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Research Application Summary

Assessment of physical purity of yellow common bean seeds in Tanzanian markets

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Abstract

Yellow bean trade is affected by limited information and unstructured system on production which has negative implications on seed supplies and other possible investments. The presence of improved varieties in seed markets shelves is a positive sign of a vibrant seed system. The aim of this study was to assess the physical purity of yellow bean varieties traded in the major markets in Tanzania using morphological traits basing on phenotypic traits of common beans. A total of 300 bean accessions were collected from Tanzanian markets and 14 inbreds reference samples were obtained from the International Centre for Tropical Agriculture (CIAT) gene bank in Uganda and the Tanzania Agricultural Research Institute (TARI). The accessions were grown to record their morphological traits to determine the level of phenotypic diversity of yellow bean varieties traded in the major markets in Tanzania. The samples were planted in augmented design and the data were collected from the growing plants at 50% of days to flowering, 50% of days of physiological maturity on number of pods per plant, number of seeds per pod, flower color and mature pod color. The analyses of variance (ANOVA), principal component analysis (PCA) and cluster analysis were conducted to characterize phenotypically the seed samples. The results of the ANOVA revealed significant ($P<0.05$) differences in six phenotypic traits out of eight traits that were studied. These were growth habit (GH), 50% of days to flowering (DF), flower color (FCOL), 50% of days to physiological maturity (DPM), mature pod color (MPDCOL) and seeds per pod (SDPD). The PCA distinguished the traits growth habit (DF), pods per plant (PDPL), 50% of days to physiological maturity (DPM), seeds per pod (SDPD) and seed weight (SW) as having greater variation. The study showed that there is phenotypic variation in yellow common beans in Tanzanian markets. Therefore, it was recommended that further investigation on the seed sold in the markets of Tanzania be done by periodically sending seed inspectors from the Seed Certifying Agency to these areas to collect seed samples from the markets for evaluation.

Key words: Breeder seed, market seed samples, physical purity, seed systems, Tanzania, yellow bean

Résumé

Le commerce du haricot jaune est affecté par des informations limitées et un système de production non structuré, ce qui a des implications négatives sur l'approvisionnement en semences et d'autres investissements possibles. La présence de variétés améliorées sur les étagères des marchés de semences est un signe positif d'un système semencier dynamique. Le but de cette étude était d'évaluer la pureté physique des variétés de haricots jaunes commercialisées sur les principaux marchés de Tanzanie en utilisant des traits morphologiques basés sur les traits phénotypiques des haricots communs. Au total, 300 accessions de haricots ont été collectées sur les marchés tanzaniens et 14 échantillons de référence consanguins ont été obtenus auprès de la banque de gènes du Centre international d'agriculture tropicale (CIAT) en Ouganda et de l'Institut tanzanien de recherche agricole (TARI). Les accessions ont été cultivées pour enregistrer leurs traits morphologiques afin de déterminer le niveau de diversité phénotypique des variétés de haricots jaunes commercialisées sur les principaux marchés de Tanzanie. Les échantillons ont été plantés en conception augmentée et les données ont été recueillies sur les plantes en croissance à 50 % des jours jusqu'à la floraison, 50 % des jours de maturité physiologique sur le nombre de gousses par plante, le nombre de graines par gousse, la couleur des fleurs et la couleur des gousses matures. . Les analyses de variance (ANOVA), l'analyse en composantes principales (PCA) et l'analyse par grappes ont été menées pour caractériser phénotypiquement les échantillons de graines. Les résultats de l'ANOVA ont révélé des différences significatives (P <0,05) dans six traits phénotypiques sur huit traits étudiés. Il s'agissait du port de croissance, de 50 % des jours jusqu'à la floraison, de la couleur des fleurs (FCOL), de 50 % des jours jusqu'à la maturité physiologique, de la couleur des gousses matures et des graines par gousse. L'ACP a distingué les caractères port de croissance, gousses par plante, 50 % de jours jusqu'à la maturité physiologique, graines par gousse et poids des graines comme ayant une plus grande variation. L'étude a montré qu'il existe une variation phénotypique des haricots communs jaunes sur les marchés tanzaniens. Par conséquent, il a été recommandé qu'une enquête plus approfondie sur les semences vendues sur les marchés de Tanzanie soit effectuée en envoyant périodiquement des inspecteurs de semences de l'Agence de certification des semences dans ces régions pour collecter des échantillons de semences sur les marchés à des fins d'évaluation.

Mots clés: Semences de sélectionneur, échantillons de semences commercialisées, pureté physique, systèmes semenciers, Tanzanie, haricot jaune

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Introduction

The common bean (*Phaseolus vulgaris* L.) is a diploid plant (2n=22) belonging to the family Leguminosae and genus Phaseolus (Pathania, 2014). The most accepted theory of the origin and evolution of common bean is that, the center of origin of common bean is located in South and Central America (Cortés, 2013). It was domesticated from Mesoamerican and Andean wild gene pools (Gepts, 1998). Major regions of common bean production are Latin America, Africa, the Middle East, China and Europe (FAO, 2001). Common bean was introduced in Eastern Africa by Portuguese traders in the 16th Century (Hillocks *et al*., 2006).

Seed is a key component of agricultural production and productivity, and the quality of the seed sown determines the upper limit of yield (Hillocks *et al*., 2006). However, the yields of common bean on smallholder farms are low, and the main reasons for these low yields include poor seed quality, pests and diseases, poor performance of the local landraces mainly due to their susceptibility to pests and diseases, soil infertility, drought and poor agronomic practices (Hillocks *et al*., 2006). In East Africa, the yellow common bean varieties are among the most preferred (49%), others being the sugar beans (21%), red mottled beans (19%), kablanketi (6%) and small whites (3%). Yellow beans are on high demand due to their quality attributes, such as early maturing, retaining about 80% of iron after boiling, and short cooking time (Lance *et al*., 2017). The main consumers of common bean as sauce are the homesteads, schools, military barracks and hospitals (Binagwa *et al*., 2019).

The reproductive biological species of common bean is from main common varieties which were developed from pure lines from Mesoamerican and Andean origins (Bitocchi *et al*., 2012) and varieties used for this research were developed from landraces such as Uyole, Njano, Gololi Wanja, Kablanketi and Masasu (Nassary *et al*., 2020). Therefore, the majority of farmers plant common bean seeds that are not genetically pure, and this is affecting the potential yields. The pure variety deteriorates along the cycles of production and during marketing. The extent of deterioration can be measured using morphological methods. The morphological methods are based on phenotypic characteristics such as shape, size, weight, color, length and width of the seed as well as the plant characteristics of germinated seeds and this study has used the phenotypic methods to confirm whether there was or not occurrence of morphologic variation. Therefore, the present research evaluated the phenotypic diversity of yellow common beans traded in Tanzanian markets.

Materials and Methods

Experimental site. The study was conducted at the Alliance of Biodiversity International and the International Centre for Tropical Agriculture based at the National Agricultural Research Laboratories (NARL)-Kawanda in Uganda. This research station is located within a bimodal rainfall region. It lies at 0 25'14.0" N of the Equator and 32 32'26.0" E of the Meridian, and at an altitude of 1200 meters above sea level (m.a.s.l). It has a tropical wet and mild dry climate and receives annual rainfall ranging between 800 and 1500 mm with slightly humid conditions (65% mean relative humidity). Average annual temperature is 22 ^{o}C and annual minimum and maximum temperatures are 16 and 28 ^{o}C, respectively. Soils are dark, reddish brown sandy loams with pH range of 5.5-6.2.

Seed sample collection and handling. A total of 300 seed samples of yellow common bean were collected randomly from the markets in five regions of Tanzania, namely Northern zone, Southern zone, Western zone, Lake zone and Coastal zone. These regions covered 30 districts which included Arusha, Momba, Kobondo, Mbulu, Babati, Siha, Moshi, Meru, Mbeya, Rungwe, Wanging'ombe, Iringa, Mufindi, Makambako, Temeke, Mbozi, Nkasi, Sumbawanga, Bukoba, Missenyi, Karagwe, Ngara, Geita, Muleba, Kakonko, Kibondo, Kasula, Kigoma, Kinondoni and Ilala. The seed samples were collected randomly from represented sacks from five markets in each district (Table 1).

Table 1. Sources, number and sizes of market seed samples that were used in the study

Region/Zone	Number of districts per region	Number of markets per district	Seed samples per market	Total
Northern	6	5	2	60
Southern	6	5	2	60
Wester	6	5	2	60
Lake	6	5	2	60
Coastal	6	5	2	60
Total	30			300

Reference seed set. A total of 14 reference seed samples were used in the study. They included ten samples collected by TARI and four samples collected from CIAT. Four samples from TARI were breeder seed of the officially released yellow bean varieties. The four reference samples from CIAT were materials maintained at the CIAT regional crop improvement hub in Uganda and they included Masindi yellow long and Masindi yellow short that are popularly marketed in Uganda, but believed to have crossed the border into the Tanzanian markets. Moore80082 is an officially released variety in Uganda (NAROBEAN3) and in Burundi, while Njano Gololi is believed to be a yellow bean landrace from Tanzania (Table 2).

Table 2. Reference seeds from TARI and CIAT Seed Bank-Uganda which were used in the study

	Sample name	Source	100 Seed weight	Seed Size	Seed Shape	Notes
1.	Selian 13	TARI	37.4	Medium	Round oval	Brown hilum
2.	Njano Uyole	TARI	30.5	Medium	Round flat	Dark hilum
3.	Uyole 16	TARI	41.2	Large	Elongate round	Brown hilum
4.	Uyole 98	TARI	39.4	Large	Elongate round	Brown hilum
5.	Vwawa Mbuzi	TARI	40.1	Large	Elongate round	Dark hilum
6.	Rusharura Bub	TARI	46.1	Large	Elongate flat	Dark hilum
7.	Uyole Njano Ndefu	TARI	42.3	Large	Kidney shaped	Dark hilum
8.	Njano Gololi Bub	TARI	38.1	Medium	Round oval	Dark hilum
9.	TZ Unlabeled 1	TARI	35.2	Medium	Round oval	Dark hilum
10.	TZ Unlabeled 2	TARI	45.2	Large	Elongate round	Dark hilum
11.	Masindi Yellow Long	CIAT	44.2	Large	Elongate round	Dark hilum
12.	Masindi Yellow Short	CIAT	29.0	Medium	Round oval	Brown hilum
13.	Njano Gololi	CIAT	35.4	Medium	Round oval	Brown hilum
14.	Moore 88002	CIAT	29.0	Medium	Round oval	Brown hilum

Experimental set up and management. For this study, the reference set were used as checks to identify the 300 seed market samples. The experiment was carried out in a screenhouse using augmented design with 14 checks and 300 market seed samples in three incomplete blocks, and 114 genotypes per block. The objective of this experimental design was to compare the morphological characteristics between reference seeds (inbreds) and market seed samples with minimal expenditure of resources such as time and money. The seeds were planted at 3-5 cm depth in a 10-litre dishpan and 10 seeds were planed in each dispan. Sandy-clay-loam soil (46:40:13) was used as the growth medium. Nitrogen, phosphorus and potassium (NPK) (20:20:20) fertilizer was applied at the rate of 0.1 g per10 kg of soil in three splits, namely at planting, flowering and at mid-pod filling stages. Weeds were uprooted by hand.

The trial received water from rains and was irrigated daily whenever necessary throughout the growth cycle of the crop, and on average, it was twice a day. The disease and pest control activities were scheduled according to vegetative development of the crop, although there were not serious incidences registered. Recommended standard agronomic crop management practices were followed.

Data collection and analysis. A total of 10 plants per pot, data were used for collection from each pot. Data were collected on phenotypic traits in growing plants and these included the growth habit, 50% of days to flowering, 50% of days to physiological maturity, flower color, mature pod color, 100 seed weight, number of seeds per pod and number of pods per plant (Table 3). Growth Habit (GH) was scored basing on visual classification by categories. Days to flowering (DF) were determined by counting the number of days from planting date up to flowering stage while days to physiological maturity (DPM) were determined by counting the number of days from planting date up to the day when the first pod started to discolor in 50% of the plants. The flower colour was observed on freshly opened flowers while mature pod colour (MPDCOL) was observed on the pods at physiological maturity. Seed weight (SW) was determined by counting manually and calculating the average weight of 1000 seeds from the weights of five 100-seed replicates (https://www.seedtest.org/upload/cms/user/OGM18-06dISTA_Rules_2019_10_weight_v2.pdf).The seed per pod (SDPD) was determined by calculating the average number of seeds per pod from five pods picked randomly from five plants. On the other hand, pod per plant was determined by calculating the average number of pods from five randomly selected plants.

Comparision of the reference seeds and market seeds was done by R-studio Software to determine the ANOVA for getting means and obtaining the F-probability (F-prob), and least significance of difference (LSD) values of DF, DPM, SW, SDPD and PDPL. The qualitative traits such as GH were scored 1 to 5, with one being determinate, two semi-determinate, three indeterminate and four climbing. Flower color (FCOL) was scored 1 to 5, with one being white, two violet, three purple and four yellow while MPDCOL was scored 1 to 5, with one being yellow, two red, three was white, and four purple. The average of frequencies of each habit and color were calculated ans subjected to ANOVA. The hierarchical cluster analysis was done using R-Studio and the dendrogram was generated using similarity matrices obtained from Euclidean distance and group link method, thus grouping the genotypes according to their similarities.

Table 3. Phenotypic traits and their description

Phenotypic trait	Property	Scale
Growth habit (GH)	Qualitative description of whether the bean plant is a climber, determinate or indeterminate	(1-5)
Days to flowering (anthesis) (DF)	Number of days from planting to the day when 50% of plants have at least one flower	(1-5)
Days to physiological maturity (DPM)	Number of days from planting to the day when first pod begins to discolor in 50% of the plants	(1-7)
Flower color (FLCOL)	Prominent color of flower	(1-5)
Mature pod color (MPDCOL)	Color of pod at physiological maturity	(1-5)
Seed weight (SW)	Weight in grams of 100 seeds	(gms)
Number of seeds per pod (SDPD)	Number of seeds counted per pod	unit by unit
Number of pods per plant (PDPL)	Number of pods counted per plant grown in a pot or the field	unit by unit

Results

Analysis of Phenotypic traits. The phenotypic analysis showed that there were significant differences at ($p<0.05$) in reference seeds and there were no significant differences in market seed samples for GH. There was significant different ($p<0.001$) in DF, FCOL, MPDCOL and SDPD. For market seed samples, there was no significant difference ($p<0.05$). However, there were significant differences at ($p<0.001$) for markets seed samples in FCOL and MPDCOL (Table 4).

Cluster analysis. The cluster analysis was represented by the dendrogram showing the clustering of the reference and market seed samples. The results showed that the reference seeds were clustered into six main groups (GI to GVI) (Figure 2). The first group (GI) comprised Masindi Yellow Long, Masindi Yellow Short and Rusharura Bub which showed high similarity. The second group (GII) was represented by only one sample, Njano Gololi Bub. The third group (GIII) was represented by Njano Gololi, TZ Unlabeled 2 and Njano Uyole. Moore88002 was in group four (GIV) alone, while Vwawa Market Mbuzi and Uyole16 were clustered together in group five (GV). Group six (GVI) compromised the highest number of samples, and these were Uyole 98, Uyole Njano Ndefu and TZ Unlabeled 1.

Table 4. Phenotypic traits of yellow common seeds from Tanzania

	Samples	GH	DF	FCOL	DPM	MPDCOL	SW	SDPD	PDPL
F-Prob	Reference seeds	0.031*	3.814***	9.144***	0.36184	2.200***	0.469	5.138***	1.000
	Markets seeds	1.000	0.772	2.200***	0.88487	2.200***	0.184	0.4669	0.151
	Interaction Reference and markets seeds	0.025*	2.925***	2.200***	0.02459*	2.200***	0.490	4.070***	1.000
	Means	1.09	32.67	1.97	57.44	4.40	43.64	6.62	3.59
	LSD $_{(0.05)}$	0.64	3.44	11.66	12.05	0.10	25.35	5.45	21.69

*=Significant at P 0.05, **=Significant at P 0.01, ***Significant at P 0.001

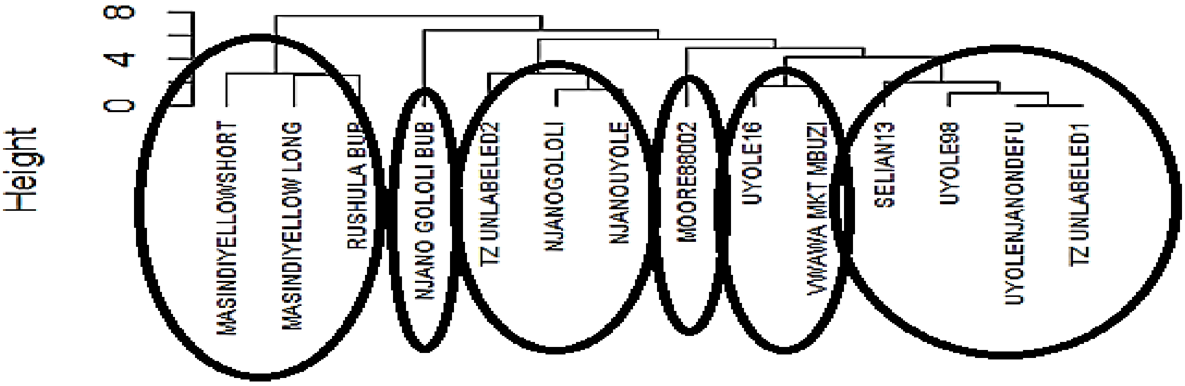

Figure 1. Dendrogram showing the morphological diversity of the reference seed samples of yellow common beans on unweighted neighbor joining clustering of Euclidean's similarity coefficients

For the case of market seeds, the dendrogram is clustered in two main groups A and B (Figure 2). Group A has two sub-groups and each one is further sub-divided into more subgroups. Group B has four sub-groups labelled 1, 2, 3 and 4, and each is further divided into more subgroups (Figure 2). The results showed that the varieties in the markets are phenotypically diverse.

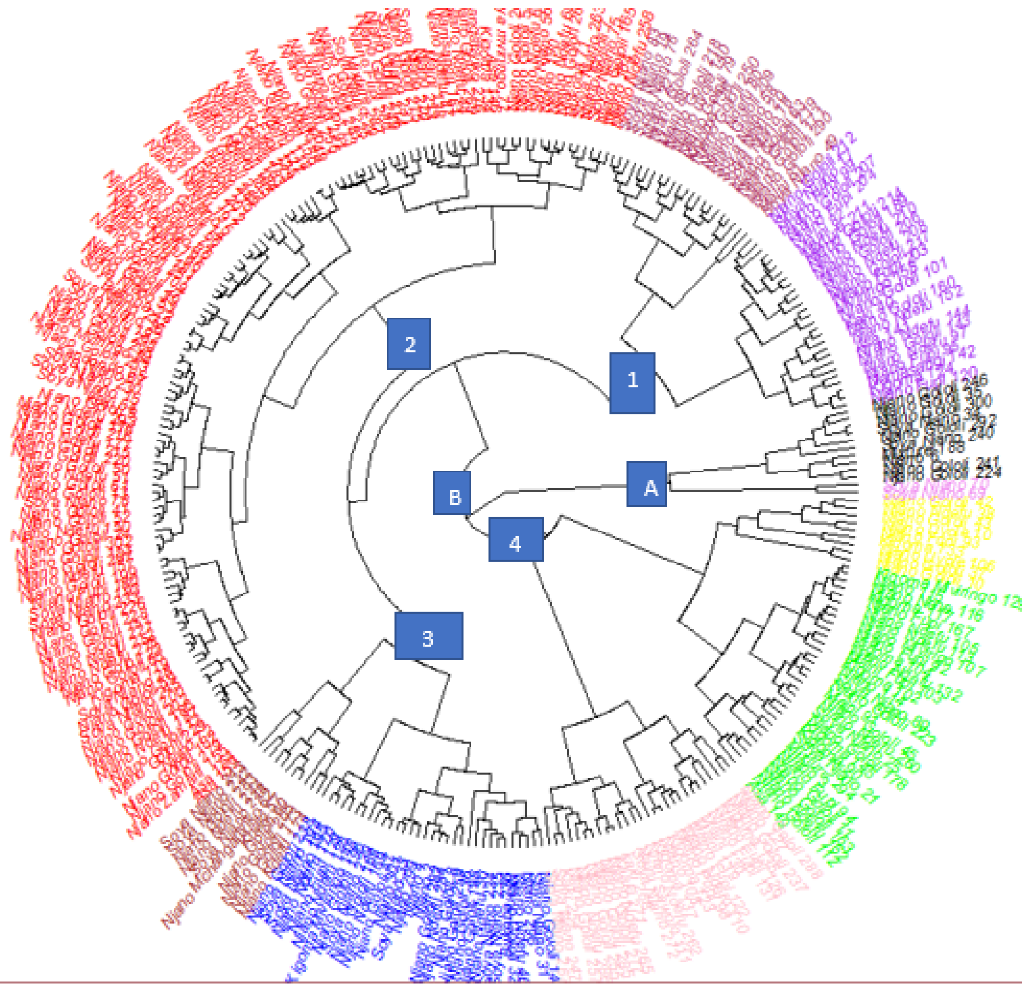

Figure 2. Dendrogram showing the morphological diversity of 300 market seeds of yellow common beans on unweighted neighbor joining clustering of Jaccard's similarity coefficients

Discussion

Analysis of variance. The results of this analysis showed that there was a level of phenotypic variation of reference seeds and markets. This variation could be attributed to morphological and environmental effects (Idris and Mohammed, 2012). These results are in line with those of Musango *et al*. (2021) who reported significant differences in days to flowering, seeds per pod and pods per plant for different phonotypic traits in common beans genotypes. Different researchers have reported significant amount of variability in different common beans. For example Bitocchi *et al*. (2012) who studied Mesoamerican origin of the common bean using sequence data. Current results suggest that the level of variation is higher in market samples, whereas reference seeds appeared as pure varieties.

Cluster analysis. The cluster analysis showed that there was phenotypic variation in the reference and market seed samples. For the reference seeds, the results were consistent with those obtained by Estelle *et al*. (2018). In this cluster analysis the Masindi yellow short, Njano Gololi Bub, TZ Unlabeled 2, Moore88002 and Selian 13 showed high phenotypic diversity and deserve attention for seed quality control and for use in breeding program.

The cluster results for market seed samples compared are in agreement with those of the study done by Fiore *et al*. (2020) who assessed the morphological and genetic variability of the Sicilian common bean germplasm. The cluster analysis revealed that the most predominant reference seeds (Njano Gololi Bub Market, Selian 13, Njano Uyole, Moore88002, TZ Unlabeled 1, TZ Unlabeled 2, Vwawa Mbuzi, Rusharura bub, Masindi yellow short and Masindi yellow long) were in the markets being commercialized in small portion and the big portion was for non-released varieties and unclear sources. According to the results, there is duplication of yellow common bean market seed samples in the collection, and this was also observed by Chiorato *et al*. (2007), Akhshi *et al*. (2014) and Rana *et al*. (2015). In fact, in the Republic of Tanzania, folk nomenclature of yellow common bean varies from one socio-linguistic group to another, and several folk varieties could be attributed to a single market seed sample and many market seed samples could have a similar name (Estelle *et al*., 2018).

The high diversity of common bean crop observed in this study may be due to farmers' customary seed exchanges. Blair *et al*. (2010) reported farmers' preference for many landraces, where diversified bean types are used for various agronomic and cultural reasons.

Conclusions

There was unstructured market system for yellow bean in Tanzania and informal seed sector was predomint. Overall, this study revealed that the yellow common beans in markets did not follow the right seed production procedures and therefore the intervention of the certifying agencies is recommended.

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