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# Phytochemical Investigation of Three Medicinal Plants Grown as Leafy Vegetables in Burkina Faso

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**Abstract** The aim of this study was to investigate the phytochemical composition and antioxidant activity of three medicinal plants whose leaves are used as vegetables in Burkina Faso: *Adansonia digitata*, *Moringa oleifera*, and *Senna obtusifolia*. The leaf powder was extracted by decoction to obtain aqueous and hydroethanolic extracts. Analyses were performed to identify the major compounds of biological interest present in each plant extract by thin layer chromatography (TLC). Total phenolics and flavonoids were determined by the Folin-Ciocalteu and $AlCl_3$ methods, respectively. The antioxidant activity was evaluated using the DPPH method. Phytochemical screening revealed the presence of chemical groups of interest in the extracts, namely tannins, flavonoids and saponosides. *Senna obtusifolia* had the highest total phenolic content with 169.31 ± 0.28 mg EAG/gES. The highest total flavonoid content was obtained with the hydroethanol extract of *Adansonia digitata* (79.72 ± 3.33 mg EQ/gES). The best antioxidant power was obtained with the hydroethanol extract of *Moringa oleifera*, with an IC50 of 13.75 ± 1.95 µg/mL, indicating its potential to neutralize free radicals. These results underscore the importance of these leafy vegetables as potential sources of phytocompounds of therapeutic interest and natural antioxidants.

**Keywords** Phytochemical study, *Adansonia digitata*, *Moringa oleifera*, *Senna obtusifolia*

## 1. Introduction

The use of natural plant resources for nutritional and therapeutic purposes is an ancestral practice deeply rooted in the cultural traditions of many parts of the world, particularly in sub-Saharan Africa. In Burkina Faso, a Sahelian country in West Africa, the local population has developed a rich empirical knowledge of the indigenous flora and has mobilized its benefits to meet their nutritional and health needs for generations.

A preliminary study carried out in the province of Oubritenga, located in the Central Plateau region of Burkina Faso, identified twenty-five (25) drought-adapted local plants with both nutritional and therapeutic virtues and high utility values [1]. Among these traditionally used plants, three species stand out for their exceptional nutritional and medicinal virtues: *Adansonia digitata*, *Moringa oleifera*, and *Senna obtusifolia*, called Tohega, Arzan tiiga, and Sogoda, respectively, in the local language (Mooré).

*Adansonia digitata*, the emblematic tree of the Sudano-Sahelian landscape, plays a central role in the culture and traditional diet of Burkina Faso. All parts of this tree (leaves, pulp, seeds, bark) are widely used in various culinary forms: Vegetables, soft drinks, cooking oil, etc. This plant has also been recognized for centuries for its medicinal properties, especially in the treatment of fever, diarrhea and stomach upset [2].

*Moringa oleifera* is a fast-growing herbaceous plant found in many regions of Burkina Faso. All parts of the plant (leaves, flowers, pods, seeds) are consumed for their exceptional nutritional qualities. *Moringa oleifera* is used in traditional medicine for its anti-inflammatory, antimicrobial and anti-hypertensive properties. This plant, nicknamed "the miracle tree", has thus established itself as one of the most nutritious and health-promoting foods in many rural communities [3].

In traditional medicine, *Senna obtusifolia* is known for its laxative and purgative properties, as well as its beneficial effects on liver and kidney function. Although its use is less widespread than that of *Adansonia digitata* or *Moringa oleifera*, *Senna obtusifolia* remains an important plant resource for traditional health care in certain regions of the country [4].

Despite their ancestral use in food and traditional medicine in Burkina Faso, scientific knowledge on the

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phytochemical composition and biological activities of these three plants is still relatively limited, especially in the specific context of the central plateau region of Burkina Faso. This is the background of the present study, which aims to characterize the phytochemical profile and antioxidant activity of these three nutritious plants cultivated in Zitenga, in the central plateau region of Burkina Faso.

# 2. Methodology

### 2.1. Plant Material

The plant material consisted of the leaves of *Adansonia digitata*, *Moringa oleifera*, and *Senna obtusifolia* as show in Figure 1, harvested from a community garden in Zitenga (Oubritenga province in the Central Plateau region of Burkina Faso). The harvested leaves were thoroughly washed and dried in an airy room away from sunlight and dust (for 2 weeks), then ground into a powder.

### 2.2. Moisture Content

The residual moisture content of each plant powder was obtained using the thermogravimetric method, the principle of which is based on loss on drying [5]. To do this, one (1) g of weighed plant powder was placed in a tared watch glass. The assembly was placed at 105℃ for one (1) hour and thirty (30) minutes in a ventilated oven. After cooling in a desiccator, the product was weighed again. The operation was repeated until a constant weight was obtained. The residual moisture content of the plant drug was determined using the following formula:

$$\text{THR}\ (\%) = \frac{\text{Pe} - \text{Pe}'}{\text{Pe}\ (\text{g})}\ \text{x}\ 100$$

**Pe** = Plant material test sample (g);
**Pe'** = Test sample after drying (g).

### 2.3. Extraction

Extracts were prepared by aqueous and hydroethanolic decoction of *Adansonia digitata*, *Moringa oleifera* and *Senna obtusifolia* leaf powders. The aqueous and hydroethanolic decoctions were prepared according to the following procedure: A 50 g test sample of the plant powder was dispersed in 500 mL of distilled water or 500 mL of a 70/30 ethanol-water mixture, which constituted the hydroethanol solvent. The mixture was boiled for 30 min. After cooling, the mixture was filtered through a fine-mesh nylon cloth. The filtrate was centrifuged at 2000 rpm for 10 min, then dried in a ventilated oven and stored for further analysis. Extraction yield was determined by dividing the mass of dry extract obtained by one hundred (100) grams of dry plant matter test sample.

$$\mathbf{R}\ (\%) = \frac{\text{M}}{\text{PE}} X\ 100$$

R: yield (%)
M: dry extract mass (g)
PE: plant material test sample (g)

### 2.4. Phytochemical Screening

The phytochemical screening was carried out on chromatoplates (60 F254, glass support 20 x 20 cm, Fluka silica gel) according to the methods described in the literature [6]. Thin layer chromatography (TLC) was used to identify the main chemical groups such as steroidal, terpene, phenolic and alkaloidal compounds. Each dry extract was solubilized in its extraction solvent at a concentration of 10 mg/mL (10 mg in 1 mL solvent) and 5 µL was applied to the TLC plate. Chromatograms were developed over an 8 cm path in appropriate solvent systems.

Several specific reagents were used to detect these groups of compounds. Sulfuric vanillin reagent and Burchard Libermann reagent for terpenes and sterols; 5% (V/V) methanolic KOH reagent for coumarins; NEU reagent for flavonoids; FeCl3 reagent for tannins and phenolics; and sulfuric anisaldehyde reagent for saponosides.

*Moringa oleifera* *Senna obtusifolia* *Adansonia digitata*

**Figure 1.** Leafy vegetables under cultivation (September 2022, Zitenga)

# 3. Evaluation of the Content of Bioactive Compounds

### 3.1. Determination of Phenolic Compounds

Phenolic compounds were determined by the Singleton method using Folin-Ciocalteu reagent (FCR) [7]. The reaction mixture consisted of 25 µL extract at 0.1 mg/mL, 105 µL FCR 0.2 N, incubated for five minutes in the dark. To this mixture, 100 µL sodium carbonate solution (75 g/L in distilled water) was added. The mixture was incubated for one (1) hour in the dark and the absorbance was measured at a wavelength of 760 nm using a spectrophotometer against a tannic acid standard curve. Assays were performed in triplicate and results were expressed as milligrams of Equivalent Tannic Acid per gram of dry extract (mg ETA/g). The total phenolic content of the extract was evaluated according to the formula:

$$T = \frac{c \times D}{Ci} X\ 100$$

T = Content in mg Equivalent Tanic Acid per gram of extract

C = sample concentration read (µg ETA/mL) on standard curve

D = Dilution factor of the sample to be assayed

Ci = initial concentration of sample solution to be assayed

### 3.2. Total Flavonoids Assay

The flavonoid assay was performed according to the method of Kumaran [8], adapted by Abdel-Hamed [9]. To 100 µL of concentrated extract (1 mg/mL), 100 µL of aluminum trichloride (2% in methanol) was added. The absorbance was read at λ = 415 nm after 40 min incubation against a blank (100 µL methanol and 100 µL $AlCl_3$). The appearance of a stable yellow color allows the evaluation of the flavonoid content of the sample by UV spectrophotometry using a BioRad spectrophotometer (model 680 Japan) in comparison with a reference solution of quercetin (0-70 µg/mL). Assays were performed in triplicate and results were expressed as milligrams of quercetin equivalent per gram of dry extract (mg EQ/g). The flavonoid content of the extract was calculated using the formula:

$$T = \frac{c \times D}{Ci} X\ 100$$

T = Content in mg Equivalent Quercetin in 100 g extract

C = sample concentration read (mg EQ/mL) on standard curve

D = Dilution factor of the sample to be assayed

Ci = initial concentration of sample solution to be assayed

### 3.3. Evaluation of Antioxidant Activity Using the DPPH Method

The DPPH (2,2-Diphenyl-picrylhydrazine) method for measuring antioxidant activity is based on the ability of a compound to reduce the DPPH° radical (Figure 2). The reduction results in a color change of the solution from purple to yellow in the presence of an anti-radical compound. The reaction is then quantified by measuring the absorbance of the solution spectrophotometrically at a wavelength of 490 nm. The color change from purple to yellow is proportional to the antioxidant power.

The ability of the extracts to reduce DPPH free radicals was determined by the method of KIM et al [10]. A series of concentrations was generated from the initial concentration (1 mg/mL) of the samples using Trolox as a reference substance. On a 96-well microplate, each concentration was filled with 200 µL DPPH solution (0.04 mg/mL) and 20 µL diluted extract or reference. After incubation for 30 minutes, the absorbance was read at a wavelength of λ = 490 nm using a Bio-Rad spectrophotometer (model 680 Japan). The blank (without sample) was prepared under the same conditions and consisted of 200 µL DPPH and 20 µL ethanol. A curve of percent DPPH inhibition was plotted as a function of sample concentration (Table 1). The concentration required to inhibit 50% of DPPH (IC50) was determined from the curve.

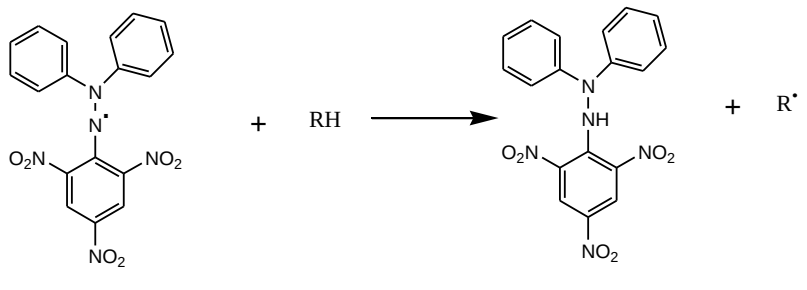
# 4. Results and Discussion

### 4.1. Moisture Content

Moisture content is a crucial factor to take into account when storing and processing plant raw materials. The results are shown in the following Table 1.

**Table 1.** Residual moisture content

| | *Adansonia digitata* | *Moringa oleifera* | *Senna obtusifolia* |
|---|---|---|---|
| **THR (%)** | 7.89 ± 0.19 | 7.20 ± 0.16 | 7.46 ± 0.57 |




DPPH· (purple) DPPH (yellow)

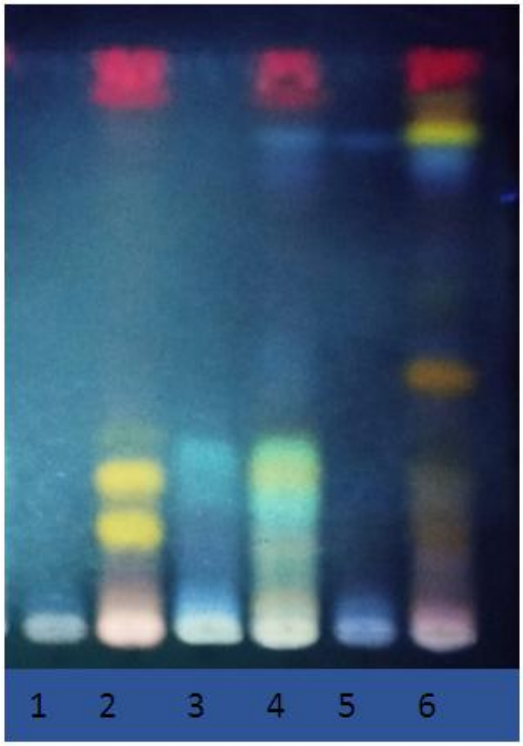
**Figure 2.** Reduction of DPPH by an antioxidant

Residual humidity levels (THR) are all below 10%. Factors such as humidity, heat, radiation, microorganisms and enzymes are often the cause of plant material degradation. The permissible content in dry plant material for good preservation should not exceed 10%. The above results show that our samples can be preserved with less risk of contamination and/or alteration of the chemical principles [11]. Drying, dehydration and relative humidity control techniques are therefore essential for maintaining optimum moisture levels in plant raw materials throughout the value chain.

### 4.2. Extraction Yield

The various plant material extraction operations gave variable yields, which are recorded in the following Table 2.

Extraction yields ranged from 8.25% to 33.52%. The highest extraction yield was obtained with the aqueous extract of *Senna obtusifolia;* while the lowest yield was obtained with the aqueous extract of *Adansonia digitata*. In terms of extraction solvents, water was the best for *Moringa oleifera* and *Senna obtusifolia*. This high yield could be explained by the hydrophilic nature of the constituents present in plant extracts. Water, the extraction solvent, carries polar compounds away. Thus, the more hydrophilic a molecule, the more easily it will pass into a decoction and/or maceration [12]. For *Adansonia digitata*, ethanol was the best extraction solvent. Water-alcohol mixtures are also known to be good extraction solvents for polar compounds [13].




*1. Adansonia digitata, aqueous extract*
*2. Adansonia digitata ethanolic extract*
*3. Moringa oleifera aqueous extract*
*4. Moringa oleifera ethanolic extract*
*5. Senna obtusifolia aqueous extract*
*6. Senna obtusifolia ethanolic* extract

**Figure 3.** TLC to detect secondary metabolites

**Table 2.** Extraction yields

| | ***Adansonia digitata*** | | ***Moringa oléifera*** | | ***Senna obtusifolia*** | |
|---|---|---|---|---|---|---|
| | Aqueous | Hydroethanolic | Aqueous | Hydroethanolic | Aqueous | Hydroethanolic |
| **Yield (%)** | **8.25** | **14.52** | **33.18** | **20.82** | **33.52** | **19.36** |

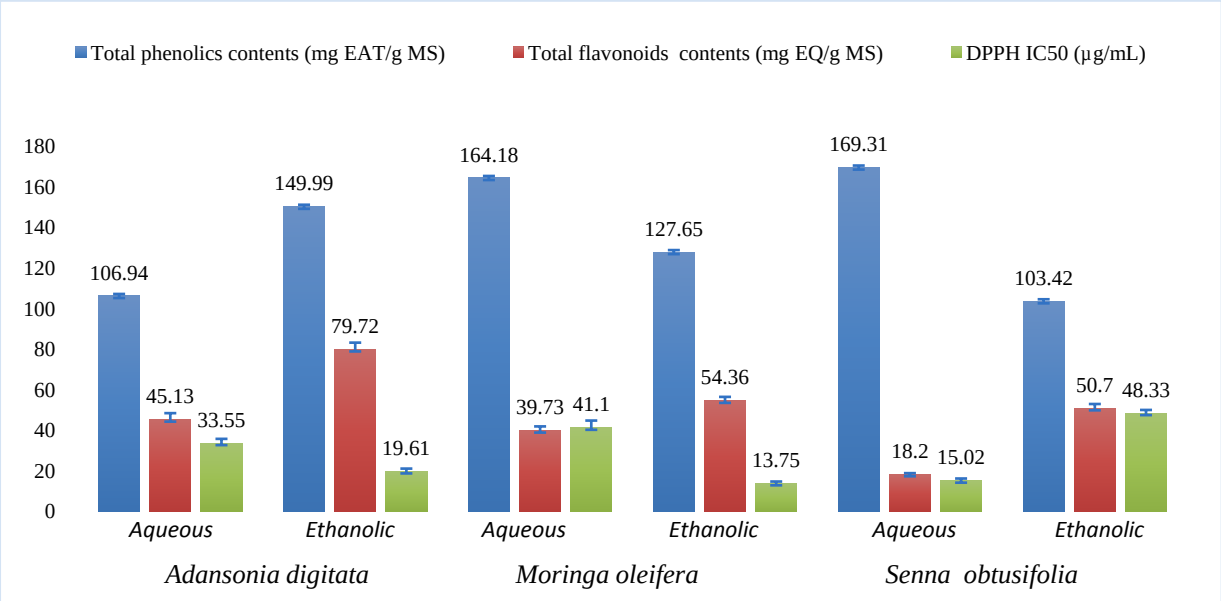



**Figure 4.** Contents of total phenolics, total flavonoids and antioxidant activity

### 4.3. Chemical Groups

The various fractions were characterized by TLC in order to highlight the chemical groups of interest. The results obtained are illustrated by thin layer chromatography (TLC) and reported in the following Figure 3.

Phytochemical screening revealed the presence of chemical groups of interest in the extracts, namely tannins, flavonoids and saponosides. These chemical groups are endowed with several biological properties, notably their antioxidant capacity [14]. Fingerprinting shows that all three plants contain numerous chemical groups. Different combinations of these plants in cooking could be a great source of secondary metabolites for our bodies.

### 4.4. Bioactive Compound Content and Antioxidant Activity

Total phenolic content, total flavonoid content and antioxidant activity were determined successively with aqueous and hydroethanolic decoctions. The results obtained are presented in histogram form in the following Figure 4.

The total phenolic content of the extracts ranged from 103.42 ± 3.9 mg EAT/g to 169.31 ± 0.28 mg EAT/g dry matter. The highest content was obtained with *the aqueous extract* of *Senna obtusifolia* at 169.31 ± 0.28 mg ETA/g DM, and the lowest with the aqueous extract of *Senna obtusifolia* at 103.42 ± 3.9 mg ETA/g. With the exception of *Adansonia digitata*, the highest levels of total phenolics were obtained with aqueous extracts. Water is a good solvent for extracting phenolic compounds, as demonstrated in the literature by several authors [15,16]. Total flavonoid content ranged from 18.2 ± 0.54 mg QE/g to 79.72 ± 3.33 mg QE/g dry matter. The highest content was obtained with the hydroethanol extract of *Adansonia digitata* (79.72 ± 3.33 mg EQ/g DM), and the lowest with the aqueous extract of *Senna obtusifolia* (18.2 ± 0.54 mg EQ/g). For all extracts, we note that the highest contents are obtained with hydroethanolic extracts. It should be noted that the literature shows that water-ethanol or water-alcohol solvent mixtures are good extraction solvents for flavonoids [17]. High levels of total phenolics and flavonoids are thus obtained both with water as extraction solvent, and with a water-ethanol mixture. The use of water in the cooking of these plants is appreciable, enabling the body to benefit from considerable quantities of phytonutrients. Antioxidants protect the body against the damaging action of free radicals. Some antioxidants are produced by our own bodies, while others, such as vitamins C, E and ß-carotene, are ingested. The DPPH method was used in our work. The 50% inhibitory concentration ($IC_{50}$), is the concentration of the extract likely to cause 50% inhibition of the DPPH radical, was determined and recorded in the Figure 4. From these analyses, the most active extract is the hydroethanol extract of *Moringa oleifera* with an $IC_{50}$ of 13.75 ± 0.86 µg/mL. The lowest antioxidant activity was obtained with the hydroethanol extract of *Senna obtusifolia*, with an $IC_{50}$ of 48.33 ± 1.55 µg/mL. Generally speaking, with the exception of *Senna obtisufolia*, hydroethanol extracts appear to be more active than aqueous extracts. Previous studies have shown a correlation between the presence of phenolic compounds in an extract and its antioxidant activity [18,19]. Indeed, phenolic compounds, notably flavonoids, are recognized as powerful antioxidants against free radicals, due to their property of donating available hydrogen atoms in the hydroxyl substituents of their phenolic groups [20]. Their protective effects in biological systems are linked to their ability to transfer electrons to free radicals, chelate metals, activate antioxidant enzymes or inhibit oxidases [21]. Plant flavonoids are recognized for their effectiveness against fever, edema and mucous inflammation [22,23]. Regular consumption of these leafy vegetables helps combat oxidative stress and prevent certain chronic diseases [24].

## 5. Conclusions

This study assessed the phytochemical profile and antioxidant activity of three leafy vegetables, *Adansonia digitata*, *Moringa oleifera* and *Senna obtusifolia*. The phytochemical screening revealed the presence of chemical groups of interest in the extracts, namely tannins, flavonoids and saponosides, known for their therapeutic properties. The results obtained provide a scientific basis for the development of improved traditional medicines or nutraceuticals for the prevention of various diseases related to oxidative stress or other diet-related chronic diseases.

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