

Improving Nutritional Quality of Cowpea (*Vigna unguiculata*) by Soaking Process



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ABSTRACT

Effect of soaking in water and acetic acid solutions to improve nutritional qualities of cowpea was studied. Cowpeas were soaked in tap water, 1 and 2% acetic acid solutions at 4, 8, 16 and 24 hours. Afterwards, phytates and minerals (Fe and Zn) content were quantified. The phytates content of soaked cowpea grain decreased with the acidity of the soaking solution and the duration of treatment. Compared to the raw material, phytates content decrease by 1.48 to 37.59% in samples soaked in tap water at 4 to 24 hours. Likewise, the phytates content decrease by 7.98 to 41.69 in samples soaked in 1 and 2% acetic acid solution at the same soaking time. The best reduction of phytates content (41.69%) is obtained with acetic acid 2% within 24 hours. Regarding minerals, a slight variation with soaking time in their content was obtained. Acetic acid solutions induced a significant removal of zinc but not for iron. The results revealed that for the two treatment, soaking with vinegar for 24h allows a better reduction of phytates with the best bioavailability of iron but low for zinc; soaking in tap water during 24h gives a good bioavailability for iron and a moderate one for zinc. So, we can say that, if intention is to obtain a product rich in iron with less phytates, it is preferable to use 2% acetic acid for 24h which can be adopted in households.

MATERIALS AND METHODS

Sampling of Cowpea Seeds.

Dry cowpea seeds were generously provided by ISRA for the experiments



Cowpea seed

Soaked Cowpea seed



Soaking of Cowpea Seeds Cowpea samples were processed from the same way for analyzes. They were soaked in order to reduce phytates in seeds. Two types of solutions were used: tap water and acetic acid (vinegar) at 1 and 2% corresponding to 1 and 2° respectively.

Acetic acid is a weak dietary acid which is found in commercial vinegar at 6° ordinary used in households. Treatments were carried out by soaking cowpea in water or acetic acid solutions with the ratio of 1:3 w/v (grain/solution), at room temperature (25° C). Four times soaking were applied: 4, 8, 16 and 24 hours. The pH of tap water was 7.0 and those of the 1 and 2% acetic acid solutions were 4.0 and 3.0 respectively. The samples obtained were named NnT (niébé not treated), NE (niébé soaked in water), NAc1 and NAc2 (niébé soaked in 1% and 2% acetic acid respectively).

Sample Preparation for Analysis

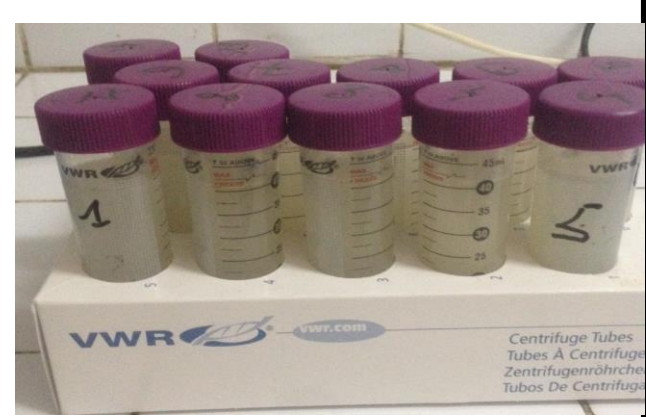
Beyond soaking essays, cowpea samples were dried at 75° C in an oven during 24 hours. They were finely ground to a fine powder (particle size of 0.5 mm) using a laboratory mill 3100 (Perten instruments) in order to be analyzed.

Phytic Acid Quantification

Phytic acid content of cowpea was determined using Latta and Eskin [15] and Vaintraub *et al.* [16] methods with some modifications. 1.2 g of each sample was weighed and introduced into 50 ml tubes. 40 ml of 2.4% concentrated HCl was added to each tube, at room temperature. Tubes were vortexed during 2 hours every 10 min for 15s. After 2 hours, the tubes were centrifuged during 30 min at 3000 rpm. The clear supernatant was used for the phytate quantification. 5 mL of Wade reagent (0.03% solution of FeCl₃·6H₂O containing 0.3% sulfosalicylic acid in water) was added in 15 mL of the supernatant; the mixture is vortexed for 5s. The new supernatant was then transferred in a cell to read absorbance at 500 nm. Distilled water was used as a blank. Phytate content were determined in triplicate.

Zinc and Iron Quantification

Minerals content were determined in triplicate. Zinc and Iron contents were determined by atomic absorption spectrophotometry after mineralization at 550° C for 4 hours. Hydrochloric acid was added to the ash obtained and then evaporated to dryness. The residue was dissolved in Nitric acid and this solution was analysed by Atomic Absorption Spectrophotometer (AAS) using the technique of flame [17].



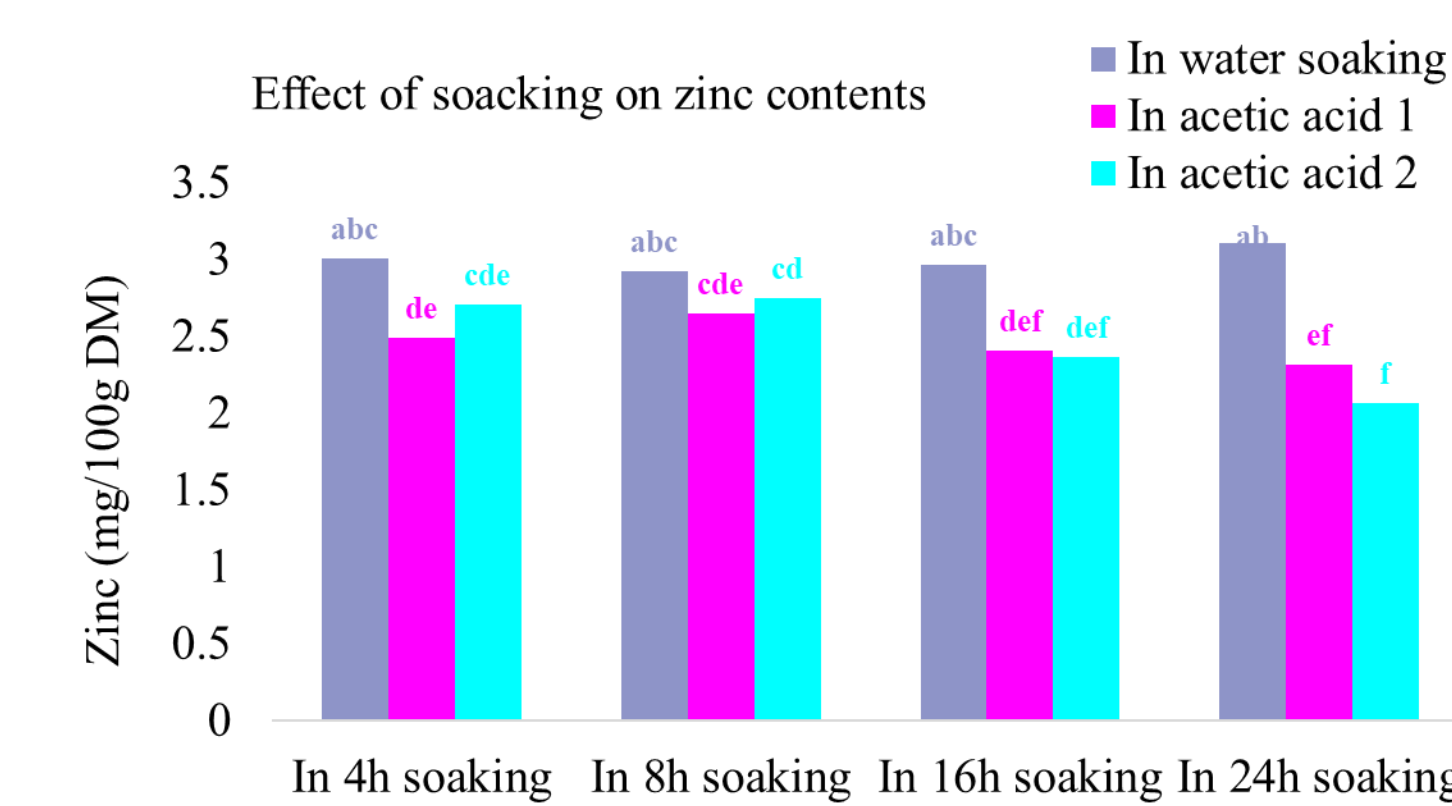
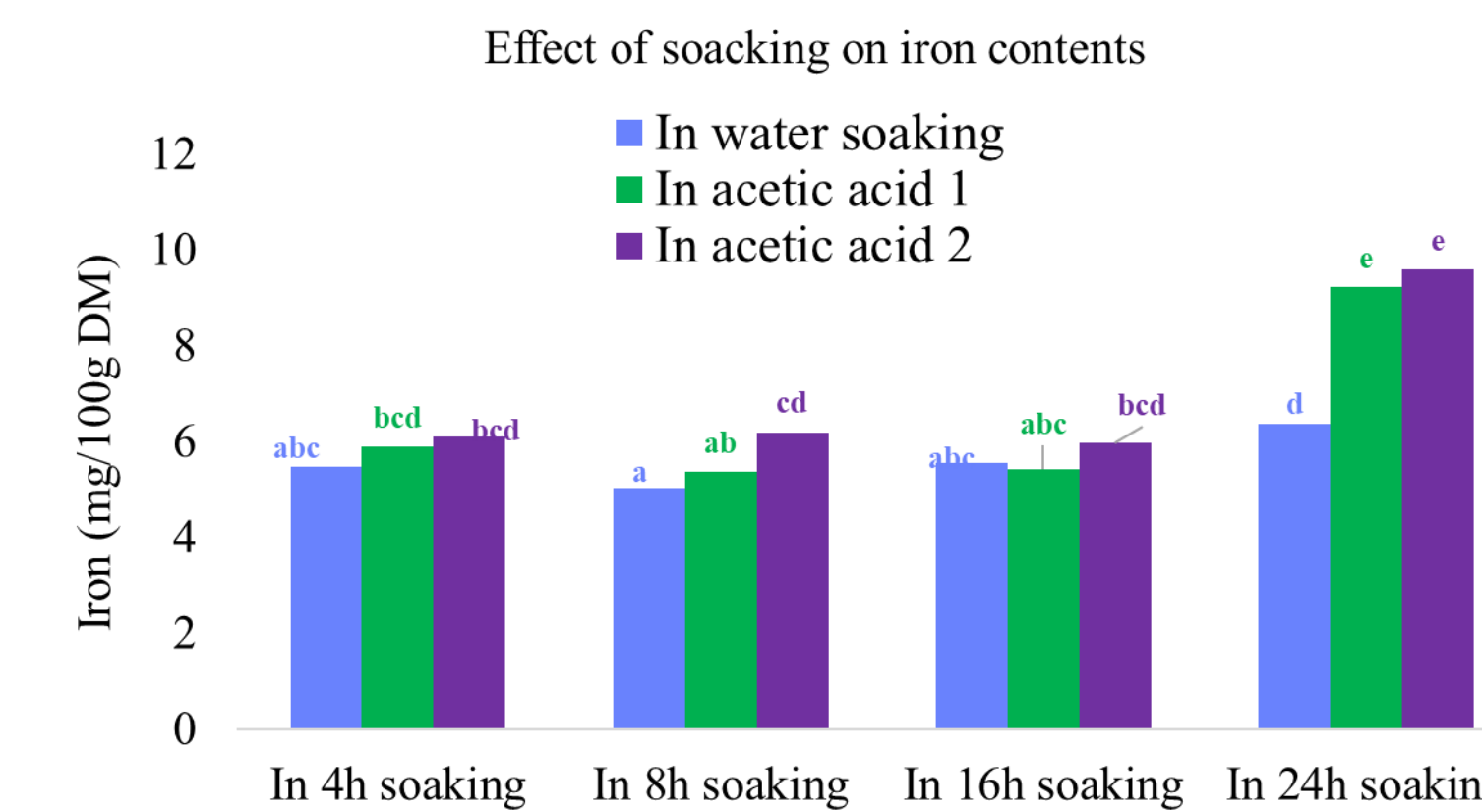
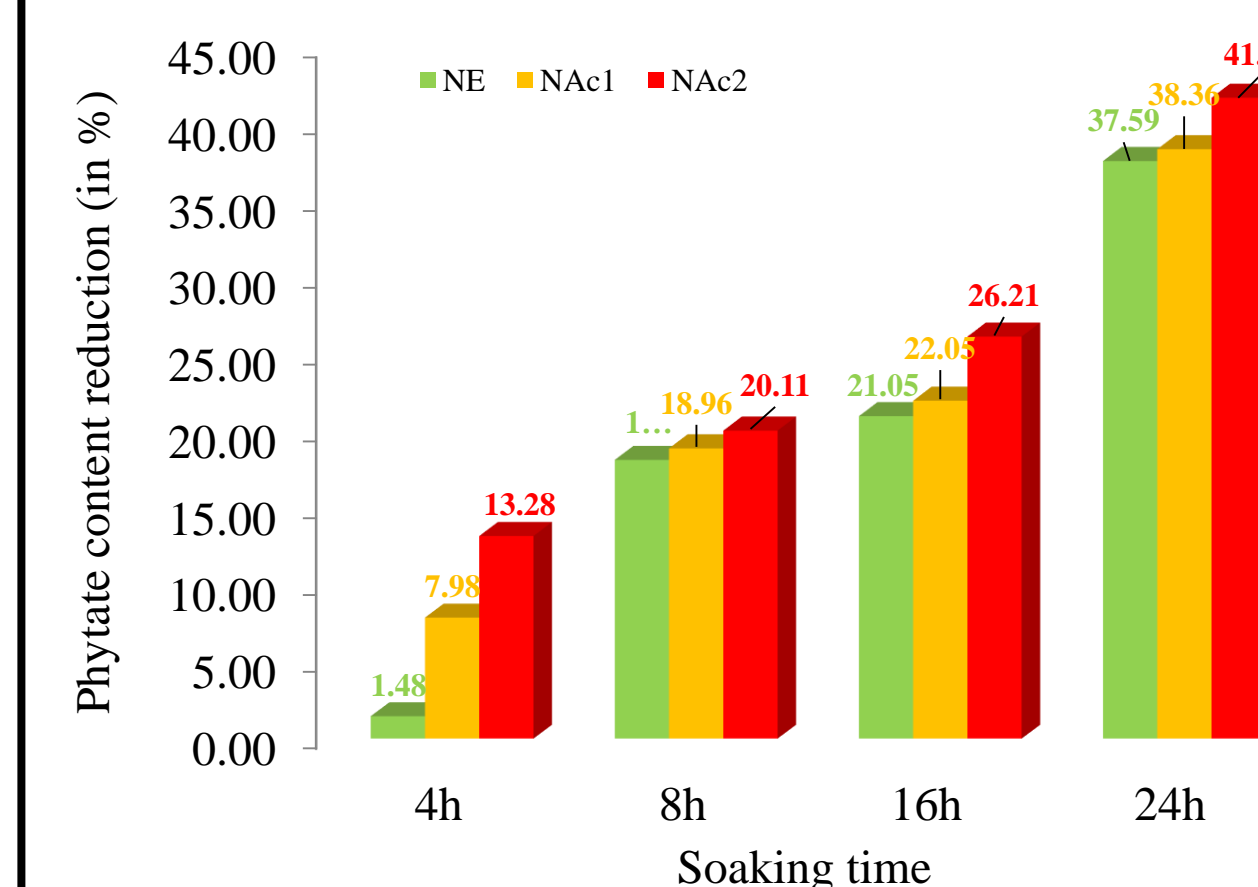
INTRODUCTION

The sub-Saharan Africa population's diet is mainly based on cereals and legumes. Legumes are sources of good quality of protein, carbohydrates, various minerals and vitamins [1]. Having less expensive proteins is an increasing demand across the world and particularly in under-developed countries [2]. Cowpea is one of the main legumes consumed in Senegal which annual production increase from 99924 tons in 2017 to 117784 tons in 2018 [3]. The protein content of cowpea is between 18 and 35 g/100 g [1,4,5]. However, the presence of several antinutritional factors (ANFs) can limit the nutritional values of this legume by reducing the bioavailability of some essential minerals. Phytic acid (phytate; myo-inositol 1,2,3,4,5,6, hexakisphosphate) is one of the ANFs among naturally occurring constituent of plant seeds, roots, tubers, some fruits and vegetables. It acts as a storage form of phosphate [6]. Specifically, phytic acid is known to build complexes with essential dietary minerals such as calcium, zinc, iron and magnesium, making those biologically unavailable for absorption. Phytic acid can also chelate vitamins and potentially contribute to their deficiency and to disease pellagra [7].

In cowpea, phytates content is 559 mg/100g DM [8]. Through previous research, several dipping methods are shown to reduce the phytate content in legume seeds [9,10,11]. Among those methods, soaking, sprouting, fermentation, extrusion cooking and steam pre-cooking are demonstrated [9,11,12]. Soaking cowpea in water during 24 hours lost 8.4% of phytates content [12]. A better reduction of 22.4 and 23.7% on two varieties of cowpea after 24 hours of soaking has been obtained [13]. Unfortunately, these techniques can remove or reduce some recommended components which may be required to enhance nutritional quality [14].

Thus, this study aims to determine optimal conditions which either reduces phytates or preserves nutrients in cowpea.

RESULTS AND DISCUSSION



The results presented in figures show that the phytates content of soaked cowpea grain decreased with the acidity of the soaking solution but also the treatment time. For minerals, results reveal that soaking water does not significantly reduce neither zinc nor iron. Acetic acid solutions induced a significant removal of zinc but not for iron. The best reduction of phytates content (41.69%) is obtained with acetic acid 2% within 24 hours.

Acetic acid soaking solution (vinegar) has a better reduction rate of phytates compared to soaking water. Thus, would be facilitated by the permeability of membrane walls which became weaker (Vijayakumari *et al.* [18]).

Acetic acid solutions induced a significant removal of zinc but not for iron. The low zinc removal is explained by strong link with protein cells [19]. The latest shows a slight increase after 24 hours. This abnormal situation should may occur with reabsorption of minerals by the grains.

To predict the inhibitory effect of phytate on iron and zinc bioavailability in cowpeas varieties, phytate/iron, phytate/zinc molar ratios were calculated (see Table).

These results suggest that the bioavailability of zinc in most samples (low contrary to iron) would be linked to higher contents of. Phytate. Referring to these values, only NE24 gives a moderate bioavailability of zinc (30%). According to Weaver [19], zinc build the most stable complex with phytate and this would affect mineral in terms of bioavailability.

Molar ratio of phytate/zinc and phytate/iron of cowpea samples

	Molar ratio phytates/zn	Molar ratio phytates/Fe
NnT	17.75	7.47
NE4	18.75	8.71
NE8	15.99	6.68
NE16	15.20	6.85
NE24	11.47	4.74
4NAc1	21.11	8.26
4NAc2	18.30	6.91
8NAc1	17.51	7.69
8NAc2	16.64	6.23
16NAc1	18.52	6.92
16NAc2	17.78	5.95
24NAc1	15.20	3.23
24NAc2	16.14	2.94

CONCLUSIONS

Soaking is treatment which is commonly used to prepare cowpea in household. The study reveals that soaking promoted significant phytate reduction in acetic acid solution which can be compare to vinegar. This treatment reduced zinc and iron but did not modified seriously their bioavailability. Then the method can be adopted in households.

REFERENCES

- Xu, B., & Chang, S. K. C. (2009). Phytochemicals in cowpea (*Vigna unguiculata*). *Journal of Agricultural and Food Chemistry*, 57(22), 10718-10731. doi:10.1021/jf902594n.
- Omenna, E. C., Olanipekun, O. T., & Kolade, R. O. (2016). Effect of boiling, pressure cooking and germination on the nutritional and antinutrients content of cowpea (*Vigna unguiculata*). *ISAB Journal of Food and Agricultural Sciences*, 6(1), 1-8.
- ANSD, 2018. (National Agency of Statistics and Demography). Monthly Bulletin of Economic Statistics. Dakar 109 p.
- Mune MA, Minka S R, Mbome L I. (2013). Response surface methodology for optimisation of protein concentrate preparation from cowpea *Vigna unguiculata* (L.) Walp. *Food Chem* 110: 735-741.
- Obasi, N. E., Unanma, N. C., & Nwofia, G. E. (2014). Effect of dry heat pre-treatment (toasting) on the cooking time of cowpeas (*Vigna unguiculata* L. Walp). *Nigerian Food Journal*, 32(2), 16-24.
- Reddy, N. R., Sathu, S. K. 2002. Food phytates. Boca Raton, Florida: CRC press. 280 p. ISBN 9781566768672.
- Gonçalves, A., Grafo, P., Barros, A., Dominguez-Perez, R., Trindade, H., Rosa, E. A., ... & Rodrigues, M. (2016). Cowpea (*Vigna unguiculata* L. Walp), a renewed multipurpose crop for a more sustainable agri-food system: nutritional advantages and constraints. *Journal of the Science of Food and Agriculture*, 96(9), 2941-2951.
- Isabelle Lestienne, ChristèleCard-Vernière, Claire Mounquet, Christian Picq, Serge Trèche. Effects of soaking whole grain and legume seeds on iron, zinc and phytate contents. *Food Chemistry*. 2005; 89(3): 421-425. Doi:10.1016/j.foodchem.2004.03.040.
- Diouf, A., Sarr, F., Sene, B., Ndiaye, C., Fall, S. M., ... & Ayessou, N. C. (2019). Pathways for Reducing Anti-Nutritional Factors: Prospects for *Vigna unguiculata*. *El-Adawy TA Nutritional Composition and Antinutritional Factors of Chickpeas (Cicer arietinum L.) Undergoing Different Cooking Methods and Germination*. *Plant Food Hum. Nutr.* 2002; 57(1): 83-97.
- Egounley M and OC AwohEffect of Soaking, Dehulling, Cooking and Fermentation with RhizopusOligosporus on the Oligosaccharides, TrypsinInhibitor, Phytic Acid and Tannins of Soybean (Glycine max Merr.). *Cowpea (Vigna unguiculata L. Walp) and Groundbean (Macrotylomogeaocarpiflora)*. *J. Food Eng.* 2003; 56(2-3): 249-254.
- Andriantsoa Z J. Evolution of antinutritional factors of two varieties of cowpea (*vignaanguiculata*), voanembamena and voanembafotsy, during germination. (DEA dissertation of Biochemistry applied to the sciences of food and nutrition). Faculty of Science: Antananarivo University. 2006.
- Razafitsalana N. Evolution of antinutritional factors of seeds of two varieties of voandrouz, mara and fotsy, during germination. DEA dissertation of Biochemistry applied to the sciences of food and nutrition). Faculty of Science: University of Antananarivo. 2006.
- Ben Souilah F. 2015. Caractérisation du comportement des micronutriments d'intérêt et des composés antinutritionnels des poischiches et du niébé au cours de transformation. Mémoire de fin d'étude pour l'obtention du diplôme de master en biologie santé, université Montpellier, 47p.
- [15] Latta M and Eskin M. 1980. *Journal of Agricultural and Food Chemistry* 28 (6), 1313-1315 DOI: 10.1021/jf00232a049.
- [16] Vaintraub I A; Laptewa N A. 1988. Colorimetric determination of phytate in unpurified extracts of seeds and the products of their processing. *Analytical Biochemistry*, 175: 227-230pp.
- [17] Asee 18th ed.rw.2007. method 968.08(4.8.02).
- [17] Udemi, E. A., Ekwu, F. C., & Ibinguzo, J. N. (2007). Antinutrient factors of vegetable cowpea (*sesquipedalis*) seeds during thermal processing. *Pakistan Journal of Nutrition*, 6, 194-197.
- [18] Vijayakumari, K., Siddharaju, P., Pugalenti, M., & Janardhanan, K. (1998). Effect of soaking and heat processing on the levels of antinutrients and digestible proteins in seeds of *Vigna aconitifolia* and *Vigna sinensis*. *Food Chemistry*, 63(2), 259-264. doi:10.1016/S0939-1467(98)00074-0.
- [19] Weaver, C. M.; Kamran, S. Phytate and mineral bioavailability. In: REDDY, N. R.; SATHI, S. K. (Eds.). *Food phytates Florida: CRC*. 2002. p. 211-223.

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