

Short Communication: Identification of structural and elemental composition of *Cassia obtusifolia*

ABDALLAH YOUSIF ADAM AHMED^{1,*}, ALI A. S. MAROUF^{2,**}

¹ Collage of Education, Geneina University, Khartoum. *email: abdalalah.yousif46@gmail.com

² Institute of Laser, Sudan University of Science and Technology, Khartoum. **email: alisaheed@sustech.edu, marouf.44@gmail.com

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Abstract. Ahmed AYA, Marouf AAS. 2017. Short Communication: Identification of structural and elemental composition of *Cassia obtusifolia*. *Bioteknologi* 14: 12-15. Kawal is a dried fermentation product of sicklepod (*Cassia obtusifolia*) leaves. The people of Sudan and Chad use kawal as a meat substitute. In this study, kawal was obtained in powder form from Forbranga, Darfour. The objective of this study was identification the chemical compositions of kawal. FTIR spectrum and atomic absorption spectrophotometer methods were used. In FTIR experimental procedure, the sample was used as a dried powder. While for the atomic absorption and inductively coupled plasma, the sample was used after combustion for extracting the minerals (K, P, Na, Mg, Ca, Zn and Fe). The investigations with FTIR showed that the fermented leaves of *C. obtusifolia* (Kawal) consisted of several types of molecular functional groups: alkyl halide, alkene, a nitro functional group, aromatic, carbonyl, alcohol, esters, ether, and acid and amides. Mineral contents of the samples were identified. About 2.50% is the presence of calcium, with the other minerals as Mg 0.66%, P 0.393%, Na 0.165% and K about 1.595%. Moreover, for extracting the iron (Fe) and zinc (Zn) minerals of fermented cassia leaves, the atomic absorption spectrophotometer was used and found that Zn was 0.665mg/L and Fe consist of the value 15.6411mg/L.

Keywords: *Cassia obtusifolia*, fermented leaves, spectroscopic

INTRODUCTION

Cassia obtusifolia (Leguminosae), also known as sicklepod, is a plant that grows wild in Africa. It is found in wastelands in the rainy season. The people from western Sudan and eastern Chad benefit from the plant, due to its leaves can be fermented to become protein-rich food. This food is known as kawal. Considered as an alternative source of meat or meat extender, kawal has had a significant role in providing protein during famine years. It has probably protected many children against kwashiorkor. However, because of its pungent, fetid odor that lingers on the fingers for hours, kawal has been refused by the elite who consider it unfit for modern social life (Algadi and Yousif 2015).

Study of the nutritional quality of *C. obtusifolia* leaves and kawal (their chemical composition, protein fractions, in vitro protein digestibility, amino acid contents, mineral content, phytic acid, tannin and total polyphenol of cassia leaves) revealed that fermentation in the kawal processing has resulted in the increase of in vitro protein digestibility, fat, protein and ash content and a decrease in fiber content. (Algadi and Yousif, 2015; Algadi, 2008). Moreover, previous studies by Nuha et al. (2010) also indicated the changes in the chemical composition, anti-nutrients, and extractable minerals among the green leaves, kawal and cooked leaves of *Cassia*. Their samples were as a dry powder. They found that the protein and ash contents increased after cooked, the total energy of both samples were decreased after fermentation process and cooked. Furthermore, anti-nutritional factors (tannin, phytate, total polyphenols) also

decreased in the treated samples, but there is an increase in the total major minerals. Total polyphenols by using spectrophotometer method by Price and Bulter (1977), minerals composition by Chapman and Pratt (1982) and HCl extractability by using Chauhan and Mahjan (1988).

Mbaiguinam et al. (2005) studied the chemical components of kawal and found aliphatic acids, and they identified as hexanoic acid (27%), butyric acid (10.4%), and valeric acid (6.3%) by using laser amounts of p-ethylphenol and p-methyl phenol. They also found a higher portion of protein fraction of the crude leaves (20.2%), compared to fermented leaves (kawal) (12.9%). They used dried samples for gas chromatography and gas chromatography-mass spectrometry (GC/MS), to analyze volatile compounds. For amino acid analysis, the kawal and leaves were hydrolyzed under nitrogen and HCl vapor by using picotag work, followed by chromatography for Amino acid identification. The objectives of this study were spectroscopic studies of the dry fermented *C. obtusifolia* leaves.

MATERIALS AND METHODS

Materials

The fermented leaves of *Cassia obtusifolia*, or kawal, were obtained from Forbranga (western Sudan). They were obtained in dry form. After being sun-dried and freed from foreign materials, they were powdered by hummer mill with the same mesh size. They were then kept in clean bottles at room temperature for further use (Figure 1).



Figure 1. Sun-dried powder of kawal (*Cassia obtusifolia*).

Methods

Samples were analyzed at room temperature. To identify the presence of certain functional groups in the sample, IR spectrum was recorded on Fourier transformer spectrometer FTIR-8400 spectrophotometer (Shimadzu, Kyoto, Japan). For further analysis, the atomic absorption spectrophotometer (model AA-6800, Shimadzu) was used to identify the mineral content (Ca, Na, K, Mg, P, Zn and Fe) composition of the sample.

FTIR analysis

In this experiment, 100mg from potassium bromide (KBr) was used additionally to 1 mg sample of the dry powder kawal. Furthermore, KBR and the sample were mixed until it became a mixture. The mixture samples were compressed and afterwards put on the piston. They were then applied to the Fourier transformer spectrometer device for detection of mineral contents of fermented cassia leaves.

Digestion method

Ca, Na, K, Mg and P digestion method was applied to prepare the sample for spectroscopy analysis. The sample was digested in $\text{HNO}_3/\text{HCl}_4$, and elements were detected by inductively coupled plasma spectroscopy (ICPS). Heater, test tube, and flasks were used as apparatus. 2g of the sample was weighed into test tubes, 8 mL HNO_3 was added to the test tubes and covered them with glass ball and left it to stand for overnight. Another HNO_3 produced fumes and was added as did it in the chamber, 3 mL of HClO_4 acid was added and heated slowly until almost dry. Moreover, the glass ball was removed and heated strongly until white fumes disappeared. The tubes were cooled and 5 mL of 0.1N HCl was added and boiled gently for 30 minutes after leaving it to cool. The solution was then transferred into the 50 mL flask. The tubes were rinsed with 10 mL of 0.1N HCl and boiled gently for 10 minutes, the solution was

poured into the previously flask and filled it to 50 mL mark with 0.1N HCl. The ICP readings were then recorded.

Atomic Spectroscopic Method

For analyzing iron and zinc, the atomic spectroscopy method was used after combusting the sample. The optic parameters were set in the following specification: lamp current low (Am) (12 and 8), wavelength (nm) 248.3 and 213.9, slit width (nm) (0.2 and 0.5) and lamp mode BGC-D2. The atomizer/gas-flow was set up as follows: fuel gas flow rate (L/min) (2.2 and 2), flame type Air-C₂H₂, burner height (mm) (9 and 7), burner lateral pos (pulse) 0 and burner angle (degree) 0. and measurements parameters are: order first, zero intercept no, unit mg/L, repetition sequence SM-M-M-..., pre-spray time (sec) 3, integration time (sec) 5 and response time 1.

RESULTS AND DISCUSSIONS

FTIR spectrum of fermented *Cassia obtusifolia* leaves revealed several regions of frequencies that can lead to identifying of molecule group. Chemical compositions of fermented kawal are shown in Figure 2. Kawal was found to consist of several types of molecules, with three functional groups. On the alkyl-halide functional group was found C-Br which has strong intensity vibration stretch, C-Cl with strong intensity of stretching moves, and C-F molecular with the same prescriptions. While the alkene functional group consist of C-H with bending strong mode of vibration, the two-C-H alkenes have bending mode of vibration. However, their intensity is variable as the three C=C alkenes have variable intensity with stretch motion and the two C-H with the strong stretch mode of vibration.

The absorption frequencies also identified N-O which belong to the nitro functional group with stretch vibration mode and strong intensity. The C=C is an aromatic molecule, stretch mode and its intensity is medium weak or multiple bands. C=O is carbonyl with strong stretch. O-H is alcohol structure which has stretch, H-bonded vibration and strong broad intensity. The two C-O are in the ester function group, with stretch vibration mode and two bonds or more intensity. The ether C-O with the same descriptions of the ester. The acid C-O stretch strong intensity of vibration. Lastly, the two N-H amides have bending modes of vibrations and their intensity are medium (Silverstein et al. 2014).

The minerals (calcium, magnesium, phosphorus, sodium, and potassium) in kawal is reported in Table 1. Calcium and potassium have relatively high proportions compared to other minerals. About 2.50% was found as Calcium and the other minerals, magnesium 0.66%, phosphorus 0.396%, sodium 0.165% and potassium about 1.595%. These results indicat the fermented leaves of *C. obtusifolia* contain the major elements needed by the human body (Table 1).

Table 1. Mineral elements contents of kawal.

The element	Amount of kawal
Calcium	2.50%
Magnesium	0.66%
Phosphorus	0.396%
Sodium	0.165%
Potassium	1.595%

Table 2. Content of Zn and Fe of sample Mg/L.

Mineral	Content (mg/L)
Zn	0.6655
Fe	15.6411

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