COMPARATIVE STUDIES OF THE AMINO ACID PROFILE OF BIOLOGICALLY TREATED AND UNTREATED CASTOR (*Ricinus communis*) SEED CAKE

O. M. Ameen[1*], M. A. Belewu[2], J. O. Ayeni[1] and T. O. Akinnagbe[1]

1. Department of Chemistry, University of Ilorin, Ilorin
2. Department of Animal Production, University of Ilorin, Ilorin

*Correspondents Author: moameen@unilorin.edu.ng; aommohd@yahoo.com; +2348035019199

Received 16 May 2018; accepted 07 January 2019, published online 15 March 2019

ABSTRACT:

Ricinus communis (Castor) seed was defatted with n-hexane and divided into two. A part was inoculated with *Penicillum expansium* for six weeks, while the other part was used as control. Both the biologically detoxified castor seed cake (BDCSC) and the undetoxified castor seed cake (UCSC) were analysed for nutritional value by carrying out proximate analyses, investigating the mineral contents and the amino acid profiles. The results revealed a reduction in the crude protein and crude fiber contents in the BDCSC, while there was increment in the moisture and ash contents after the treatment, but the fat contents was not affected. Though, there was a decrease in the crude protein contents, the amino acid profile of the BDCSC is however comparable with those of soybean as well as FAO/WHO recommended minimum daily limits. This research has revealed that treating castor seed cake biologically may improve the availability of the cake for animal feeds. Nonetheless, the detoxified seed cake needs to be analysed further in order to determine the level of its anti-nutrient contents.

KEYS: Castor seeds, *Penicillum expansium*, Detoxification, Proximate analysis, Amino Acid Profile

INTRODUCTION

The daily increase in demand for protein-rich food and feedstock fascinated studies and exploitation of diverse non-edible organic materials that contain high protein content among which is the residual cake of the de-oiled castor seed [1]. Castor plant (*Ricinus communis*) is an angiosperm plant belonging to the kingdom plantae, and the family of Euphorbiacea (the fourth largest of the angiosperms comprising over 300 genera and about 7500 species widely distributed in tropical Africa) and sole member of the genus Ricinus and sub-tribe of Ricininae. It is an oilseed shrub of significant economic importance because of its several potentials in industry and medicine [2].

Initially, oil extraction has been the major concern of the countries that grow castor plants but not until recent times that researchers began to ponder on the bulkiness, economic values and uses of the residual cake generated after extraction of the oil. For example, for each ton of castor seed pressed for oil extraction, a half-ton of residue seed cake is obtained [3]. This exploitation and development of castor bean cake as substitute for protein source for animals could offer a good scope to meet dynamic increasing demand in large quantity, especially in developing countries. The seed cake of *R. communis* contains micro and macro-element components like N, P, K, Na, Mg and S which supplement plant nutrient, however, the uses of the cake is not limited to nutrition but can also be used as fertilizer, because of its high organic constituent [3]. Meanwhile, the presence of toxins in the cake restricts its usefulness in feed nutrition. In order to satisfy feedstock requirement, detoxification of the toxins is inevitable [4-5]. Various methods including physical, chemical and biological treatment have been employed to detoxify the seed cake of industrial castor oil production to be used for feeding or other purposes [6 – 10]. In view of these, the present research was designed to study the effects of *Penicillum expansium* treatment to eliminate the antinutrional factors in defatted castor seed cake.

MATERIAL AND METHODS

Preparation of Seed cake

Samples of matured dried castor seeds (*R. communis*) were collected from Ibadan, Western Nigeria. The seeds were prepared dried and crushed into paste. The paste was defatted according to Standard Official and Tentative Method of Oil Chemists Society procedure [11]. The defatted seed cake was dried and kept for analysis.

Biological treatment

The defatted cake was divided into two and a portion was detoxified according to the method described earlier [12].

Chemical Analysis

The proximate analysis of both the Biologically Detoxified Castor Seed Cake (BDCSC) and Undetoxified Castor Seed Cake (UCSC) were carried out according the procedure of Association of Official Agricultural Chemists (AOAC) [13]. The ash and moisture were determined by the procedure of AOAC [11], while the crude fibre content was determined by the procedure of AOAC [13]. The crude proteins were determined using modified Biuret method [14].

Mineral Analysis

The mineral analysis was determined using the procedure of AOAC [13].

Amino Acid Profile Analysis

The Amino Acid profile of the BDCSC and UCSC were determined using methods reported in the literature [15]. The Applied Biosystems PTH Amino Acid Analyzer was used for the analysis.

RESULTS AND DISCUSSION

Proximate analysis

The proximate composition of crude protein, crude fibre, fat, ash contents and the dry matter of the BCDSC and UCSC are presented in Table 1.The results obtained for the BDCSC are slightly different when compared with the UCSC which is similar to those reported elsewhere [2]. The dry matter constituent of the biologically detoxified castor seed cake (93 %) is higher than those obtained from the hot water (90.83 %), lye (90.41 %), and fermentation (90.44 %) treatments respectively as presented in related studies [2].Whereas, the dry matter (95.25 % of the UCSC is 2.36 % greater than those of BDCSC. The crude protein (8.12±0.1), crude fibre (7.8±0.01), fat (47.4±0.01) and ash (9.1±0.01) contents of the BDCSC are lower than those of the UCSC (Table 1).

Also, the crude protein content (8.12 %) of the biologically detoxified seed cake in this report is lower than the crude protein content of seed cake reported in related studies [2, 9] using other treatment methods. It is also less than the crude protein of *Jathropha curcas* bio-detoxified cake [16 – 17]. As ascertained in literature, the effect of the detoxification on the seed cake reduced the amount of crude protein present, [16 – 17], which agree with the result obtained in this report.

Table 1: Proximate analysis of treated and untreated seed cake

PARAMETERS	BDCSC (%)	UCSC (%)
Crude protein	8.12±0.1	15.42
Crude fibre	7.8±0.01	12.65
Ash content	9.1±0.01	2.51
Fat content	47.4±0.01	47.4
Moisture content	7.0±0.005	4.75

Amino Acid Profile

The quality of any protein source is related to its amino acid composition, digestibility, bio-availability and ability to supply the essential amino acid in the amount required by the organism consuming it. Based on these facts, there are two methods of assessing the quality of proteinous species, and amino acid analysis [18]. Table 2 presents the results of the amino acid analysis of the BCDSC and the UCSC. Results presented in Table 2 indicate that there was a decrease in the amino acids of the BDCSC, the only exception being alanine whose value was the same for both the BDCSC and UCSC (4.59 g/100g), this could be as result of the fermentation process. These results also compared favourably with those reported in literature

[1, 19] in which hot water was used for the detoxification of castor seed cake.

Table 2: Amino acid compositions of treated and untreated of castor seed cake

S/N	Amino acid	Concentration (g/16gNg)		Laufey and Hans [2]	Onwuliri and Anekwe [19]
		UCSC	BCDSC	g/16 g N	g/16g N
1	Leucine	6.39	5.28	6.42	5.1
2	Lysine	3.10	2.63	2.68	7.3
3	Isoleucine	5.24	4.91	4.68	3.8
4	Phenyalanine	4.70	3.90	4.02	4.0
5	Tryptophan	1.13	0.92	0.31	ND
6	Valine	5.38	4.80	5.44	3.1
7	Methionine	1.50	1.28	1.51	1.0
8	Proline	3.55	3.25	3.74	9.7
9	Arginine	9.98	9.21	8.61	7.4
10	Tyrosine	2.75	2.58	2.82	2.3
11	Histidine	1.72	1.60	1.25	2.2
12	Cysteine	0.24	ND	0.68	ND
13	Alanine	4.59	4.59	4.26	3.8
14	Glutamic acid	13.17	12.34	18.87	13.6
15	Glycine	0.31	ND	4.31	5.7
16	Threonine	3.19	2.89	3.44	4.8
17	Serine	3.40	3.24	5.44	4.1
18	Aspartic acid	8.93	8.34	9.67	12.4
19	Hydroxyproline	ND	ND	0.28	ND

ND= Not Detected, Norleucine= standard

Table 3 presents the essential amino acid of the UCSC and BCDSC in comparison with those of soybean and WHO/FAO Standard. Table 3 revealed that the levels of essential amino acids for the BCDSC were higher than the WHO/FAO recommended standards with the exception of lysine, tyrosine, tryptophan and methionine in which lower values were obtained, while cysteine was not detected. On the other hand, in comparison with the amino acid profile of soybean, it is observed that the values obtained for the BCDSC were lower than those for soybeans except isoleucine and methionine which recorded higher values while for valine the same value was recorded for both BCDSC and soybean (Table 3). This observation indicates that the nutrition value of biological detoxified castor seed cake compare favourably with that of soybeans.

It has been reported that the utilization of lysine and isoleucine in protein is affected by the amount of leucine present. A leucine to lysine ratio greater than 4.6 will hinder the utilization of lysine [18]. However, the ratio of Leucine to Lysine in this report is 2.0 and 2.1 for BCDSC and UCSC respectively, so the utilization of Lysine for both seed cakes is ideal.

The Total essential amino acid of *R. communis* for BCDSC (37.45 g/100g) and UCSC (42.33 g/100g) which corresponds to (52.19%) and (53.40%) respectively showed that castor seed cake in rich in essential amino acid. The total amino acid of castor seed cake is greater than that of *Anarcadium occidentale* concentration (35.3g/100g) reported elsewhere [20]. Also the Essential Aliphatic Amino Acid (EAAA), Isoleucine, Leucine and Valine, for BCDSC which constitute the hydrophobic regions of protein (14.99g/100g protein) is less than the EAAA (16.4g/100g protein) of *Anarcadium occidentale* (Cashew nut) as reported by other workers [20]. However, the total amino acid of BCDSC (37.45 g/100g) is lower while the UCSC (42.33 g/100g) is higher than the treated castor seed cake (38.36 g/100g) reported by other workers [1]. Also, the total amino acid of both the BCDSC and UCSC are lower than the total amino acids of some Nigeria legume concentrates; liman bean (44.88g/100g protein), pigeon pea (48.11g/100g protein) and African yam bean (48.28g/100g protein) [21]. Nevertheless the protein of *R. communis* to a reasonable level satisfied the FAO requirements (Table 3) [22] for the essential amino acids.

Table 3: Essential amino acid compositions of treated and untreated of castor seed cake, soybean and essential amino acid pattern suggested by FAO/WHO (g/100 g protein)

	UCSC	BCDSC	Soybean [23]	WHO/FAO standard
Isoleucine	5.24	4.91	4.8	4.2
Leucine	6.39	5.28	8.0	4.2
Lysine	3.10	2.63	6.4	4.2
Phenyalanine	4.70	3.90	4.8	2.8
Tyrosine	2.75	2.58	3.2	2.8
Cystine	0.24	ND	0.8	2.0
Methionine	1.50	1.28	0.9	2.2
Threonine	3.19	2.89	4.0	2.8
Tryptophan	1.13	0.92	1.3	1.4
Valine	5.38	4.80	4.8	4.2

ND= Not Detected

Mineral Analysis:

The result of the mineral contents of the BCDSC in comparison with UCSC is presented in Table 4. The table revealed considerable decrease in the mineral contents of the treated castor seed cake with the exception of potassium, sodium and iron whose values in BCDSC were higher than that of UCSC.

Table 3: Mineral contents of the treated and untreated of castorseed cake

Metals	Concentration (mg/kg)

	UCSC	BCDSC
Ca	13.17	2.2 ±0.1
Cr	0.006	0.1 ±0.03
Fe	1.57	1.8 ±0.2
Cu	0.22	0.04 ±0.006
Cd	0.003	0.03±0.01
Zn	10.63	1.8 ±0.4
Pb	0	0.1 ±0
Mg	700	265±60
Na	5.27	64 ±0.8
K	17.7	57 ±10

The level of these mineral contents in the biologically detoxified castor seed cake is an indication that the seed cake would be a very good alternative in animal feeds because calcium is a very vital element in human metabolism. It is the chief element in the production of very strong bones and teeth in mammals. Its tolerance limit is high relative to other bio-useful metals. Magnesium also is an important electrolytic constituent of the blood which plays other vital role in body [24]. The high concentrations of sodium, potassium and magnesium make castor seed cake a good source of fertilizer for soil improvement as reported elsewhere [2].

Heavy metals must be minimal in concentration due to their toxicity at higher concentrations, heavy metals such as Cd, Pb, Cr, Cu, can cause gastrointestinal (GI) disorders, diarrhoea, stomatitis. The concentrations of these heavy metals found in this study agreed with those of Duruibe *et al*., [24].

CONCLUSION

Oil seed plants recently have been focused for source of protein for animal feed in which *R. communis* seed is not exempted. Studies in animal nutrition have shown that *R. communis* bean meal can be used as a protein supplement for ruminants. However, its utilization in monogastrics, especially poultry, has been limited because of possible deleterious effect due to the presence of ricin, and thermo stable castor allergens which are not easily removed.

The nutritional value of *R. communis* BCDSC agreed favourably (to a certain level) with the conventional oil seed and protein source (soybean) whereas, the UCSC agreed more but cannot be used as feed for animals because of the presence of toxins (Ricin a water soluble protein). However, the deficient amino acids can be supplemented for improved growth. Thus, for efficient and total use of castor seed cake in animal feed formulation, with high expectancy of effective utilization, treat the raw cake in order to make it suitable for animal consumption and this would invariably reduce competition between man and livestock for the conventional sources of proteins. Thus, cultivation of this potentially rich plant is encouraged.

REFERENCES

1. V. Laufey and F. Hans (1971) Castor Bean Meal as a Protein Source for Chickens: Detoxification and Determination of Limiting Amino Acids, *The Journal of Nutrition*, **101(9)**: 1185–1192. https://doi.org/10.1093/jn/101.9.1185
2. T. O. Akande, A. A. Odunsi, O. S. Olabode, T. K. Ojediran (2012), Physical and Nutrient Characterisation of Raw and Processed Castor (*Ricinus communis* L.) Seeds in Nigeria, *World Journal of Agricultural Sciences*, **8(1)**: 89 – 95.
3. V. F. Keysson and L. T. M. Olga (2012), Approaches for the Detection of Toxic Compounds in Castor and Physic Nut Seeds and Cakes, In Z. Fang (ed). Biodiesel - Feedstocks, Production and Applications, InTech, DOI: 10.5772/52332. Also available from: https://www.intechopen.com/books/biodiesel-feedstocks-production-and-applications/approaches-for-detection-of-toxic-and-allergenic-compounds-present-in-castor-and-physic-nut-seeds-an
4. A. O. Ani and A. U. Okorie (2005). The effects of graded levels of dehulled and cooked castor oil bean (*Ricinus communis*, L) meal on performance of Broiler starters, *Nigeria Journal of Animal Production*, **32(1),** 54 – 60.
5. A. O. Ani and A. U. Okorie (2009), Response of broiler finishers to diets containing graded levels of processed castor oil bean (*Ricinus communis L.*) meal, *Journal of Animal Physiology and Animal Nutrition*, **93(2),** 157 – 164. doi: 10.1111/j.1439-0396.2007.00796.x.
6. P. A. dos Santos, M. C. M. M. Ludke, J. V. Ludke, C. B. V. Rabello, M. J. B. dos Santos, and T. R. Torres (2015), Characterization and Digestibility of Detoxified Castor Oil Meal for Japanese Quails, *Brazilian Journal of Poultry Science*, 65 – 72. DOI: http://dx.doi.org/10.1590/1516-635xSpecialIssue
7. D. E. Faria Filho, A. N. Dias, W. A. Carneiro, C. F. D. Bueno, J. B. Matos Júnior, A. L. C. Veloso and P. A. Rodrigues (2016), Detoxified Castor Seed Cake for Broilers, *Brazilian Journal of Poultry Science*, **18(1)**: 069 – 072. DOI: http://dx.doi.org/10.1590/1516-635x1801069-072
8. N. L. Sousa, G. B. Cabral, P. M. Vieira, A. B. Baldoni and F. J. L. Aragão (2017) Bio-detoxification of ricin in castor bean (*Ricinus

communis L.) seeds, Scientific Reports, **7**, 15385. DOI:10.1038/s41598-017-15636-7

9. J. O. A. Okoye, C. A. Enunwaonye, A. U. Okorie and F. O. I. Anugwa (1987), Pathological effects of feeding roasted castor bean meal *(Ricinuscommunis)* to chicks, *Avian Pathology*, **16(2)**, 283 – 290. DOI: 10.1080/03079458708436375
10. M. C. Ojinnaka and P. C. Ojimelukwe (2013) Study of the Volatile Compounds and Amino Acid Profile in *Bacillus* Fermented Castor Oil Bean Condiment, *Journal of Food Research*, **2(1)**, 191 – 203. doi:10.5539/jfr.v2n1p191
11. AOAC (Association of Official Analytical Chemists) (1990). Official Methods of Analysis. 15th Ed. Arlington, VA: Association of Official Analytical Chemists.
12. M. A. Belewu, O. Ahmed and S. O. Ibrahim (2011), Solid state fermentation of *Jatropha curcas* kernel cake with cocktail of fungi. *International Journal of Bioscience*, **1(1)**: 12 – 19.
13. AOAC (Association of Official Agricultural Chemists) (1964) Official Method of Analysis of the AOAC, W. Horwitz Editor Eighteen Edition, Washington; D. C., AOAC.
14. A. J. Pinckney (1961). The biuret test as applied to the estimation of wheat protein, *Cereal Chemistry*, **38**: 501 – 506.
15. L. V. Benitez (1989). Amino Acid and fatty acid profiles in aquaculture nutrition studies, (23-35). In S.S. De Silva (ed.) Fish Nutrition Research in Asia. Proceedings of the Third Asian Fish Nutrition Network Meeting. Asian fish. Society Special Publication. 4, 166. Asian Fisheries Society, Manila Philippines.
16. M. D. Nunes, J. M. R. da Luz, S. A. Paes, D. P. Torres and C. M. M. Kasuya (2014), Bio-detoxification of *Jatropha curcas* seed cake by *Pleurotus ostreatus*, *African Journal of Microbiology Research*, **8(11)**: 1148 – 1156. DOI: 10.5897/AJMR2014.6617
17. M. C. M. Kasuya, J. M. R. da Luz, L. P. S. Pereira, J. S. da Silva, H. C. Montavani and M. T. Rodrigues (December 3rd 2012), Bio-Detoxification of Jatropha Seed Cake and Its Use in Animal Feed, Biodiesel, Zhen Fang, IntechOpen, DOI: 10.5772/52157. Available from: https://www.intechopen.com/books/biodiesel-feedstocks-production-and-applications/bio-detoxification-of-jatropha-seed-cake-and-its-use-as-animal-feed

18. O. M. Ameen, L. A. Usman, N. O. Muhammed, O. F. Okeola, E. O. Boluwarin and O. O. Fadeyi, (2014), Effect of Heat and Alkaline Hydrolysis on the Amino Acid Profile of *Jatropha curcas* Seed Cake, *African Journal of Food, Agriculture, Nutrition and Development*, **14(2)**; 1-13.
19. V. A. Onwuliri and G. E. Anekwe (2001), Amino Acids and Other Biochemical Components of *Ricinus communis* (Variety Minor), an Anti-conceptive Seed. *Pakistan Journal of Biological Sciences*, **4:** 866 – 868. **DOI:** 10.3923/pjbs.2001.866.868
20. M. O. Aremu, I. Ogunlade and A. Olonisakin (2007), Fatty Acid and Amino Acid Composition of Protein Concentrate from Cashew Nut (*Anarcadium occidentale*) Grown in Nasarawa State, Nigeria, *Pakistan Journal of Nutrition*, **6(5)**: 419 – 423.
21. A. A. Oshodi, K. O. Esuoso and E. T. Akintayo (1998), Proximate and amino acid composition of some under-utilized Nigerian legume flour and protein concentrates, *La Rivista Itatiana Delle Sostanze Grasse*, **75(8-9)**: 409 – 412.
22. FAO/WHO/UNU (2002), Protein and amino acid requirements in human nutrition: report of a joint FAO/WHO/UNU expert consultation. In a Joint FAO/WHO/UNU Expert Consultation on Protein and Amino Acid Requirements in Human Nutrition (2002: Geneva, Switzerland), WHO technical report series; no. 935
23. K. A. Kuiken, C. M. Lyman, M. Bradford, M. Trant and S. Dieterich (1949), Essential Amino Acid Composition of Soy Bean Meals Prepared From Twenty Strains of Soy Beans, *Journal of Biological Chemistry*, **177**: 29 – 36.
24. J. O. Duruibe, M. O. C. Ogwuegbu, J. N. Egwurugwu, (2007), Heavy metal pollution and human biotoxic effects, *International Journal of Physical Sciences*, **2(5):** 112 – 118. Available online at http://www.academicjournals.org/IJPS