Tracking African beans' diversity:

a chemical and cytogenomic study of *Vigna unguiculata* (L.) Walp. and *Phaseolus vulgaris* L.

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In sub-Saharan Africa, pulses, and legumes in general, are essential food sources for alleviation of malnutrition and can play an important role in sustainable agriculture, due to their ability to fix atmospheric nitrogen. The common bean (*Phaseolus vulgaris* L.) and cowpea (*Vigna unguiculata* (L.) Walp.), are among the most important pulses in the dry areas of tropical Africa. Based on field surveys performed across three African countries (i.e., Angola – Western Africa; Mozambique – Eastern Africa and Cabo Verde Islands), we aim to compare the chemical composition and the genome size variation of the *Vigna unguiculata* and *Phaseolus vulgaris*, in order to evaluate the patterns of diversity between native and introduced species, respectively. According to our results, chemical composition of *Vigna unguiculata* has, higher content of B, Mg, S, and Zn, while *Phaseolus vulgaris* had more content of Fe, Ca, and Cu. Respecting the nuclear DNA content, on average, *Vigna unguiculata* presents significantly higher values than *Phaseolus vulgaris*, showing the first much more genome size variations. We conclude that both species are important resources in Angola, Mozambique and Cabo Verde Islands, but native *Vigna* species are often disregarded, even though they have generally good alimentary properties. To the best of our knowledge, this is the first comparative chemical and cytogenomic study, focusing on wild and crop species, collected across different African countries. It is highlighted that interdisciplinary approaches and new data are need for the sustainable use of African plant genetic resources, contributing to achieve some of the Sustainable Development Goals, as the reduction of hunger and increasing human health.

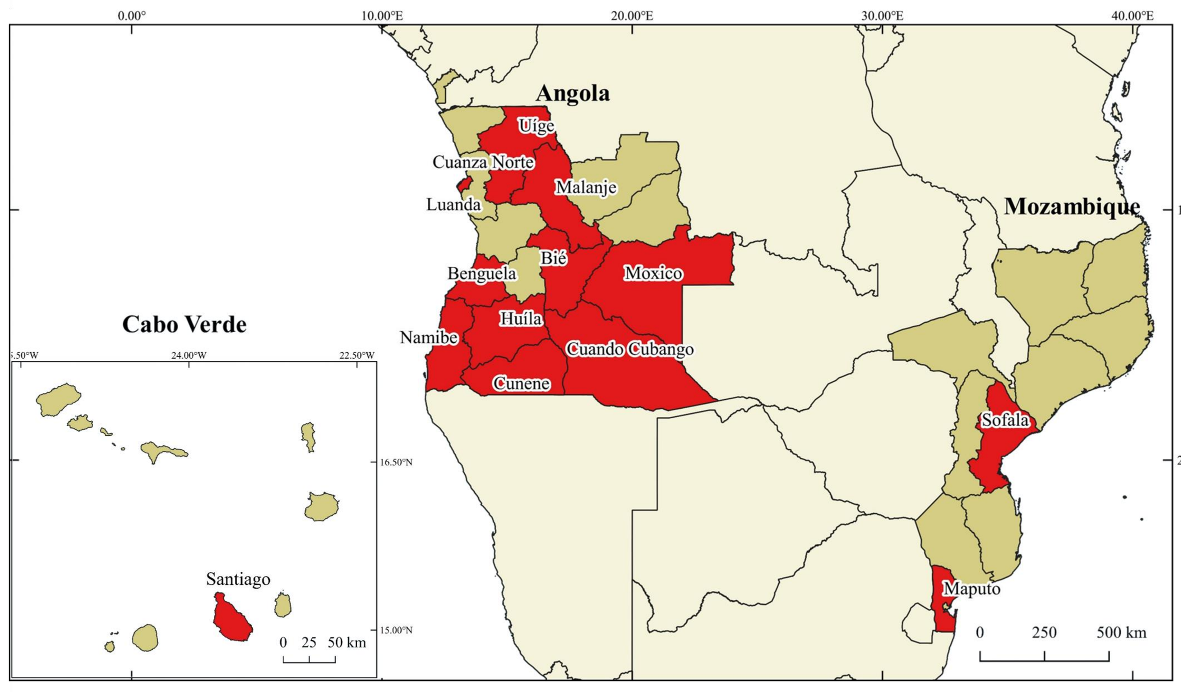
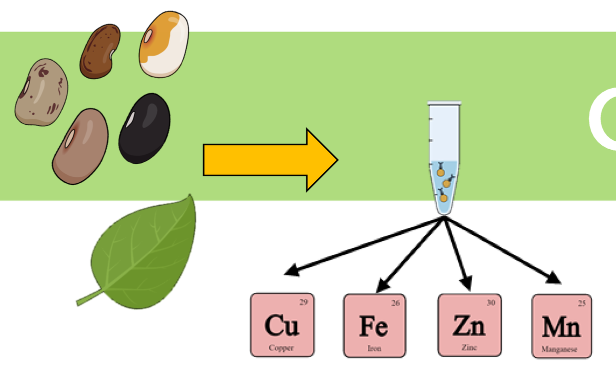


Figure 1. Countries and provinces where samples were collected (in red color): Cabo Verde (Santiago), Angola (Uíge, Cuanza Norte, Malanje, Benguela, Namibe, Bié, Moxico, Huíla, Cunene, and Cuando Cubango), and Mozambique (Sofala and Maputo).

CHEMICAL DIVERSITY



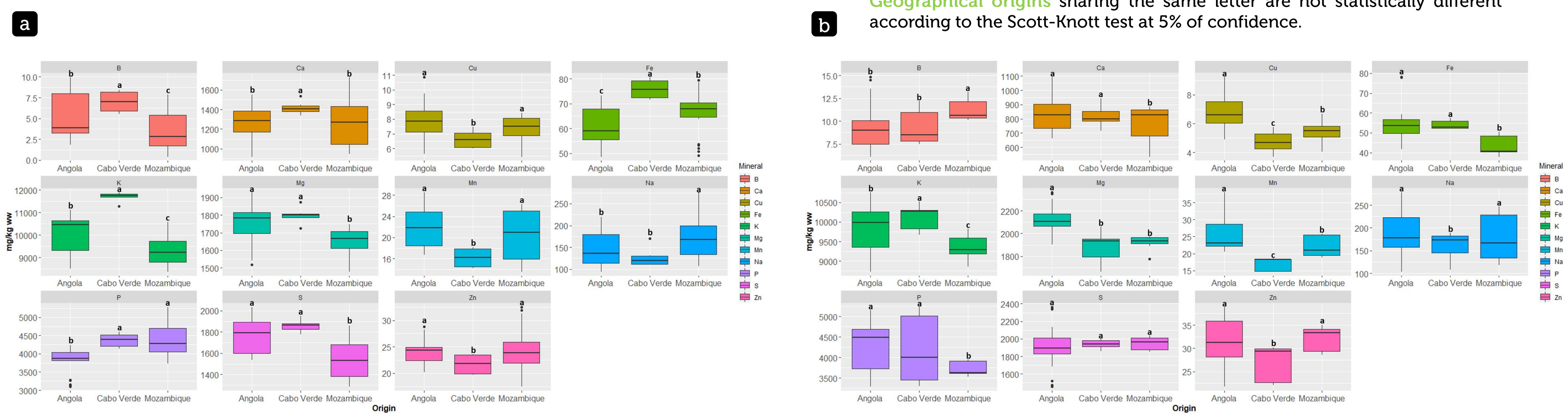


Figure 2. Variation of mineral content (mg/kg ww) and the geographical origin for the studied samples: **(a)** *Phaseolus vulgaris* (21 accessions), and **(b)** *Vigna unguiculata* (17 accessions).

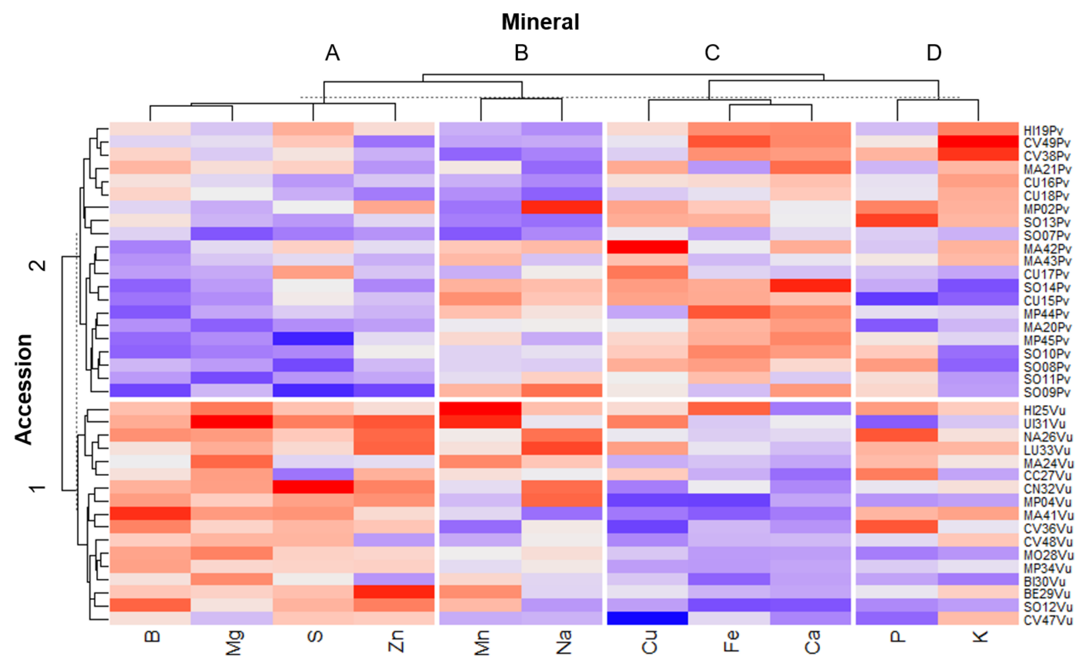
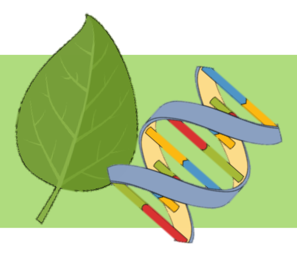


Figure 3. Heatmap of the 21 *Phaseolus vulgaris* and 17 *Vigna unguiculata* accessions obtained from the chemical characterization data for the content of 11 minerals. Red and blue boxes indicate high values and low values respectively.

CYTOGENOMIC DIVERSITY



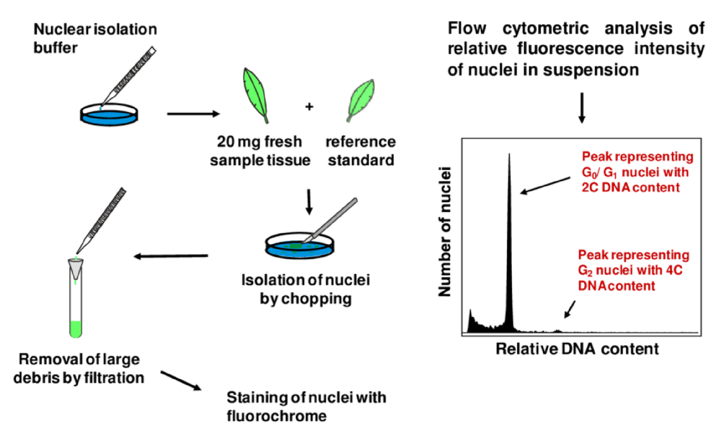


Figure 4. Sample preparation procedure for flow cytometry measurements developed. Figure from Loureiro (2007).

Table 1. Comparison of the average genome size of the *Phaseolus vulgaris* and *Vigna unguiculata* accessions estimated by flow cytometry.

Species	Genome size (Mbp)		H.G.[b]
	Average	stDev[a]	
Vigna unguiculata	1414.7	86.2	A
Phaseolus vulgaris	1337.4	33.3	B

[a] Standard deviation; [b] Homogeneous groups: accessions sharing the same letter for each mineral are not statistically different according to the Scott-Knott test at 5% of confidence.

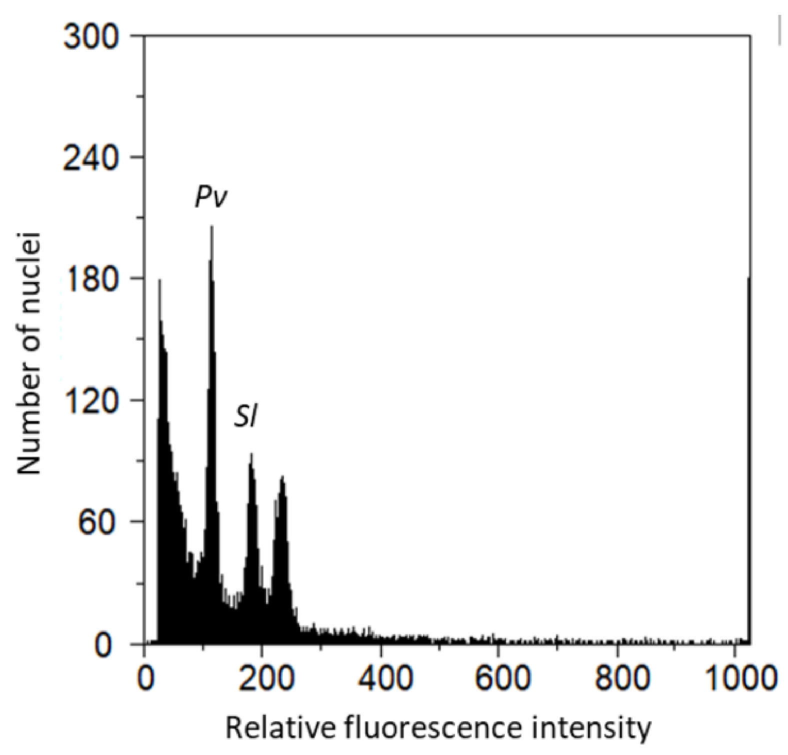


Figure 5. Flow cytometric histogram of relative fluorescence intensities from propidium iodide-stained nuclei of *Phaseolus vulgaris* sample MA20Pv (Pv) and *Solanum lycopersicum* L. (Sl), used as an internal reference standard.

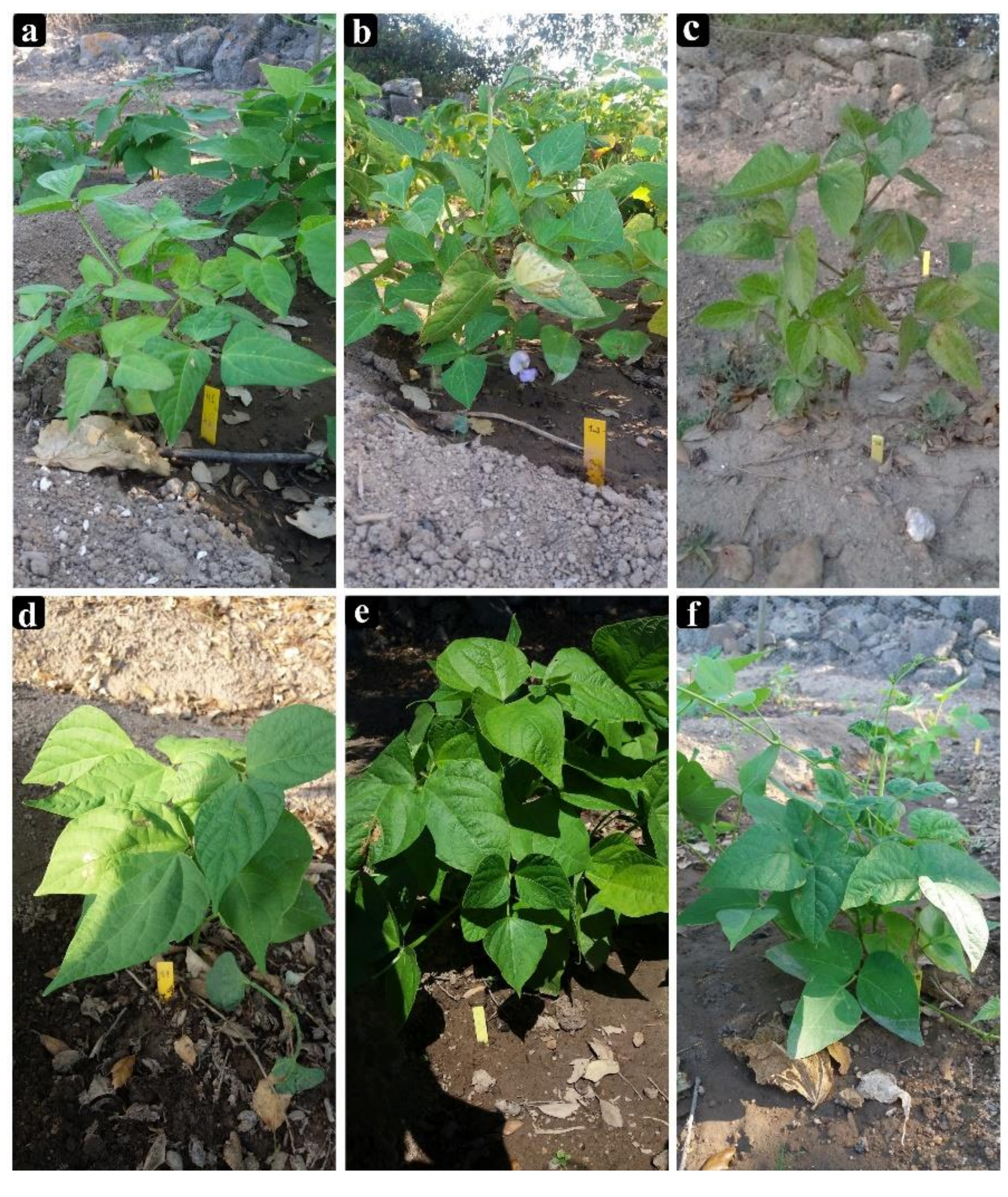


Figure 6. Cultivated samples collected in different African countries: **(a)** *Vigna unguiculata* from Malanje, Angola (MA41Vu); **(b)** *V. unguiculata* from Sofala, Mozambique (SO12Vu); **(c)** *V. unguiculata* from Santiago, Cabo Verde (CV36Vu); **(d)** *Phaseolus vulgaris* from Cunene, Angola (CU17Pv); **(e)** *P. vulgaris* from Sofala, Mozambique (SO10Pv); and **(f)** *P. vulgaris* from Santiago, Cabo Verde (CV38Pv). Plants growth under controlled conditions, between June 2019 and February 2020, at Instituto Superior de Agronomia of the University of Lisbon (ISA/UL).

NEW DATA contributes to the valorization of plant genetic resources & food security in Africa!

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REFERENCES:

Brilhante, M., Varela, E., P Essoh, A., Fortes, A., Duarte, M. C., Monteiro, F., Ferreira, V., Correia, A. M., Duarte, M. P. and & Romeiras, M. M. (2021). Tackling Food Insecurity in Cabo Verde Islands: The Nutritional, Agricultural and Environmental Values of the Legume Species. *Foods, 10*(2), 206.

Broughton, W. J., Hernandez, G., Blair, M., Beebe, S., Gepts, P., & Vanderleyden, J. (2003). Beans (Phaseolus spp.)–model food legumes. *Plant and soil, 252*(1), 55-128.

Catarino, S., Brilhante, M., Essoh, A. P., Charrua, A. B., Rangel, J., Roxo, G., Varela, E., Moldão, M., Ribeiro-Barros, A., Bandeira, S., Moura, M., Talhinhas, P., & Romeiras, M. M. (2021). Exploring physicochemical and cytogenomic diversity of African cowpea and common bean. *Scientific Reports, 11*(1), 12838. https://doi.org/10.1038/s41598-021-91929-2.

Catarino, S., Duarte, M. C., Costa, E., Carrero, P. G., & Romeiras, M. M. (2019). Conservation and sustainable use of the medicinal Leguminosae plants from Angola. *PeerJ, 7*, e6736.

Iqbal, A., Khalil, I. A., Ateeq, N. & Khan, M. S. Nutritional quality of important food legumes. *Food Chem. 97*, 331-335 (2006).

Lonardi, S., Muñoz-Amatriaín, M., Liang, Q., Shu, S., Wanamaker, S. I., Lo, S., ... & Close, T. J. (2019). The genome of cowpea (Vigna unguiculata [L.] Walp.). *The Plant Journal, 98*(5), 767-782.

Loureiro, J. C. M. (2007). Flow cytometric approaches to study plant genomes (Doctoral dissertation, Universidade de Aveiro (Portugal)).

Maxted, N., Mabuza-Dlamini, P., Moss, H., Padulosj, S., Jarvis, A., & Guarino, L. (2004). An ecogeographic survey: African Vigna, Systematic and ecogeographic studies of crop genepools 10. *IPGRI, Rome.*

Singh, B. B. (2014). Cowpea: the food legume of the 21st century. Madison, WI: Crop Science Society of America. *Inc. doi, 10*(2135), 2014.

Vidigal, P., Romeiras, M. M., & Monteiro, F. (2019). Crops diversification and the role of orphan legumes to improve the Sub-Saharan Africa farming systems. In *Sustainable Crop Production*. IntechOpen.

Yeken, M. Z., Akpolat, H., Karaköy, T., & Çiftçi, V. (2018). Assessment of Mineral Content Variations for Biofortification of the Bean Seed. *International Journal of Agricultural and Wildlife Sciences, 4*(2), 261-9.