.16^{h}	499.37 ± 0.56^{b}
FCR	ST1(32)	180.05±0.94^{i}	240.00 ± 1.46^{h}	ND	1650.00 ± 2.22^{d}	500,45 ± 1,46^{b}
	ST2(38)	190.05±1.13^{e}	258.00 ± 2.48^{g}	ND	1500.00 ± 1.58^{h}	500,75 ± 0,95^{b}
	ST3(45)	200.02±1.39^{h}	270.02 ± 0.68^{f}	ND	1600.00 ± 1.36^{e}	499,13 ± 0,43^{b}
	ST4(52)	230.05±0.82^{b}	280.00 ± 0.90^{e}	ND	1679.72 ± 0.98^{c}	575,53 ± 0,81^{a}
FCB	ST1(32)	180.00±0.74^{i}	400.00 ± 0.50^{d}	ND	1900.00 ± 1.42^{a}	599.93 ± 0.86^{a}
	ST2(38)	200.02±0.66^{h}	403.27± 1.25^{c}	ND	1800.00 ± 1.10^{b}	600.35 ± 1.00^{a}
	ST3(45)	220.35±0.75^{c}	409.75 ± 1.10^{b}	ND	2000.05 ± 0.46^{g}	599.60 ± 0.49^{a}
	ST4(52)	239.97±0.77^{a}	420.25 ± 0.50^{a}	ND	2000.03 ± 1.32^{g}	600.00 ± 1.09^{a}

Mean ± SD, n=3; in columns, means marked with different letters indicate significant differences at the threshold (P < 0.05). FCW (White Cultivar Flour), FCR (Red Cutivar Flour) and FCB (Black Cultivar Flour)

3.4. B-Vitamin Composition of Seeds From Three *Phaseolus lunatus* Cultivars Consumed in Ivory Coast

The B vitamin composition of the seeds of the three *Phaseolus lunatus* cultivars is shown in the (Table 5). Concentrations of thiamine (B1) and riboflavin (B2) increase during ripening, with the exception of the vitamins pyridoxine (B6) and folate (B9), which remain constant. Thiamine (B1) content increases significantly ($P < 0.05$) in all cultivars. It varies from 160.15 ± 1.66 to 210.80 ± 1.11µg/100g dry matter (white cultivar); from 180.05 ± 0.94 to 230.05 ± 0.82µg/100g dry matter (red cultivar) and from 180.00 ± 0.74 to 239.97 ± 0.77µg/100g dry matter (black cultivar). Vitamin B2 content increased significantly ($P < 0.05$) in all cultivars. Concentrations range from 130.05 ± 2.19 to 189.90 ± 0.84µg/100g dry matter (white cultivar); from 240.00 ± 1.46 to 280.00 ± 0.90µg/100g dry matter (red cultivar) and from 400.00 ± 0.50 to 420.25 ± 0.50µg/100g dry matter (black cultivar). Pyridoxine (B6) and folate (B9) concentrations did not increase significantly ($P > 0.05$) during ripening. They vary respectively from 1471 ± 1.47 to 1500.97 ± 2.16 µg/100g dry matter (white cultivar), from 1650.00 ± 2.22 to 1679.72 ± 0.98 µg/100g dry matter (red cultivar) and from 1900 ± 1.42 to 2000.03 ± 1.32 µg/100g dry matter (black cultivar). Seeds from the red cultivar contained the highest concentration of vitamin B6, compared with seeds from the white and black cultivars. Vitamin B9 content did not vary significantly ($P > 0.05$) across cultivars and ripening stages. The highest vitamin B9 concentration was obtained in the seeds of the black cultivar (599.93 ± 0.86 to 600 ± 1.00µg/100g dry matter) at stage 4 (52 days) of ripening. The black cultivar produces seeds rich in vitamins B1, B2, B6 and B9 at stage 4 (52 days).

3.5. Evolution of Phytochemicals and Antioxidant Activity as a Function of Maturity Stage

Analysis of the phytochemical content of seeds from white, red and black cultivars of *Phaseolus lunatus* (L.) showed a significant decrease ($P < 0.05$) in polyphenols as a function of maturity level. (Table 6). From stage 1 (32 days) to stage 4 (52 days) of harvest, total polyphenols varied from 1310.96 ± 1.00 to 705.85 ± 0.95 mg/100 g DM. The highest polyphenol content was observed in white seeds (1310.96 ± 0.96 to 860 ± 0.85 mg/100 g DM). It was obtained at stage 1 (32 days) of maturity. Phytate concentrations in seeds of white, red and black *P. lunatus* cultivars ranged from 105.36 ± 0.36 to 124.66 ± 0.61 mg/100 g dry matter during ripening. Phytate content increased significantly ($P < 0.05$) from 109.76 ± 0.76 to 120.23 ± 0.92 mg/100 g dry matter in the black cultivar from stage 1 (32 days) to stage 4 (52 days) of ripening. From stage 1 (32 days) to stage 2 (38 days) of maturity, this content increases respectively from 111.38 ± 0.75 to 117.23 ± 0.56 mg/100 g dry matter) in white seeds and from (124.66 ± 0.61 to 130.95 ± 0.17 mg/100 g dry matter) in red seeds before decreasing until 52 days of harvest. The phytate content of the red cultivar, ranging from 124.66 ± 0.61 to 124.37 ± 0.35 mg/100 g dry matter, did not vary significantly ($P < 0.05$). At stage 2 (38 days) of pod harvest, seeds of the red variety contained more phytates than seeds of the white and black cultivars. Oxalate concentrations of all three *P. lunatus* varieties decreased significantly ($P < 0.05$) with harvest stage. The oxalate concentrations of the three *P. lunatus* cultivars ranged from 396.53 ± 0.61 to 615.93 ± 0.90 mg/100 g dry matter. At harvest stage 1 (32 days), white seeds contained more oxalates than red and black seeds. On the other hand, at stage 4 (52 days) of harvest, oxalate concentration is higher in red seeds. The tannin content of *P. lunatus* seeds increases significantly ($P < 0.05$) with maturity stage. They range from 82.75 ± 1.05 to 133.81 ± 0.77 mg/100 g dry matter. Tannin content higher in white seeds. It ranged from 94 111.16 ± 0.69 to 133.81 ± 0.77 mg/100 g dry matter. Seeds of the white cultuvar have higher tannin concentrations at stage 4 of maturity than seeds of the red and black cultivars. Analysis of flavonoid levels shows an increase in the seeds of all three *P. lunatus* cultivars from stage 1 to stage 4 of harvest. They range from 3.68 ± 0.27 to 13.62 ± 0.50 mg/100 g dry matter. Black seeds contain a high concentration of flavonoids, ranging from 5.42 ± 0.15 to 13.62 ± 0.50 mg/100 g dry matter, compared with white and red seeds. At stage 4 of maturity, black seeds contain high levels of flavonoids. Carotenoid levels in *Phaseolus lunatus* seeds increase significantly ($P< 0.05$) with harvest stage. They range from 2.83 ± 0.15 to 17.83 ± 0.22 mg/100 g dry matter. Harvest stage 4 shows high carotenoid levels with black seeds, ranging from 4.70 ± 0.11 to 17.83 ± 0.22 mg/100 g dry matter. Chlorophyll content decreased significantly ($P< 0.05$) according to harvest stage with all three *Phaseolus lunatus* varieties. It varied from 3.80 ± 1.96 to 0.20 ± 0.71 mg/100 g fresh matter. The hydrocyanic acid content of *P. lunatus* seeds increased significantly ($P< 0.05$) with harvest stage. It varied from 3.06 ± 0.03 to 4.26 ± 0.06 mg/100 g dry matter according to maturity stage. Red seeds contain more hydrocyanic acid (3.25 ± 0.02 mg/100 g dry matter) than black and white seeds at harvest stage 4.

Antioxidant activity increases in *Phaseolus lunatus* (L.) seeds during ripening, ranging from 81.72 ± 1.96 to 95.30 ± 0.71 in percentage inhibition. Antioxidant activity is highest at stage 4 of ripening in black seeds. It varied from 82.60 ± 0.65 to 95.30 ± 0.71 % DM.

3.6. Anti-Nutritional/Mineral and Mineral/Mineral Ratios of Seeds From Three Phaseolus Lunatus Cultivars as a Function of Maturity Stage

Phytate/iron ratios for seeds from cultivars (white, red, black) range from 10.19 - 16.33; 9.67 - 13.65 and 12.66 - 17.26 respectively. Phytate/Ca ratios for white, red and black cultivars ranged from 0.21 - 0.30; 0.18 - 13.65 and 0.20 - 39. Oxalate/Ca ratios for seeds of the white (CB), red (CR) and black (CN) cultivars ranged from 0.72 to 1.55. Oxalate / (Ca+Mg) ratios ranged from 0.58 to 1.12. Seeds from all three cultivars have Na/K values below 1 and Ca/P values above 1. Phytate/Ca and Oxalate/Ca ratios are below 2.5 for all maturity stages and cultivars

Table 6. Phytochimical composition and Antioxidant Activity of seeds from three *Phaseolus lunatus* cultivars consumed in Ivory Coast (mg/100 g dry matter)

Flours	States of maturity (days)	Polyphenols	Phytates	Oxalates	Tannins	Flavonoids	Caroténoids	hydrocyanic acid	Antioxidant Activity (%)
FCW	St1(32)	1310,96±1,00^{a}	111,38±0,75^{h}	615,93±0,90^{a}	111,16±0,69^{g}	3,68 ±0,27^{c}	2,83± 0,15^{f}	3,11± 0,02^{h}	81,72 ±1,96^{edb}
	St2(38)	911,33 ±0,51^{e}	117,23±0,56^{c}	574,27±0,75^{b}	118,18±0,54^{f}	5,57 ±0,20^{g}	3,91 ± 0,10^{e}	2,14 ± 0,04^{f}	84,86 ±9,03^{edc}
	St3(45)	750 ,86±0,80h	101,12±1,12^{f}	556,38±0,97^{e}	127,78±1,07^{e}	6,99 ±0,30^{f}	6,77 ± 0,28^{h}	4,21 ± 0,01^{g}	86,52±3,19^{edca}
	St4(52)	860,10 ±0,85^{f}	105,36±1,16^{e}	525,20±0,72^{g}	133,81±0,77^{a}	8,74 ±0,14^{e}	8,43 ± 0,21^{g}	4,92 ± 0,11^{c}	89,98 ±1,11^{dca}
FCR	St1 (32)	1046,33±0,76^{c}	124,66±0,61^{g}	563,41±0,36^{c}	110,76±0,21^{g}	5,12 ±0,15^{g}	4,80 ± 0,20^{i}	3,15 ± 0,02^{h}	76,56 ±1,69 ^{b}
	St2 (38)	747,66 ±0,76^{i}	130,95±0,17^{a}	499,24±0,74^{h}	116,13±0,17^{f}	6,87 ±0,12^{f}	8,76 ± 0,30^{g}	4,23 ± 0,02^{g}	79,32 ±0,93^{eb}
	St3 (45)	732,16 ±1,04^{j}	124,80±0,08^{g}	528,13±1,20^{f}	122,15±0,15^{b}	8,66 ±0,22^{e}	13,82 ±0,41^{c}	5,36 ±0,09^{b}c	83,26±3,66^{edcb}
	St4(52)	803,90 ± 0,90^{i}	124,37±0,35^{g}	560,50±0,50^{d}	128,67±1,02^{e}	10,28±0,03^{d}	16,21 ±0,16^{b}	6,37 ± 0,03^{a}	85,06±4,50^{edcb}
FCB	St1 (32)	1247,73±0,50^{b}	109,78±0,51^{h}	444,69±0,33^{i}	82,75 ±1,05^{d}	5,42 ± 0,15^{g}	4,70 ± 0,11^{i}	3,06 ± 0,03^{h}	82,60±0,65^{edcb}
	St2 (38)	925,40 ± 1,00^{d}	111,31±1,00^{h}	352,43±0,51^{l}	99,45 ± 0,31^{h}	7,71 ± 0,08^{b}	7,50 ± 0,30^{h}	3,52 ± 0,22^{e}	86,60±0,70^{edca}
	St3 (45)	609,59 ± 0,52^{l}	114,52±0,53^{d}	411,77±0,25^{j}	100,4 ± 0,65^{h}	9,88 ± 0,28^{d}	12,25 ±0,45^{d}	3,86 ± 0,05^{d}	92,10 ±0,65^{ab}
	St4 (52)	705,85 ± 0,95^{k}	120,23±0,92^{b}	396,53±0,61^{k}	108,42±1,15^{c}	13,62±0,50^{a}	17,83 ±0,22^{a}	4,26 ± 0,06^{g}	95,30± 0,71^{ca}

Mean ± SD, n = 3; in columns, means marked with different letters indicate significant differences at the threshold (P < 0.05). FCW (White Cultivar Flour), FCR (Red Cultivar Flour) and FCB (Black Cultivar Flour).

Table 7. Rapports facteurs antinutritionnels/minéraux et minéraux/minéraux des graines de trois cultivars de *Phaseolus lunatus*

Cultivars	Stades de maturité	Phytate/Fer	Phytate/Ca	Oxalate/Ca	Oxalate/Ca+Mg	Na/K	Ca/P
FCW	ST1(32)	16,33	0,21	1,16	0,86	0,05	1,15
	ST2(38)	15,73	0,24	1,19	0,90	0,04	1,20
	ST3(45)	13,58	0,25	1,38	1,01	0,05	1,21
	ST4(52)	10,19	0,30	1,54	1,09	0,05	1,31
FCR	ST1(32)	13,65	0,18	0,84	0,67	0,05	1,17
	ST2(38)	10,14	0,24	0,94	0,72	0,04	1,23
	ST3(45)	11,49	0,26	1,10	0,84	0,04	1,40
	ST4(52)	9,67	0,34	1,55	1,12	0,04	1,43
FCB	ST1(32)	17,26	0,20	0,82	0,63	0,04	1,14
	ST2(38)	16,71	0,24	0,76	0,58	0,04	1,12
	ST3(45)	14,68	0,31	1,14	0,83	0,05	1,12
	ST4(52)	12,66	0,39	1,31	0,92	0,05	1,30

Mean ± SD, n = 3; in columns, means marked with different letters indicate significant differences at the threshold (P < 0.05). FCW (White Cultivar Flour), FCR (Red Cultivar Flour) and FCB (Black Cultivar Flour)

4. Discussion

4.1. Pods and Seeds Change Color According to Maturity

All three varieties had pod values (L*) between 100 (white) and -100 (black). The pods of the red variety are green (a*) between stage 1 (32 days) and stage 4 (52 days). The pods of the white and black varieties are green (a*) from stages 1 to 3, but turn red in stage 4. All pods of the three varieties were positively (yellow) (b*). (L*) seed values ranged from 0 to 100 (white), for white cultivars and decreased (black) for red and black cultivars. The (a*) seed value of the white variety is green, while the (a*) seed value of the red and black varieties is green at maturity stage 1 (32 days) and turns red at maturity stage 4 (52 days). Seed values (b*) were positive for all three varieties. The pods and seeds change color as they mature. The CIE parameters (L, a* and b*) showed that the pod color of all three cultivars changed from green at stage 1 to yellow at stage 3 and brown at stage 4. Positive b* values decrease during ripening and are responsible for the brown color of stage 4 pods. The white color of the seeds of white varieties is explained by the increase in L* value during ripening. The red seed color of red varieties can be explained by positive a* values. The black seed color can be explained by a decrease in L* value during ripening. The development of pod and seed color can be explained by the breakdown of chlorophyll, which on the one hand leads to the loss of green color, and on the other hand to the new synthesis of colored pigments such as carotenoids, anthocyanins, etc. ([30,31]. Chlorophyll

degradation occurs through the action of ethylene [30,32]. The maturation process has been studied by, enzymes involving only chlorophyllase. The activity of the enzyme increases during maturation, which Associated with reduction of chlorophyll and loss of green color [31,33].

4.2. Biochemical Composition of *Phaseolus lunatus* (L.) Seeds During Maturity Stage

Moisture content decreases during seed maturation. This decrease in water content of *P. lunatus* seeds is most likely due to the utilization of water in various metabolic activities caused by environmental conditions (high) temperature, wind, etc.) [34]. The dehydration of *P. lunatus* seeds is similar to this similar. Consistent with what he observed during his study of *Anethum graveolens* (L) [35]. The seeds of red varieties were higher in carbohydrates than black and white varieties regardless of maturity stage. Seeds of all three *P. lunatus* cultivars showed a significant decrease in carbohydrate content at maturity at the 5% threshold (73.17 – 64.16% dry matter). The decrease in carbohydrate content was due to conversion of seed starch by phosphorylases during ripening caused by soluble sugars [36]. According to [37], the variation in carbohydrate content among three month old pine cultivars was influenced by cultivars. According to the work of [38], the carbohydrate content of the seeds of the three species varied similarly to that of the Crotalaria Enable species, whose dry matter content varied between 66.21 % and 82.16 %. The carbohydrate content of *P. lunatus* seeds is similar to that of Crotalaria, making them a good source of calories against wasting and can be used for infant feeding [38]. At stage 4 of maturity (52 days), seeds of the red cultivar contained more fat than the black and white cultivars.

The fat content in the seeds *Phaseolus lunatus* cultivars varied widely during ripening, ranging from 1.01 to 2.6 g/100 g dry matter. The fat content is higher in the seeds of the black cultivar (1.1 to 2.6% dry matter) than in the white and red cultivars. The decrease in water content in *P. lunatus* seeds is responsible for the increase in seed oil content during ripening [39]. According to the work of [40], the lipid content of the seeds of the three *P. lunatus* cultivars is comparable to that of some lima bean cultivars (1.3-2.3 g/100 g dry matter). Lipids are important compounds in the human diet and play several roles in the human body. Lipids are the main source of energy, more than the energy provided by proteins or carbohydrates [41]. The lima bean is not rich in lipids like most species of the Phaseolus genus, unlike oilseeds such as soy, which are rich in protein and lipids. At stage 4 (52 days), the seeds of the white variety contain more protein than those of the black and red varieties. Its content ranges from 19.30 to 25.06% of dry matter. The protein content in the seeds of the three *Phaseolus lunatus* cultivars increased significantly ($P < 0.05$) with maturity and differed between cultivars. In the seeds of the three *P. lunatus* cultivars, the protein content varied from 17.2 to 25.06 g/100 g dry matter). [42] showed a significant positive correlation ($P < 0.05$) between increasing seed nitrogen content and crude protein content in mungbean seeds during ripening. During ripening, the mass of the seed increases, followed by the accumulation of large amounts of protein, each of which accounts for 40% of the dry mass [43,44] have shown that the crude protein content of tumbleweed, which is 11.2% of dry matter at the beginning of flowering, increases with ripening. The protein content of the seeds harvested after 52 days from the three varieties is higher than that of pulses like *Cicer arietinum* (20.70% DM), *Vigna mungo* (23.60% DM) and *Vigna radiata* (24.50% DM); *Phaseolus vulgaris* (22.4% DM) and Cajanus cajan (22.7% DM) [37]. These values are comparable to the protein contents of the three Vigna mungo varieties. They range from 24.37 to 26.22% dry matter [45]. The protein content of *P. lunatus* white cultivar seeds is lower than that of *Mucuna pruriens var. utilis* white cultivar seeds (28.82% dry matter) [46]. The plant protein from the cultivars of *P. lunatus* is an alternative source of protein in the human diet.

The highest energy value is found in the seeds of the red cultivar, whatever the ripening stage. The energy value decreases significantly at the 5% threshold with increasing maturityThe energy value of the seeds of three cultivars of *P. lunatus* varied between 314.67 and 310.02 Kcal / 100 g of dry matter during the ripening period. In this study, this low energy value can be attributed to the low lipid and carbohydrate content during seed ripening. The energy values obtained with *P. lunatus* seeds are low in comparison with those of seeds of the same species, which are respectively 370.96 Kcal/100 g for the cultivated species and 376.26 Kcal/100 g dry matter for the wild species [47]. The energy value of the seeds of the three *P. lunatus* cultivars is comparable to that of soybean and bean [48], ranging from 1318-1394 Kj (315-333.17 Kcal/100 g dry matter).

Ash content increases significantly with maturity ($P<0.05$). The relatively high ash content of the seeds of all three cultivars is thought to be due to the high concentration of minerals in the seedsThis value is slightly higher than that found in wild and cultivated seeds of *P. lunatus* [49], which are 3.42 g and 2.91 g per 100 g of dry matter, respectively. *Cassia obtusifolia* seeds [50] and Vigna species [51] showed similar results. The ash content of a food is a very important parameter for the assessment of mineral content. At stage 4 (52 days), black seeds are rich in sodium, copper and zinc, whereas red seeds at the same stage are rich in potassium, iron and calcium. At stage 4 of maturity, the white variety produces seeds rich in phosphorus. There is a significant increase ($P<0.05$) in the content of minerals such as sodium, potassium, iron and copper during the ripening period. They vary from 38.68 - 60.80 mg/100 g; 72.26 - 110.43 mg/100 g; 6.80 - 10.33 mg/100 g and 1.84 - 2.4 mg/100 g dry matter, respectively. With ripening, the content of phosphorus, magnesium and calcium decreases. Variety seeds contain phosphorus (55.52 - 37.18 mg/100 g dry matter), magnesium (55.72 - 45.44 mg/100 g dry matter) and calcium (63.32 - 48.24 mg/100 g dry matter). Potassium (K) content in kale leaves increases during development according to [52]. On the other hand, according to the same author, phosphorus tends to decrease during ripening. Phosphorus (P) is an essential element for plant growth and development. The decrease in minerals such as phosphorus, magnesium and zinc during ripening could

also be explained by their use as activators of enzyme-catalysed reactions [53]. Their use in photosynthesis, carbohydrate and nucleic acid metabolism could explain the decline in calcium (Ca) and magnesium (Mg) [54]. Magnesium is an essential component of chlorophyll [55], which contributes to fruit (pod) ripening. Changes in the mineral profile of the seeds of the three cultivars can be explained by genetic variability and the level of mineral uptake from the soil [56], plant metal fertility [57] and soil salinity [58]. The levels of iron and phosphorus are high *Phaseolus lunatus* (L.) seeds contain high concentrations of iron, which can help carry oxygen to tissues at the haemoglobin level. Potassium regulates the body's water balance. Together with sodium, it helps maintain acid-base balance. It is an activator of certain enzyme systems necessary for protein synthesis, glycogen storage, nerve fibre excitability and muscle joints. The Na/K and Ca/P ratios of the seeds of all three varieties are less than or greater than 1, irrespective of the stage of maturity. The Na/K ratio is very important for the proper functioning of the body. It helps to prevent high blood pressure. A food product is a good source of Ca and P if the Ca/P ratio is greater than 1 [59]. The Na/K and Ca/P ratios of the seeds of all three cultivars are less than or equal to 1. They can therefore be recommended for human consumption. *Phaseolus lunatus* seeds can be recommended in the diet of both men and women as their content exceeds the standards proposed by [60] of 1.37 mg/day and 2.94 mg/day, respectively. In addition, according to the work of [61], the seeds of the three *P. lunatus* cultivars can be used in the human diet to reduce anaemia affects several million people worldwide.

4.2.1. Vitamins B

Seeds from the black cultivar contain more B vitamins than seeds from the red and white cultivars at stage 4 (52 days) of maturity. The B-vitamin concentrations of *Phaseolus lunatus* cultivars differ from one cultivar to another. There was a significant difference ($P < 0.05$) in the evolution of thiamine (B1) and riboflavin (B2) concentrations during seed ripening. On the other hand, pyridoxine (B6) and folate (B9) levels did not increase significantly ($P > 0.05$) with seed ripening. Seeds of the white cultivar are rich in thiamine (B1), while those of the black cultivar are rich in riboflavin (B2) and pyridoxine (B6). Pyridoxine (B6) content ranges from 1900 to 2000.03 µg/100g dry matter. P. lunatus seeds have high levels of pyridoxine (B6), as well as thiamine (B1), riboflavin (B2) and folate (B9). Vitamin B content depends on the ripeness of the fruit and vegetables [62]. Vitamins are essential organic compounds found in fruit and vegetables. Soluble B vitamins are necessary for cellular metabolism, especially carbohydrate metabolism. According to [63], the recommended level of vitamin B6 (pyridoxine) in the human diet is 1.3 mg/day or 1300 µg/day of dry matter. Seeds from all three *P. lunatus* cultivars can be used in adult diets, as their pyridoxine (B6) content exceeds the level recommended by [63]. A deficiency in B vitamins can lead to metabolic disorders, manifested by more or less characteristic symptoms (pellagra and beri-beri). *P. lunatus* seeds can therefore be a good source of folates (B9) to prevent megaloblastic anemia in undernourished or malnourished populations, mainly in Africa and specifically in Côte d'Ivoire.

4.3. Polyphenols and Antioxydant Activity

The white cultivar contains more polyphenols than the red and black cultivars at all stages of ripening. Polyphenol levels in Phaseolus lunatus (L.) seeds from the three cultivars range from 1310.96 ± 1.00 to 705.85 ± 0.95 mg/100 g dry matter, depending on ripening stage. Values range from 1310.96 ± 1.00 - 860.10 ± 0.85 mg/100 g dry matter. According to studies by [64], the difference in total polyphenols between bean cultivars can be attributed to genetic differences, agronomic practices, climatic conditions and ripening stages. Levels are lower than those found by [46], which are 3130 ± 0.01 mg /100 g dry matter in the white variety and 2840 ± 0.01mg/100 g dry matter in the black variety of dry bean Mucuna pruriens var. utilis (Wall ex Wight). The seeds of all three P. lunatus cultivars are a good source of polyphenols. They may possess anti-inflammatory properties ([65,66] and be able to modulate immune system function. Consumption of P. lunatus seeds, rich in polyphenols, can protect human cells against oxidation, thus preventing their premature destruction and body wrinkles [67]. The use of these seeds in food recipes should be encouraged. The hydrocyanic acid content of Phaseolus lunatus seeds increases from 3.06 ± 0.03 to 6.37 ± 0.03 mg/100 g dry matter during maturity. The hydrocyanic acid content of the seeds is below the toxic level of 10 mg HCN Eq/kg dry matter recommended for cassava by [68] and comparable to that of Vigna sinensis and Pisum sativum [69]. P. lunatus seeds can be used for human consumption because the hydrocyanic acid is destroyed by heat. Seeds of the red cultivar contain more phytates than those of the white and black cultivars at stage 4 of maturity. Phytate levels increase significantly in the seeds of the black cultivar at the 5% threshold from (109.78 ± 0.51 to 120.23 ± 0.92 mg /100 g dry matter) during ripening, and remain constant in the seeds of the white and red cultivars. Phytates accumulate in the seeds of P. lunatus cultivars during the ripening period in the form of phosphate and inositol [70]. This increase in phytate levels in the seeds is explained by the accumulation of phytates in the form of phosphorus in legume seeds, which can reach up to 80% of the total phosphorus content [71]. According to [72], during seed development, phytic acid is deposited in the organelles. The average phytate content of P. lunatus seeds, ranging from 105.36 ± 1.16 to 130.95 ± 0.17 mg/100 g dry matter, is within the range of values recorded for Dioscorea alata yam (58.6 to 198 mg/100 g dry matter) [73]. Seeds of the white cultivar contain more tannin than those of other cultivars. Tannin levels increase significantly ($P < 0.05$) in the seeds during ripening. Tannin levels in Phaseolus lunatus seeds ranged from 82.75±1.05 to 133.81 ±0.77 mg/100 g dry matter. An increase in seed tannin levels during ripening was observed by [74], who showed that polymerization of tannins in potatoes at stage 7 of ripening. According to [75], tannins are widely distributed in plants, particularly in fruit and cereal seeds. Seeds of the red cultivar contain the highest levels of total oxalates. The total oxalate content of Phaseolus lunatus seeds decreased significantly

($P < 0.05$) from 615.93 ± 0.90 to 396.53 ± 0.61 mg/100 g dry matter during ripening. The oxalate content obtained in P. lunatus seeds is higher than that reported by [76] for Artocarpus altilis flours, which is 66.84 ± 1.54 mg/100 g dry matter. The evolution of oxalate content is explained by a rapid accumulation of oxalates followed by a decrease at the end of ripening [77]. Oxalate content varies according to variety and species. The lethal oxalate content of a feed is between 2000 and 5000 mg oxalates/100 g feed dry matter [78], indicating that consumption of P. lunatus seeds would be safe. Seeds of the black cultivar are richer in flavonoids at stage 4 (52 days) of ripening. Flavonoid levels increase significantly at the 5% threshold during seed ripening for all three Phaseolus lunatus cultivars, regardless of ripening stage. They range from 3.68 ± 0.27 to 13.62 ± 0.50 mg/100 g dry matter. Flavonoid values are higher in the seeds of the red cultivar than in the white and black cultivars. Changes in flavonoid composition during ripening correlate with increased antioxidant activity. Our results are similar to those of [79], who showed an increase in flavonoid levels in Thymus transcaspicus (Klokov) fruits during ripening. Bean seed color is determined by the presence and concentration of flavonol glycosides, anthocyanins and condensed tannins [80]. Results found by [81] showed an increase in flavonoid levels during ripening of Ocinum américanum (L) cultivars ranging from 10 to 749 mg CA/100 g dry matter. This difference is probably due to anthocyanins, a well-known group of water-soluble colorants, which contribute significantly to seed coloration [82]. All plant phenolics possess antioxidant activity. [83] have shown that the antioxidant power of flavonoids can be attributed to the synergy of phenolic compounds (phenolic acids, tannins and flavonoids). The increase in antioxidant activity in the seeds of three P. lunatus plants illustrates the findings of [83]. The results obtained show an increase in flavonoids ranging from 3.68 to 13.62 mg/100 g dry matter, followed by high antioxidant activity (81.72 - 95.30%) during ripening. Interestingly, flavonoids in the seeds of all three P. lunatus cultivars are able to inhibit and/or reduce the production of reactive oxygen species (ROS) by neutrophils [84]. Calcium, magnesium, zinc and iron are divalent cations chelated by oxalates and phytates, reducing their bioavailability [85]. Phytates/Ca, oxalates/Ca, Phytates/Ca and oxalates/Ca+Mg ratios in Phaseolus lunatus seeds are below the critical value of 2.5, whatever the stage of maturity, implying good absorption of these minerals by the organism, according to [86]. As for the phytate/iron ratio, which is higher than the critical value of 0.4 whatever the stage of maturity, this shows the non-bioavailability of iron after consumption of Phaseolus lunatsu seeds.

5. Conclusion

The aim of this study was to determine the pod harvesting stage of three Phaseolus lunatus cultivars, so as to obtain seeds of good nutritional quality. The results showed that most biochemical parameters are at their maximum at stage 4, i.e. 52 days after fertilization, especially in the seeds of the red cultivar. This cultivar is rich in carbohydrates and has a good energy value. Protein levels are higher in the seeds of the white cultivar than in those of the red and black cultivars. The seeds of all three cultivars are rich in minerals, but more potassium. Seeds from the black cultivar contain more B vitamins than seeds from the white and red cultivars. Flavonoid, carotenoid and antioxidant activity levels increase during ripening in the seeds of all three Phaseolus lunatus cultivars. Antioxidant activity is high in seeds harvested at stage 4 of ripening. At stage 4 (52 days), the seeds of all three cultivars can be used as a dietary supplement by Ivorian populations.

References

[1] Chávez-Mendoza, C.; Sánchez, E. (2017). Bioactive compounds from Mexican varieties of the common bean (Phaseolus vulgaris): Implications for health. Molecules, 22, 8. [CrossRef] [PubMed]

[2] Gutierrez-Zavala, A.; Ledesma-Rivero, L.; García-García, I.; Grajales-Castillejos, O. (2007). Capacidad antioxidante total en alimentos convencionales y regionales de Chiapas, México. Rev. Cubana Salud Pública, 33, 1.

[3] Tchumou. (2017): Ethnobotanical survey and physicochemical characterization of bean seeds, Phaseolus lunatus (Fabaceae) consumed in the South and East of Côte d'Ivoire according to maturity level and cooking time, 224p.

[4] El-Gohery, S. (2021). Effect of different treatments on nutritional value of lima bean (*Phaseolus lunatus*) and its utilization in biscuit manufacture. Food Nutr. Sci. 12: 372-391.

[5] Tope, A.K. 2014. Effect of fermentation on nutrient composition and anti-nutrient contents of ground Lima bean seeds fermented with Aspergillus fumigatus, Rhizopus stolonifer and Saccharomyces cerevisiae. Int. J.

[6] Farinde, E.O., O.T. Olanipekun and R.B. Olasupo. (2018). Nutritional composition and anti-nutrients content of raw and processed lima bean (Phaseolus Lunatus). Ann. Food Sci. Technol. 19: 250-2.

[7] Farinde, E., V. Obatolu and S. Fasoyiro. (2017). Microbial, nutritional and sensory qualities of baked cooked and steamed cooked lima beans. Am. J. Food Sci. Nutr. 5:156-161.

[8] Bonita, L.C., G.A. Shantibala-Devi and C. H. Brajakishor Singh. (2020). Lima Bean (Phaseolus Lunatus L.) A Health Perspective. Int. J. Sci. Technol. Res. 9:5638-5649.

[9] Darbie M.G., Williams T.K. and George B. (1999). Lima beans, commercial vegetable production. Georgia Extension services publication, Circular, pp : 13-17.

[10] Daisy E.K. (1979). Food legumes TPI crop product digest No 3. Tropical Product Institute, London.

[11] Van de Maessen, L.G.J and S. Sadikin. (1989). Plant resources of South Eastern Asia., (1):56-60.

[12] Lyman S.M, Baudoin J.P and Hildago R. (1985).Lima beans (*Phaseolus lunatus*) In: Grain Legume Crops. RJ Summerfield and EH Roberts (Eds.) London: Williams Collins sons & Co Ltd LondonUnited Kingdom.: P.477-519.

[13] Mehta C.J., Kuhad M.S., Sheoran I.S. and Nandwal A.S. (1993). Studies on seed development and germination in chickpea cultivars. Seed Res., 21(2): 89-91.

[14] Tekrony DM, Egli DB. (1997). Accumulation of seed vigour during development and maturation.

[15] Ghassemi-Golezani K, Hosseinzadeh-Mahootchy A. (2009). Changes in seed vigour of faba bean (Vicia faba L.) cultivars during development and maturity. Seed Sci. Tech 37: 713-720.

[16] Chinma C.E. and Igyor M.A. (2007). Micro-nutriments and anti-nutritional content of select tropical vegetables grown in south-east, Nigeria. Nig. Food., 25.111-115.

[17] Papadakis S.E., Abudal-Malek S., Kamden R.E. and Yam K.L. (2000). A versatile and inexpensive techniques for measuring colour of foods. Food Technol. 54 (12): 48-51.

[18] Al-Said F.A., Opara U.L. and Al-Yahyai R.A. (2009). Physico-chemical and textural quality attributes ofpomegranate cultivars (Punica granatum L.) grown in the Sultanate of Oman. J. Food Eng. 90: 129–134.

[19] Yellavila, S.B., J.K. Agbenorhevi, J.Y. Asibuo and G.O. Sampson. (2015). Proximate composition, minerals content and functional properties of five lima bean accessions. J. Food Secur. 3: 69-74

[20] AOAC. (1990). Official methods of analysis of the Association of Official Analytical Chemists, 15th ed, Washington DC, 1230p.

[21] Bernfeld. (1955). Amylase β and α, In: method in enzymology 1, Colowick S.P. and Kaplan N.O., Academic Press, pp149-154.

[22] FAO/INFOODS. (2015). FAO/INFOODS Guidelines for verifying food composition data before publication of a user table/database-Version 1.0. FAO, Rome.

[23] Fatima Ismail., Farah N., Talpur. & Memon A.N. (2013). Determination of Water Soluble Vitaminin Fruits and Vegetables Marketed in Sindh Pakistan. Pakistan Journal Nutrition, 12: 197-199.

[24] Atwater and Rosa. (1899). A new respiratory colorimeter and the conservation of energy in human body. Physiol. Rev., 9: 214-251.

[25] Food and Agriculture Organization of the United Nations. (2011). Dietary Protein Quality Evaluation in Human Nutrition. Report of FAO Expert Consultation, Auckland, 31 March-2 April 2011, 27.

[26] Bainbridge Z.K. Tomlins & A. Westby. 1996. Analysis of condensed tannins using acidified vanillin. Journal Food Science Agriculture, 29: 77-79.

[27] Latta M. & Eskin M. (1980). A simple method for phytate determination. Journal Agriculture and Food Chemistry, 67: 1313-1315.

[28] Day R. A. & Underwood A. L. (1986). Analisis Kimia Kuantitatif, Edisi Kelima, Penerbit Erlangga, Jakarta Hal, 388- 390.

[29] Brand-Williams, W., Cuvier, M. E., and Berset, C. (1995). "Use of a free radical method to evaluate antioxidant activity". Lebens-Wissen Technology, 28. 25-30.

[30] Treibitsh T., Goldschmidt E.E. & Riov J. (1993). Ethylene induced de novo synthesis of chlorophyllase, a chlorophyll degrading enzyme in citrus fruit peel. Processing National Academy Science USA, 90: 9441-9445.

[31] Ketsa S., Phakawatmongkol W. & Subhadrabhandu S. (1999b). Peel enzymatic activity and color changes in ripening mango fruit. Journal. Plant physiology, 154: 363-366.

[32] Guis M., Botondi R., Ben-Amor M., Ayub R., Bouzayen M., Pech J.C. & Latché A. (1997). Ripening-associated biochemical traits cantaloupe charentais melons expressing an antisense ACC oxidase transgene-J. American Society Horticulture Sciences, 122: 748-751.

[33] Azuma R., Kurata H., Adachi M. & Shimokawa K. (1999). Degreening of citrus unshiu fruit via ethylene-induced soluble chlorophyllase. Journal Japan Society Hortical Science, 68: 558-562.

[34] Sreeramulu N., Tesha A.J. & Kapuya J.A. (1992). Some biochemical changes in developing seeds of bambarra groundnut (Voandzeia subterranea Thouars). Indian Journal Plant Physiology, 35: 191-194.

[35] Egli D.B. (1997). Seed Biology and the Yield of Grain Crops. CABI International. Wallingford 178pp.

[36] Germain P. & Linden G. (1981). Activités enzymatiques. In: Deymier, B., Multon, J.L., Simon, D (eds) Analyse des constituants alimentaires. Techniques d'Analyse et de contrôle dans les industries agrolalimentaires, Tec. Et Doc Lavoisier, Paris, 4: 211-244.

[37] Apata D.F., & A.D. Ologhobo. (1994b). Proximate composition of some nutritionally valuable minerals and functional properties of three varieties of Lima beans (*Phaseolus lunatus* L.) flour. International Journal Food Sciences Nutrition, 43: 181-191.

[38] Vadivel V. & Janardhanan K. (2000). Chemical composition of the underutilized legume Cassia hirsuta L. Plant Foods Human Nutrition, 55: 369-381.

[39] Granito M., Brito Y. & Torres A. (2007). Chemical composition, antioxidant capacity and functionality of raw and processed (Phaseolus lunatus). Journal Sciences Food Agriculture, 87: 2801–2809.

[40] Pious Soris Tresina. & Veerabahu Ramasamy Mohan. (2012). Comparative assement on the nutritional and antinutritional attributes of the underutilized legumes, Canavalia gladia (JACQ.) DC, Erythrina indica LAM and *Abrus precatorius* L. Tropical Subtropical Agroecosystem, 15: 539–556.

[41] Bhuiyan M. A. H. (2004). Evaluation of introducing mungbean into cereal based cropping Pattern for sustainable soil fertility and productivity. Ph.D. Thesis. Department of Soil Science Bangladesh Agricultural University, Mymensingh, Bangladesh 1-217p.

[42] Baud S., Boutin J. P., Miquel M., Lepiniec L. & Rochat C. (2002). An integrated over view of seed development in Arabidopsis thaliana ecotype WS Plant Physiology. Biochemistry; 40: 151–160.

[43] Kamalak A., Canbolat O., Gurbuz Y., Erol A. & Ozay O. (2005). Effect of maturity on the chemical composition, in vitro and in situ dry matter degradation of tumbleweed hay (Gundelia tournefortii L.) Small Ruminant Reserch, 58(2): 149-156.

[44] Bravo L., Siddhuraju P. & Sauvo-Calixto F. (1999). Composition of under exploited Indian pulses. Comparison with common legumes. Food Chemistry, 64: 185-102.

[45] Kalidass C. & Mahapatra A. K. (2014). Evaluation of the proximate and phytochemical compositions of an underexploited legume Mucuna pruriens var. utilis (Wall ex Wight) L.H. Bailey. International Food Research Journal, 21: 303-308.

[46] De la Vega A. & Sotelo A. (1986). The nutritional quality and toxin content of wild and cultivated lima beans (*Phaseolus lunatus*) Qual. Plant-Plant Foods Hum. Nutrition, 36: 75-83.

[47] Rao N., Deosthale B.S., Pant Y.G. & K.C. (1989). Nutritive Value of Indian Foods. Hyderabad, India: National Institute of Nutrition, Indian Council of Medical Research.

[48] Rajaram N. & Janardhanan K. (1993). Biochemical composition of Lima bean (*Phaseolus lunatus* L.) seeds. Ibid, 21: 39-43.

[49] Vijayakumari K., Siddhuraju P. & Janardhanan K. (1993b). Nutritional and antinutritional properties of certain underexploited legume seeds. International Journal Food Science Nutrition, 44: 181-189.

[50] Kalidass C. & Mohan V.R. (2012b). Nutritional composition and anti-nutritional of factors of little-known species Vigna, Tropical and Subtropical Agroecosystems, 15: 525–538.

[51] Miller-Cebert R.L., Sistani N.A. & Cebert E. (2009a). Comparative mineral composition among canola cultivars and other cruciferous leafy greens. Journal Food Comp Anal, 22: 112-116.

[52] Ayaz A.F., Glew R.H., Millson M., Huang H.S., Chuang L.T., Sanz C. & Hayirlioglu Ayaz S. (2006). Nutrient contents of kale (Brassica oleraceae L. var. acephala DC.) Food Chemistry, 96: 572–579.

[53] Russel E.W. (1973). Soil conditions and plant growth. Supergene Zone, M. 19p.

[54] Tirasoglu E., Cevik U., Ertugrul B., Apaydin G., Baltas H. & Ertugrul M. (2005). Determination of trace elements in cole (Brassica oleraceae var. acephale) at Trabzon region in Turkey. J. Quantitative, Spectroscopy, Radiative Transfer, 94: 181-187.

[55] Nunez-gonzalez M. A. & al. (2001). Genotypic variability in absorption of minerals among bean (*Phaseolus vulgaris* L.) cultivars exposed to low nutrient stress. Crop Research, 22 (3): 408-423.

[56] Sadiq M. & Hussain G. (1994). Effect of chelate fertilizers on dry matter and metallic composition of bean plants in a pot experiment. Journal Plant Nutrition, 17: 1477-1488.

[57] Carbonell-Barrachina A. A., Burlo F. & Mataix J. (1998). Response of bean micronutrient nutrition to arsenic and salinity. Journal of Plant Nutrition, 21 (6): 287-299.

[58] Nieman D.C., Butterworth. & Nieman C.N. (1992). Nutrition, WmC. Brown publishers. Dubugue, USA, 237-312pp.

[59] Siddhuraju P., Becker K. & Makkar H.S. (2001). Chemical composition, protein fractionation, essential amino acid potential and antimetabolic constituents of an unconventional legume, Gila bean (Entada phaseoloides Merrill.) seed kernel. Journal Science of Food Agriculture, 82: 192–202.

[60] Geissler C.A. & Powers H.J. (2005). Human Nutrition. Elsevier, Churchull, Livingston.

[61] Toma R.B. & Tabeckia M.M. (1979). HPLC analysis of B-vitamins in rice and rice products. Journal Food Science, 44:263-266.

[62] FAO/WHO. (2002). Human vitamin and mineral requirements. Report of a joint FAO/WHO expert consultation, Bangkok, Thailand. World Health Organization Food and Agriculture Organization of the United Nations Rome (Accessed 2010.07.04).

[63] Luthria D. L. & Pastor-Corrales M. A. (2005). Phenolic acid content of fifteen dry edible beans (Phaseolus vulgaris L.) varieties. Journal of Food Composition and Analysis, 19: 205- 211.

[64] Da Silva E.J.A., Oliveira A.B., Lapa A.J. (1994). Pharmacological evaluation of the antiinflammatory activity of a citrus bioflavonoid,

hesperidin, and the isoflavonoids, duartin and claussequinone, in rats and mice. Journal Pharm Pharmacological, 46(2): 118-220.

[65] Read, M. A., (1995). Flavonoids: naturally occurring anti-inflammatory agents Vascular. Am. Journal. Pathological, 147(2) : 235-7.

[66] Oomah B. D., Cardador-Martinez A. & Loarca-Pina G. (2005). Phenolics and antioxidative activities in common beans (Phaseolus vulgaris L.). Journal of the Science of Food and Agriculture, 85: 935-942.

[67] FAO/WHO. (1992). Codex standard for Edible cassava flour-African Regional standard. Rome: FAO/WHO Food Standards Programme.

[68] Montgomery R.D. (1980). Cyanogens. In: Toxic constitutes of Plant Food Stuffs, 2nd edn, ed. IE Liener, New York: Academic Press 158 – 160pp.

[69] Loewus F. (2002). Biosynthesis of Phytate in Food Grains and Seeds. In: Food Phytates (Eds.) Reddy N. R. and S. K. Sathe, CRC Press. Florida, USA: pp.53–61.

[70] Raboy V. (1990). The biochemistry and genetics of phytic acid synthesis in higher plants. In: Morre, E.J.; Boss, W.S. and Loewus, F.A. Inositol metabolism in plants. New York: John Wiley and Sons, 1: 55-76.

[71] Lott J. N. A., Randal P. J., Goodchild D. J. & Craig S. (1985). Occurrence of globoid crystals in cotyledonary bodies of Pisum sativum as influenced changes in experimentally induced changes in Mg, Ca and K contents of seeds. Australian Journal Plant Physiology, 12: 341-353.

[72] Wanasundera J.P.D. & Ravindran G. (1994). Nutritional assessment of yam (Dioscorea alata) tubers. Plant Foods for Human Nutrition, 46: 33-39.

[73] Alves R. E., Bezerra F. C., Abreu F. A. P. & Filgueirras H. A. C. (1999). Development and maturation of apple of early dwarf cashew tree CCP-76. Acta Horticulturae, 485: 225 – 230.

[74] Oulaï, S. F.; Koné, F. M. T.; Amedée, A. P.; Gonnety, J. T.; Faulet, B. M.; Kouamé, L. P., 2014. Impact of cooking times on some nutritional and anti-nutritional factors of Ivorian breadfruit (*Artocarpus altilis*) flour. Int. J. Rec. Biotech., 2 (3): 34-46.

[75] Davis A. M. (1981). The oxalate, tannin, crude fiber, and crude protein composition of young plants of some Atriplex species.J. Range Manage, 34:329-331.

[76] Munro A. & Bassir D. (1969). Oxalate in Nigerian vegetables. West African journal of biological and applied chemistry, 12: 14-18.

[77] Narjes Zamani., Manijeh Mianabadi*. & Ahmad Abdolzadeh. (2011). Changes in anti-oxidant activity of Thymus transcaspicus (Klokov) during growth and developmental stages Journal of Cell and Molecular Research, 3 (1):12-18.

[78] Reynoso C., Ramos G. & Loarca P. (2006). Bioactive components in common beans (Phaseolus vulgaris L.). Research Signpost, 2: 37- 61.

[79] Viera R.F., Grayer R.J. & Paton A.J. (2003). Chemical profiling of Ocimum americanum using external flavonoids. Phytochemistry, 63: 555-67.

[80] Horbowicz M., Kosson R., Grzesiuk A. & Dębski H. (2008). Anthocyanins in fruit and vegetables their occurrence, analysis and role in human nutrition. Vegetable Crops Res Bull, 68: 5–22.

[81] Liu R.H. (2004). Potential synergy of phytochemicals in cancer prevention : mechanism of action. Journal Nutrition 134: 34795-34855.

[82] Limasset B., Le Doucen C., Dore J.-C., Ojasoo T., Damon M. & Crastes de Paulet A. (1993). Effects of flavonoids on the release of reactive oxygen species by stimulated human neutrophils. Multivariate analysis of structure-activity relationships (SAR). Biochemical Pharmacological, 46: 1257-1271.

[83] Sandberg A.S. (2002). Bioavailability of minerals in legumes. Bristish Journal of Nutrition, 88:281-285.

[84] Coelho J.V. & Lajolo F.M. (1993). Total phenolic compounds and tannins in seeds of Phaseolus vulgaris during development. Arch Latinoam Nutrition, 43: 61–65.