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Comparison of Phytochemicals and Anti-Nutritional Factors in Some Selected Wild and Edible Bean in Nigeria

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Abstract

This work aims at analyzing the bioactive and anti-nutritional compounds of edible and wild beans when unprocessed and malted. Qualitative screening of phytochemicals in the various bean samples was determined in ethanol and petroleum ether solvents. Results of the anti-nutritional compositions of unprocessed wild bean extracted with petroleum ether showed there were no traces of saponin and polyphenol, in *Feregede* and also in edible bean-IT07K-243-1-10 which also had no traces of saponin and tannin. After malting, saponin was totally absent in *Pakala*, *Mucuna*, IT97k-499-35, IT07k-243-1-10, and IT04k-333-2 respectively. Polyphenol was also found to be absent in IT07k-243-1-10. *Mucuna* has the highest phytic acid level (7.8867 ± 0.011) while *Feregede* has the lowest phytic acid level (2.9810 ± 0.004). *Otili* has the highest anti-trypsin level (12.001 ± 0.0013). This study showed varying levels of anti-nutrients on the respective bean samples when unprocessed but decreased marginally after malting. It was keenly noted that values derived, either before and after malting were not significantly different (P ≤ 0.05) from each other. In all, this study had further shown that malting process enhanced removal of anti-nutrients which invariably would lead to availability of nutrient for animal and human consumption.

Keywords

Beans, Saponin, Tannin, Trypsin, Polyphenol

1. Introduction

Legumes (family: *Fabaceae*) have been recognized to be the second most valuable plant source for human and

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animal nutrition [1]. Legumes are designated to be the third largest family among flowering plants, consisting of approximately 650 genera and 20,000 species [2]. A significant part of human population relies on legumes as staple food for subsistence, particularly in combination with cereals. Even though several common proteinaceous edible legumes (soyabean, cowpea, and others) are available on the market, in most instances, production rate compared with consumption (as food and feed) has remained unmet, and an ever-increasing demand has been witnessed [3]. They are unique foods because of their rich nutrient content including starch, protein dietary fiber, oligosaccharides, phyto-chemicals and minerals. Their nutritional contents contribute to many health benefits to the human beings [4] [5].

Preparation of legume-based foods has remained, to some extent an art, and their nutritional quality has been of interest to both professionals and laymen. The malting process aids in improvement of the organoleptic quality of edible legumes and enhances nutritional quality. Finley *et al.* and Mitchel *et al.* [6] [7] reviewed the use of malting to improve the nutritional value of legumes, with particular reference to the generation of functional foods and functional ingredients. Malting is presented as alternatives that are able to reduce or inactivate anti-nutritional factors in legumes, to preserve and possibly enhance the content of isoflavones in legumes, and improves the potential of legumes as functional foods and as ingredients for use in functional foods.

Some of the common anti-nutrients interfere with the use of wild legumes include antivitamins, allergens, cyanogenic glycosides, flatulence factors, goitrogens, hydrogen cyanide, lectins, lysinoalanine, oligosaccharides, phytate, protease inhibitors, saponins, trypsin inhibitors, and several high-molecular-weight phenolic compounds [8]-[12]. However, the concentration and level of these anti-nutrients may vary between legumes and also among subaccessions of the same legume source depending on the location of collection, stage of development, and availability. The greatest impediment to utilize wild or underutilized legumes as a food and feed is the presence of certain anti-nutritional factors, which may not only be toxic, but also can be lethal in extreme situations. For a food researcher, removal of the anti-nutrients from the wild and underutilized legumes with minimal compromise on the nutritional qualities has been a great challenge [13].

Considering the above, it has become imperative for governments along with food researchers and nutritionists to search for cheap, reliable, and safe plant-based resources to accomplish the demand for protein-rich foods. For alleviating hunger and overcoming malnutrition, particularly in children and pregnant women, there is an increased demand in developing countries to explore underutilized legumes [14]-[16]. Exploring wild underutilized legumes can be of high significance for food security, meeting nutritional requirements, and agricultural development. Many of the known wild and underutilized legumes possess adequate amounts of protein, essential amino acids, polyunsaturated fatty acids (PUFAs), dietary fiber, and essential minerals and vitamins comparable to other common legumes, along with the presence of beneficial bioactive compounds. Hence, the present study is geared towards gathering useful information on our localized natural wild and underutilized legumes, which have remained underutilized and unexplored.

2. Materials and Methods

2.1. Collection of Cultivars

Dry, wild bean—*Sphenostyles stenocarpa* (*Otili*), *Cajanus cajan* (*Feregede*), *Phaseolus lunatus* (*Pakala*), were sourced from the bush of Ado Ekiti environ while the wild type-*Mucuna cochindunum* (*Mucuna*), and edible type bean-*Phaseolus vulgaris* tagged, IT845-2246-4, IT97K-499-35 and IT04K-333-2 were brought from International Institute of Tropical Agriculture (IITA), Ibadan Nigeria. Authentication was carried out at the herbarium of the Plant Science Department, Ekiti State University Ado Ekiti, Nigeria.

2.2. Physical Characteristic Test

On all the ten samples collected, bean characteristics which include colour, seed classification and number of seed per grams were determined by modifying the method of Rooney and Miller [17]. From each sample, one hundred whole seeds were randomly selected and placed in a plastic container. The weight of the hundred seeds was determined with a top loading weighing balance. Three replicates were averaged and recorded. The germinative properties were determined according to the method of Institute of Brewing [18]. This was done by taking a graduated meter rule and manual counting of percentage growth of each of the beans during germination (**Table 1**).

Table 1. Table of colour, seed classification by shape, weight by 100 beans, shoot length and percentage growth per seed after 5th day germination.

Sample	Colour	Classification of Seed	Weight of 100 Beans	Shoot Length	Percentage Growth PER 10 Seeds
Otili	Grey	Oval shape (Bigger)	26.10 ± 0.4	2.80 ± 0.3	80
Feregede	Grey	Oval Shape (Smaller)	10.65 ± 0.1	3.47 ± 0.1	100
Pakala	Variegated	Kidney	38.70 ± 0.1	4.37 ± 0.1	100
Mucuna	Black	Oval	78.11 ± 0.6	3.50 ± 0.3	70
IT99K-573-2-1	Off white	Oval	19.80 ± 0.3	6.37 ± 0.1	100
IT99K-573-1-1	Off white	Oval	17.90 ± 0.4	5.75 ± 0.3	100
IT97K-499-35	Off white	Oval	18.70 ± 0.1	7.43 ± 0.1	90
IT07K-243-1-10	Off white	Oval	20.10 ± 0.1	3.36 ± 0.1	80
IT04K-333-2	Off white	Oval	19.30 ± 0.3	5.50 ± 0.1	70
IT845-2246-4	Brown	Oval	16.30 ± 0.3	7.20 ± 0.1	90

2.3. Malting

Slight modification as described by Awoyinka and Adebawo [19] was adopted. The ten samples were steeped to give a grain/water ratio 3:4 for 65 h in a cycle comprising 6 h wet and 3 h dry. After the steeping regime the cultivars were spread in germination boxes in the dark and allowed to grow for five days in an atmosphere of near water saturation at room temperature to encourage sprouting. At the end of the germination period samples were dried in oven at 45˚C for 24 h.

2.4. Phytochemical Screening

Phytochemical screening for major constituents was undertaken using standard qualitative methods as described by Sofowora method [20] using petroleum ether and ethanol as solvent respectively. The test for alkaloids was carried out by subjecting 0.5 g aqueous extract in 5 ml 1% HCl, boiled, filtered and Mayer's reagent added [21] [22]. For cardiac glycosides, Killer Kiliani test [22] was adopted (0.5 g of extract was added to 2 ml acetic anhydrate plus H_2SO_4 [20]. The presence of flavonoids was determined using 1% aluminum chloride solution in methanol concentrated HCl, magnesium turnins, and potassium hydroxide solution [23] [24]. For Sterols 1 ml of extract was treated with drops of chloroform, acetic anhydride and conc. H_2SO_4 and observed for the formation of dark pink or red colour. Terpernoids was detected by Salkowki's test, 1 ml of chloroform was added to 2 ml of each extract followed by a few drops of concentrated sulphuric acid. A reddish brown precipitate produce immediately indicate the presence of Terpenoids. Well for presence of Quinones, a small portion of extract with conc. HCl and observed for the formation of yellow precipitate**.** However, for Oxalate 3 ml portion of extract were added a few drops of ethanoic acid glacial. A greenish black colouration indicates the presence of Oxalate [20].

3. Anti-Nutritional Analysis

3.1. Determination of Tannins

This was determined by Folin Denis colometric method as modified by Nwosu [25]. Five grams (5 g) of the sample was put inside a volumetric flask and 50 ml of distilled water was dispensed inside the volumetric flask. The mixture was shaken for 30 minutes at room temperature and filtered to obtain the extract. A standard tannic acid solution was prepared, 2 ml of the standard solution and equal volume of distilled water were dispersed into a separate 50 ml volumetric flasks to serve as a standard and reagent blank respectively. Then 2 ml of each of the sample extracts was put in their respective labeled flask. The content of each flask was mixed with 35 ml distilled water and 1 ml of the Folin Denis reagent was added to each. This was followed by 2.5 ml of saturated Na_2CO_3 solution. Therefore, each flask was diluted to the 50 ml mark with distilled water and incubated for 90 minutes at room temperature. Their absorbance was measured at 760 nm in a spectrophotometer with the reagent

blank at zero.

The tannin content was calculated as shown below:

$$\%\ \text{Tannin} = \frac{100}{W} \times \frac{au}{as} \times C \times \frac{Vt}{Va}$$

where, W = weight of sample

au = absorbance of test sample

as = absorbance of standard tanning solution

C = Concentration of standard tannin Solution

Vt = Total volume of extract

Va = Volume of extract analysed

3.2. Determination of Saponins

This was done by the double solvent extraction gravimetric method [21]. Five grams (5 g) of the sample was mixed with 50 ml of 20% aqueous ethanol solution and incubated for 12 hours at a temperature of 55˚C with constant agitation. After that, the mixture was filtered through Whatman filter paper. The residue was re-extracted with 50 ml of the ethanol solution for 30 minutes and the extracts weighed together.

The combined extract was reduced to about 40 ml by evaporation and then transferred to a separating funnel and equal volume (40 ml) of diethyl ether was added to it. After mixing well, there was a partition and the other layer was discarded while the aqueous layer was reserved. This aqueous layer was re-extracted with the ether after which its pH was reduced to 4.5 with dropwise addition of dilute NaOH solution. Saponin in the extract was taken up in successive extraction with 60 ml and 30 ml portion of normal butanol. The combine extract (ppt) was washed with 5% NaCl solution and evaporated to dryness in a previously pre-weighed evaporating dish. The saponin was then dried in the oven at 60˚C (to remove any residual solvent) and re-weighed. The saponin was determined and calculated as a percentage of the original samples.

$$\%\ \text{saponin} = \frac{W_2 - W_1}{W} \times 100$$

where W = weight of sample used

W_1 = weight of empty evaporation dish

W_2 = weight of dish + saponin extract

3.3. Determination of Trypsin Inhibitor

This was done using the spectrophotometric method, described by Babar *et al*., [26]. Five grams (5 g) of the test sample was dispersed in 50 ml of 0.5 m NaCl solution and stirred for 30 minutes at room temperature. It was centrifuged and the supernatant filtered through Whatman filter paper. The filtrate was used for the assay. Standard trypsin was prepared and used to treat the substrate solution (N-benzol-D1-arginine-p-anilide; BAPA). The extent of inhibition was used as a standard for measuring the trypsin. In the tube containing 2 ml of extract, 10 ml of the substrate (BAPA) was added. Also the 2nd part of the standard trypsin solution was added in another test tube containing only 10 ml of the substrate. The latter served as the blank.

The content of the tubes were allowed to stand for 30 minutes and then absorbance of the solution were measured spectrophotometrically at 410 nm wavelength. One trypsin activity unit inhibited is given by an increase on 0.01 absorbance unit at 410 nm.

$$\text{Trypsin unit inhibited}/100\ \text{g} = \frac{Au}{As} \times 0.01 \times F$$

where, Au = Absorbance of standard (uninhibited sample)

$$F = \text{Experimental factor given as } \frac{Vf}{Va} \times \frac{1}{W}$$

where,

Vf = Total volume of extract
Va = Volume of extract analysed

3.4. Phytate Determination

This was carried out according to A.O.A.C. [27]. 2 g of the sample was weighed into a test tube. About 10 ml of distilled water was added. The sample was extracted using 2 ml of 0.2 M HCl (aq). About 0.5 ml of the extract was pipetted into a test tube fitted with glass stopper. Then, 1 ml of the solution was added in the tube and covered with stopper. The tube was heated in a boiling water bath for 30 min and the tube was covered very well with the stopper for the first 15 min. Then the test tube containing the solution was cooled in ice water for 15 min and allowed to adjust to room temperature. Then the content of the test tube was mixed very well and centrifuged for 30 min. About 1 ml of the supernatant was transferred into another test tube and about 1.5 ml of the solution was added.

The absorbance at 420 nm against distilled water was measured.

$$\%\ \text{Phytate} = \frac{Au}{As} \times \frac{C}{W} \times \frac{100}{VA} \times Vf$$

Au = absorbance of test sample
As = Absorbance of standard solution
C = concentration of standard solution
W = Weight of sample used
Vf = Total volume of extract
Va = Volume of extract

3.5. Estimation of Polyphenols

Polyphenol substances were estimated by Folin-Denis method as modified by Nwosu [25]. About 200 mg defatted material was taken in a 250 ml round bottomed flask and 100 ml of 1% HCl in methanol was added. The contents were refluxed for 2 h, cooled, filtered and the volume was made up to 100 ml with acid-methanol after few washings 0.2 ml of extract was taken and 7.5 ml of water and 0.5 ml Folin-Denis reagent were added and mixed. To this, 1 ml of saturated sodium carbonate solution was added and volume made up to 10 ml with water, mixed and the absorbance was measured at 760 nm after 30 min. The results were calculated as mg tannic acid equivalents/g sample and expressed as mg/100 g dry weight.

4. Results and Discussion

The seeds of beans (*Otili*, *Feregede*, *Pakala*, and *Mucuna*) analyzed in this present study are rich in phytochemicals (**Table 2-5**). As shown in the results, presence or absence of certain bioactive compounds depends on the solvent of extraction used (petroleum ether or ethanol) because of their polar and non-polar nature or the bean form (dry or malted). For instance Oxalate was found absent only in *Pakala*, and some common beans such as (IT97K-499-35, IT04K-333-2 and IT845-2246-4) when phytochemical screening of ethanol extracts of malted samples was carried out but absent in all bean sample when phytochemical screening of petroleum ether extracts of malted dry samples was performed. The results of the phytochemical screening of petroleum ether and ethanol extracts of malted and dry samples contained phytochemicals such as alkaloids, flavonoids, saponin, terpenoids, quinones, sterols, tannins, phenols (**Table 2-5**). All these phytochemicals present in these legumes compared favorably with those reported from some medicinal plants found in Nigeria [20]. Alkaloids, comprising a large group of nitrogenous compounds are widely used as cancer chemotherapeutic agents [28]. In the report of John *et al.* [29] alkaloids also interfere with cell division hence the presence of alkaloids in the plant makes it a possible remedy in the treatment of cancer. They further reported that pure flavonoid compounds such as anthocyanins, quercetin glycosides and tannins, present in the seed displayed antioxidant and anticarcinogenic activity. Presence of tannins suggests the ability to play a major role as antidiarrhoec and antihaemorrhagic agent [30]. The presence of Saponins, another common class of anti-nutrient compounds has been reported to show hypocholesterolemic as well as anti-carcinogenic effects [31]. It is also of immense significance as antihypercholesterol, hypotensive and cardiac depressant properties [22] [32].

Table 2. Phytochemical screening of ethanol extracts of dry samples.

Samples	Alkaloid	Cardiac glycosides	Flavonoid	Phenol	Saponin	Sterol	Tanin	Terpenoid	Quinones	Oxalate
Otili	P	P	P	P	P	P	p	P	P	A
Feregede	P	P	A	A	A	A	P	P	P	A
Pakala	P	P	P	P	P	P	P	P	P	A
Mucuna	P	P	P	P	P	A	P	P	P	A
IT99K-573-2-1	P	P	P	P	P	P	P	P	P	A
IT99K-573-1-1	P	P	P	P	P	P	P	P	P	A
IT97K-499-35	P	P	P	P	P	P	P	P	P	A
IT07K-243-1-10	P	P	A	A	P	A	A	P	P	A
IT04K-333-2	P	P	P	P	P	P	P	P	P	A
IT845-2246-4	P	P	A	P	P	P	P	P	P	A

Note: P = Present; A = Absent.

Table 3. Phytochemical screening of ethanol extractsof malted dry samples.

Malted Samples	Alkaloid	Cardiac glycosides	Flavonoid	Phenol	Saponin	Sterol	Tanin	Terpenoid	Quinones	Oxalate
Otili	P	P	P	P	P	P	P	P	P	A
Feregede	P	P	A	P	P	A	P	P	P	A
Pakala	P	P	P	P	P	A	P	P	P	A
Mucuna	P	P	P	P	P	A	P	P	P	A
IT99K-573-2-1	P	P	P	P	P	P	P	P	P	A
IT99K-573-1-1	P	P	P	P	P	P	P	P	P	A
IT97K-499-35	P	P	P	P	P	P	P	P	P	A
IT07K-243-1-10	P	P	A	A	P	P	A	P	P	A
IT04K-333-2	P	P	P	P	P	P	P	P	P	A
IT845-2246-4	P	P	A	P	P	P	P	P	P	A

Note: P = Present; A = Absent.

Table 4. Phytochemical screening of petroleum ether extracts of dry samples.

Samples	Alkaloid	Cardiac glycosides	Flavonoid	Phenol	Saponin	Sterol	Tanin	Terpenoid	Quinones	Oxalate
Otili	P	P	P	P	P	P	p	p	P	P
Feregede	P	P	P	A	A	P	P	A	P	P
Pakala	P	P	A	P	P	P	P	A	P	A
Mucuna	P	P	P	P	P	P	P	P	P	P
IT99K-573-2-1	P	P	A	P	P	P	A	P	A	P
IT99K-573-1-1	P	P	P	P	P	P	P	P	P	P
IT97K-499-35	P	P	P	P	P	P	P	P	P	A
IT07K-243-1-10	P	P	P	A	A	P	P	A	P	P
IT04K-333-2	P	P	P	A	A	P	P	P	P	A
IT845-2246-4	P	P	A	A	P	A	P	A	P	A

Note: P = Present; A = Absent.

Table 5. Phytochemical screening of petroleum ether extracts of malted samples.

Samples	Alkaloid	Cardiac glycosides	Flavonoid	Phenol	Saponin	Sterol	Tannin	Terpenoid	Quinones	Oxalate
Otili	P	P	P	P	P	P	P	A	P	A
Feregede	P	P	A	P	P	A	P	P	P	A
Pakala	P	P	P	P	A	A	P	P	P	A
Mucuna	P	P	P	P	A	A	P	P	P	P
IT99K-573-2-1	P	P	A	P	P	P	P	P	P	A
IT99K-573-1-1	P	P	A	P	P	P	P	P	P	A
IT97K-499-35	P	P	P	P	A	P	P	P	P	A
IT07K-243-1-10	P	P	A	A	A	P	A	P	P	A
IT04K-333-2	P	P	A	P	A	P	P	P	P	P
IT845-2246-4	P	P	A	P	P	P	P	P	P	A

Note: P = Present; A = Absent.

Moreso in the anti-nutritional analysis shown in **Table 6** malted ethanolic extract, IT99K-573-2-1 (edible bean) had the highest Saponin with the value 1.22 ± 0.01 and found not available in edible bean sample IT07K-243-1-10. Phytate content was found to be very high in *Mucuna* with the value 11.8 ± 0.02 while the lowest values are found in both *Feregede* and *Pakala* with the values 4.3 ± 0.1. Tannin was not found more in *Mucuna* with the value 1.28 ± 0.01 than any other bean sample. Also trypsin inhibitor was found to be significantly higher in *Otili* with the value of 19.92 ± 0.02 compares to other sample. Well polyphenol was not detected in *Feregede* and Edible bean IT97K-499-35 while *Pakala* had the highest value of 0.5 ± 0.02 while *Mucuna* had the lowest with value 0.33 ± 0.01.

From the result of anti-nutritional factors in malted samples with petroleum ether extract (**Table 7**), it was observed that saponin level was highest in edible bean IT845-2246-4 with value of 0.87 ± 0.01 sample B had the lowest value of 0.039 ± 0.001. Phytate level in *otili* was higher when compared to other bean sample with a value of 6.41 ± 0.01 while the lowest was *Feregede* with the value 2.98 ± 0.01. *Mucuna* had the highest Tannin with the value of 0.5 ± 0.01 while the lowest value was found in the edible bean IT97K-499-35 with the value of 0.08 ± 0.0008. Trypsin inhibitor was found to be highest in *Otili* with the value of 12 ± 0.001 while the lowest was IT97K-499-35 with the value of 3.002 ± 0.001. Polyphenol was found to be highest in *Pakala* with a value of 0.59 ± 0.001 while the lowest was IT97K-499-35 with a value of 0.348 ± 0.001. Tannin was not found more in *Mucuna* with the value 1.28 ± 0.01 than any other bean sample. Also trypsin inhibitor was found to be significantly higher in *Otili* with the value of 19.92 ± 0.02 compares to other sample. Well polyphenol was not detected in *Feregede* and Edible bean IT97K-499-35 while *Pakala* had the highest value of 0.5 ± 0.02 while edible bean IT99K-573-2-1 had the lowest with value 0.33 ± 0.01. Important biological activities have been described for fibers, phenolic compounds, lectins, trypsin inhibitors, and phytic acid from beans like enhancement of the bifidogenic effect [33] [34]; antioxidant [35]; anticarcinogenic effects [36]. Nevertheless, it should be noted that some of the commonly considered anti-nutrient compounds like phenols and tannins are now being considered as potential antioxidants with health promoting effects [37]-[40]. Even phytic acid, widely considered to be a major anti-nutrient affecting the bioavailability of minerals and essential trace elements, has now been shown to possess rich antioxidant, anti-carcinogenic, and hypoglycemic activities [41] [42] Hence, depending on the consumer preferences retaining or eliminating these compounds could be facilitated.

5. Conclusion

These data provided preliminary evidence that consumption of beans diet such as the localized wild—*Otili*, *Feregede*, and *Pakala* competed favorably with the common edible bean *Phaseolus vulgaris* in bioactive compounds constituents. This present work provided information on their possible health implication when consumed by human in order to accomplish the demand for protein rich food.

Table 6. Anti-nutritional factors in dry pulverised samples with absolute ethanol extract.

Bean sample	Saponin	Phytate	Tannin	Trypsin	Polyphenol
Otili	0.583 ± 0.08	10.5 ± 0.1	0.43 ± 0.02	19.92 ± 0.02	0.3 ± 0.01
Feregede	ND	4.3 ± 0.1	0.25 ± 0.03	6.74 ± 0.3	ND
Pakala	0.413 ± 0.01	4.36 ± 0.1	0.98 ± 0.002	7.83 ± 0.3	0.5 ± 0.02
Mucuna	0.85 ± 0.001	11.8 ± 0.002	1.28 ± 0.01	5.28 ± 0.1	0.4 ± 0.02
IT99K-573-2-1	1.22 ± 0.01	6.84 ± 0.04	0.83 ± 0.01	8.39 ± 0.01	0.33 ± 0.01
IT99K-573-1-1	0.83 ± 0.02	7.76 ± 0.04	0.8 ± 0.01	6.75 ± 0.03	0.43 ± 0.02
IT97K-499-35	0.79 ± 0.02	7.97 ± 0.1	0.72 ± 0.01	6.13 ± 0.1	0.34 ± 0.01
IT07K-243-1-10	ND	7.60 ± 0.1	ND	7.14 ± 0.04	ND
IT04K-333-2	0.93 ± 0.02	6.5 ± 0.2	0.99 ± 0.05	5.3 ± 0.16	0.42 ± 0.01
IT845-2246-4	0.09 ± 0.06	9.13 ± 0.17	0.99 ± 0.06	6.78 ± 0.02	0.42 ± 0.01

NB: Samples in mg/g.

Table 7. Anti-nutritional factors in malted pulverised samples with petroleum ether extract.

Bean sample	Saponin	Phytate	Tannin	Trypsin	Polyphenol
Otili	0.40 ± 0.001	6.41 ± 0.01	0.298 ± 0.01	12.00 ± 0.001	0.375 ± 0.001
Feregede	0.39 ± 0.001	2.98 ± 0.01	0.090 ± 0.01	5.9 ± 0.005	0.4 ± 0.001
Pakala	ND	3.17 ± 0.01	0.380 ± 0.12	4.267 ± 0.001	0.59 ± 0.001
Mucuna	ND	7.86 ± 0.01	0.500 ± 0.10	4.68 ± 0.001	0.38 ± 0.009
IT99K-573-2-1	0.84 ± 0.01	4.84 ± 0.01	0.210 ± 0.002	4.10 ± 0.001	0.42 ± 0.001
IT99K-573-1-1	0.811 ± 0.01	5.18 ± 0.01	0.400 ± 0.001	3.3 ± 0.001	0.445 ± 0.001
IT97K-499-35	ND	5.66 ± 0.01	0.080 ± 0.001	3.002 ± 0.001	0.348 ± 0.001
IT07K-243-1-10	ND	5.8 ± 0.001	ND	5.018 ± 0.001	ND
IT04K-333-2	ND	4.81 ± 0.01	0.401 ± 0.005	5.4 ± 0.001	0.5 ± 0.001
IT845-2246-4	0.870 ± 0.01	6.00 ± 0.01	0.410 ± 0.001	4.86 ± 0.002	0.47 ± 0.001

Note: ND = Not determined.

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