Partial phytochemical characterization of the fruits of *Saba senegalensis* (Apocynacaea) and *Landolphia heudolotii* (Apocynaceae) .

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ABSTRACT

The wild tree species define themselves as species pushing to the wild state. These species have a non negligible contribution in the alimentation in farming environment. The investigations led in three regions of Burkina allowed to list about forty species used as ailments Among these, two species were selected for chemical analysis: *Saba senegalensi* (Apocynaceae) and *Landolphia heudeolotii* (Apocynaceae).

Their phytonutrients and fat soluble vitamins. were determined.

Carotenoïds, anthraquinones, sterols, triperpenes as phytonutrients, and the β-carotene (provitamin A) = 1559 µg/100g, the vitamin K_1 = 889 µg/100g as fat soluble vitamins were found in the fruits of *S. Senegalensis*

While carotenoïds, anthraquinones tannins, coumarins, sterols, triterpenes as phytonutrients, and β-carotene = 275 µg/100g., the vitamin E = 4 603 µg/100g, the vitamins K_1 = 195 µg/100g and the vitamin K_3 = 304 µg/100g as fat soluble vitamins were found in the fruits of *L. heudeolotii*..

Key words: phytonutrients, vitamins, metabolic diseases, *S. senegalensis L. heudolotii*

INTRODUCTION

In Burkina Faso (West-Africa), as in a lot of tropical countries, the wild plants foods play an important role in the diets of local residents. Often consumed at the natural state, they contribute to improve the quality of the food through the micronutrients that they contain. The ignorance of their nutritional potential, or even their therapeutic effects, is a limitation to their consumption.

Previous studies have documented the nutritional contents of wide range of types of plants foods that are eaten in the republic of Niger (West Africa) (Humphry,1993; Kim,1997; Glew ,1997; VanderJagt,2000; Cook,2000). Fruits are important sources of minerals, fiber and vitamins, and they provide essential nutrients for the human health. In addition, it is known that some fruits have the so-called 'anti nutritional' factors (e.g. phytic acid, tannins) that can diminish the nutrient bioavailability, especially if they are present at high levels (Spiller,1993; Czajka-Narins,1995; Badui-Dergal,1999; Preet,2000). Nevertheless, it has been reported that these anti-nutritional factors could help prevent and treat several important diseases; remarkably, the anti-carcinogenic activity of phytic acid has been demonstrated by *in vitro* and *in vivo* assays (Lachance,1997; Shamsuddin,2002).

The results of these research showed that many of them had considerable nutritional attributes for human. Much of these research deals with the minerals components, proteins lipids and glucids. The nutrient information reported in this study should enhance efforts to promote the wider use of wild plant food.

The aims of this study is to characterize and to value the content in fat vitamins, mineral compound and phytonutrients of two wild tree : *Saba senegalensis* (Apocynaceae) and *Landolphia heudeelotii* (Apocynaceae) fruits. *S. senegalensis* fruit juice is widely consumed whereas it is not the case of *L. heudolotii* fruit juice.

A better knowledge of the chemical composition and the toxicity doorstep of these fruits will help to remedy the problems of people often victims of the malnutrition.

MATERIALS AND METHOD

Materials

The mature fruits of *Saba senegalensis* and *Landolphia heudeelotii* have been collected in the studies zones from January to June 2008 and have been kept to 4°C in view of the different analyses.

The material of analysis: TLC-densitometer (Shimadzu Dual-wavelength TLC Scanner CS-930) with a variable wavelength UV-visible detector.
For determination of mineral contents, Atomic absorption spectroscopy was used.

Chemical

For the chemical screening, the used standards were: α.-amyrin, β-sitosterol, provide by ExtraSynthèse (France). Quercetin, cyanindin and benzoquinone are Merck products (Germany). The reagents of characterizations are also Merck products (Germany). The solvents for extraction and TLC are chromatographic grade from Fluka Chemie products (Switzerland).
For the assessment of the contents in fat-soluble vitamins (β-carotene, E and K), the standards were: trans-retinol, α-tocophérol, phylloquinone (K_1) and the menadione (K_3) provided by Fluka Chemie (Switzerland). HPTLC plates 20x20 cm; RP-18 RP-18 F_{254s} from Merck.

Sample preparation

For the chemical screening, juices were extracted from these two wild tree fruits, according to Ciulei (1982) and H. Wagner methods (Hildebert,1996).
For the measurement of the proteins, the Kjeldahl method was used.
For the measurement of total minerals contents, the method of the incineration at 550° C in an oven Heraeus Hanau KR model 260 E, according to the NF norm V 03-760: 1981, was used. All the mineral elements have been quantified using atomic absorption spectroscopy.

For the measurement of fat soluble vitamins, juices were extracted according to Folch and Torre methods (Folch,1957; Torre,2001).
To 0.250 g of juice, 4 ml of acetone is added. Extraction of vitamin E and K was performed by probe sonication for 3 min. the suspension obtained is centrifuged at 2000 g for 3 min. the supernatant were filtered through a 0.45 µm pore size nylon filter and the kept for TLC-densitometer analyse.
For β-carotene extraction, the carotenoids was extract after the preliminary saponification of the sample by adding 20 ml of KOH solution (28g in 100 ml of ethanol/water mixture (9:1 v/v) on 1 g of juice and then refluxing it on a water bath at 70°C for 30 min. the carotenoids are then extracted by using hexane of chromatographic grade. After triple extraction, the hexane extracts are combined and dried over anhydrous sodium sulphate. The

solvent was then evaporated in a rotary evaporator under vacuum (temperature no higher than 40°C). The dry residue was dissolve in methanol for TLC-densitometer analyse.

Standard solutions

Calibration standard samples containing:

- (5, 4, 3, 2, and 1µg/ml) of α- tocopherol as vitamin E,
- (5, 4, 3, 2, and 1 µg /ml) of β-carotene
- (5, 4, 3, 2, and 1µg /ml) of phylloquinone as vitamin K_1
- (5, 4, 3, 2 and 1µg/ml) of menadione as vitamin K_3.

Measurement of fat soluble vitamins

20 µl of each concentration of the vitamin standard solutions were spotted on HPTLC plates and develop using mixture of acetonitrile/methanol/water (12: 12: 1 v/v/v) as mobile phase. The spots areas are measuring by setting the wavelength:

- at λ = 295 nm for vitamin E,
- at λ = 245 nm for vitamins K,
- at λ = 449 nm for β-carotene.

RESULTS AND DISCUSSION

The results of the chemical screening, presented in table I, show that *S. senegalensis* contain: carotenoids, anthraquinones, sterols and triterpenes. Whereas *L. Heudolotii* fruits contain: carotenoids, flavonoïds, tannins and anthraquinones. The alkaloids and cardiac glycosids are not found in these fruits.

These chemical groups, gifted of pharmacological properties and present in these studied edible fruits, can be beneficial for the man in the treatment or the prevention of some pathologies.

Indeed, the sterols and triterpenes are gifted of anti-inflammatory activity (Bruneton, 1993). Bolleddula(2006) showed the anti hyperglycemias and antihypercholesterolemias activities of the anthocyanosids of the fruits of *Cornus mas*, Anacardiaceae. Cichewicz (2002) showed the anti parasitize activity of some anthraquinones on the *S. mansoni cercariae*. According to Bruneton (1993), tannins and flavonoïds have some antibacterial and antihypertensive activities. These results demonstrated therapeutic or diet potential properties of fruits of these species.

For the fat soluble vitamins determination, calibration standards curves were established. The linearity of the curve obtained by plotting each regression equations (fig. 1), confirmed the precision of this assay. The correlation coefficient ($r^2 = 0.9934$ for vitamin E, $r^2 = 0.9964$ for β–carotene and $r^2 = 0.9834$ for vitamin K) value gotten with the different vitamins took like reference, show that the results of the fat-soluble vitamins E, K and β-carotene are reliable.

Landolphia heudolotii fruits contain more vitamins (E, K_3, K_1 and β-carotene) than *S. senegalensis* (β-carotene and K_1). One notes that the fruits of the *Saba senegalensis* are a real source of provitamin A (β -carotene = 1559 µ g/g). According to Vanisha, 6 µg of β-carotene equal 1 µg of retinol (vitamin A) (Vanisha,2008). Vitamin A (retinol) solves the problems of blindness and β -carotene would play an important role in the prevention of the cancer (Kennth,1985; Krinsky,1989).

According to Ball (1998), vitamin E would have some effects against barrenness and the vitamin K would have some anti hemorrhagic properties. These results allow to appreciate the nutritional potential of these wild plants food in the treatment of some deficiency diseases.

CONCLUSION

All these data on the pharmacological properties of these chemical substances identified in these fruits, showed that these wild tree fruits are worthy of interest. The fruits of these studied species could be counselled in the diet.

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Table I: phytonutrients in *S senegalensis and L.heudolotii* fruits**.**
Legend: presence (+), absence (-) , trace (+/-)

Phytonutrients	*Saba senegalensis*	*Landolphia heudolotii*
carotenoids	+	+
alcaloids	-	-
flavonoids	+	+
Coumarines	-	-/+
anthocyans	+	+
tanins	-	+
anthraquinones	-	+
saponosids	-	-
Sterols - triterpenes	+	+
Cardiac glycosids	-	-
fibber	+	+

Table 2 : Contents in fat-soluble vitamins of *S senegalensis and L.heudolotii.*

Name	Organs	Vitamins (µg/100g)
Saba senegalensis(A. DC.) Pichon (Apocynaceae)	Juice of the fruit	β-carotene = 1559 vit K_1 = 889
Landolphia heudolotii A. DC. ***(***Apocynaceae***)***	Juice of the fruit	β-carotene = 275 vit K_1 = 195 Vit K_3 = 304 Vit E = 4 603

Figure 1**:** curve of standardization of the E vitamin