

Original Research Article

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# Antioxidant Activity and Phytochemical Composition of the Leaves of *Microdesmis keayana* J. Leonard, a Pandaceae Species Traditionally Used for Newborn Health Care in Man, Western Côte d'Ivoire

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## ABSTRACT

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This study contributes to the promotion of medicinal plants from the neonatal health pharmacopeia. In this context, we evaluated the antioxidant activity of aqueous and hydro-ethanolic extracts prepared from the leaves of *Microdesmis keayana*, a plant used in the city of Man in western Côte d'Ivoire to strengthen newborns' immune systems. Screening for radical scavenging activity on thin layer chromatography (TLC) revealed that the extracts have antioxidant activity. Spectrophotometer measurement of the extracts' DPPH scavenging power showed that the hydro-ethanolic and aqueous extracts of *Microdesmis keayana* are highly active, with IC<sub>50</sub> values of about 0.014 and 0.025 mg/ml, respectively. These results were confirmed by assay tests, which showed that the *Microdesmis keayana* extracts contained total polyphenol contents of 433.18 and 412.03 mg GAE/gE for the hydro-ethanolic and aqueous extracts, respectively and in condensed tannins (207.83 and 203.66 mgEC/gE in the same order) as well as a significant total flavonoid content of about 46 mgEQ/gE. Thus, the considerable antioxidant activity of this species is attributed to its richness in these phytochemicals.

## Introduction

General disorders in newborns are caused by various factors, including oxidative stress, which is when the cell

can no longer control the excessive presence of toxic oxygen radicals (Pincemail *et al.*, 1999). The use of synthetic antioxidant molecules is currently being questioned because of potential toxicological risks,

which can be very harmful to neonatal health. Nowadays, new plant sources of natural antioxidants are being sought (Tadhani *et al.*, 2007). So after giving birth, many mothers systematically turn to herbal medicine. This use of medicinal plants is due to the natural substances they contain. As a result, many natural antioxidants are already being commercially exploited, and their likely role in preventing diseases related to oxidative stress has been proven around the world. The Ivorian flora, due to its rich plant diversity (Yao, 2013), is not left out of this research. However, such studies are rare in the city of Man.

In an effort to contribute to the promotion of medicinal plants for neonatal health in the Mountain District, *Microdesmis keayana* J. Leonard, a plant used in traditional medicine in the city of Man to strengthen newborns' immune systems, was chosen. It is a short dioecious shrub about 6 m tall and around 8 cm in diameter. *M. keayana* is a plant species from the Pandaceae family that is quite widespread in tropical and subtropical African regions, notably in Ghana, the Republic of Congo, Ivory Coast, Nigeria, Sierra Leone, Burundi, Gabon, and Rwanda. In Ivory Coast, it is only found in the forest zone (Abidjan, Agboville, Dabou, Man) where it is used as an aphrodisiac and to treat rheumatism (Okeke *et al.*, 2023). It was selected based on its frequent use in traditional medicine, its effectiveness in treating neonatal diseases, the lack of scientific data, and its availability in the Man Department. This study is to evaluate the antioxidant activity of aqueous and hydro-ethanolic extracts prepared from the leaves of *Microdesmis keayana* then look for the plant compounds involved in this antioxidant activity and measure them.

## Plant Material and Methods

### Plant Material

The plant material consists of leaves of *Microdesmis keayana* (Figure 1), collected in March 2024 in the village of Glayogouin, in the Man Department. The leaves were washed with tap water and dried under constant air conditioning at 18 °C for two weeks, then ground. The resulting powder was then stored in a tightly sealed glass container at room temperature. The plant species was identified at the National Center of Floristics at Félix Houphouët Boigny University, Abidjan (Côte d'Ivoire).

## Methods

### Extraction

For the preparation of plant extracts, two methods were used: decoction according to traditional preparations for aqueous extracts, and maceration for hydro-ethanolic extracts.

### Aqueous extraction

25 g of plant powder were mixed with 250 mL of distilled water, then the mixture was brought to a boil for 30 minutes with mechanical stirring. The resulting solution was then filtered under vacuum using a funnel and Whatman No. 1 filter paper. The obtained extract was subsequently dried in an oven at 50 °C for 24 hours. The resulting dry extract was weighed and the extraction yield was calculated.

### Hydro-ethanolic extraction

To 25 g of plant powder, 250 mL of a 70% ethanol solution was added, and the mixture was then left to macerate under mechanical stirring for 24 hours. The macerate was vacuum filtered using a funnel and Whatman No. 1 filter paper. The filtrate was slowly dried in an oven at 40 °C for one day. The resulting dry extract was weighed, and the extraction yield was calculated.

### Determination of the antioxidant activity of the extracts

The antioxidant activity was evaluated by thin-layer chromatography (TLC) and by spectrophotometry according to Blois' method (1958) with slight modifications.

The percentage of DPPH reduction was calculated using the equation:

$$\% \text{ DPPH inhibition (PI)} = [1 - (\text{Extract Absorbance} / \text{Control Absorbance})] \times 100 \quad (2)$$

### Search for Major Groups of Chemical Compounds by TLC

To characterize the groups of chemical compounds present in the extracts, thin-layer chromatography and assays were used to quantify phytochemicals.

- Total polyphenol contents were determined using the Folin-Ciocalteu colorimetric method (Singleton *et al.*, 1999).
- Total flavonoid content was measured according to the method used by Arvouet-Grand *et al.*, (1994).
- Condensed tannin content in the various extracts was determined according to the method described by Heimler *et al.*, (2006).

## Results and Discussion

### Extract Yields

The calculated yields are 25.44% and 28.24%, respectively, for the aqueous extract and the hydro-ethanolic extract. The hydro-ethanolic extract had the higher extraction yield. Table.1 shows the mass and yield of the different extracts after drying.

### Evaluation of Antioxidant Activity

#### Radical Scavenging Activity by TLC

Compounds showing radical scavenging activity appear as pale yellow spots on a violet background. These results show that all tested extracts have antioxidant activity.

#### Antioxidant Power by Spectrophotometry

All tested extracts showed DPPH inhibition. This inhibition was even more pronounced as the extract concentration increased, which corresponds to a dose-dependent effect (Figure 2). The determination of IC<sub>50</sub> shows that the species *Microdesmis keayana* has good antioxidant activity, with IC<sub>50</sub> values around 0.025 mg/ml and 0.014 mg/ml for the aqueous and hydro-ethanolic extracts, respectively. Ascorbic acid or vitamin C (IC<sub>50</sub> = 0.007 mg/ml) showed higher antioxidant power than the extracts.

### Phytochemical tests

#### Phytocompounds detected by TLC

The 10% ferric chloride solution indicates the presence of tannins, identified by black spots visible in all extracts. Yellow-orange spots visible to the eye confirm the presence of anthraquinones and anthrones after spraying

with 5% potassium hydroxide in ethanol. The yellow fluorescence observed at 365 nm and in visible light in the presence of 5% aluminum chloride reveals the presence of flavonoids. Observation under UV light at 365 nm showing yellow fluorescence after spraying with 25% ammonia solution confirms the presence of coumarins. All these results are recorded in Table.2.

### Extracts Quantification

#### Total Polyphenol Contents

The results obtained show that *Microdesmis keayana* extracts are rich in total polyphenols. Its hydro-ethanolic extract is the richest, with a content of 433.18 mg GAE/gE compared to 412.03 mg GAE/gE for the aqueous extract.

#### Total Flavonoid Contents

The flavonoid quantification was done using the aluminum trichloride method, with quercetin as the standard. The results also show a difference in flavonoid content among the studied extracts (46.54 mg QE/gE for the aqueous extract and 46.96 mg QE/gE for the hydro-ethanolic extract).

#### Condensed Tannins Contents

The results of the condensed tannin quantification indicate that their contents vary depending on the extract and the species. Overall, the species *Microdesmis keayana* is the richest in condensed tannins, with levels of 207.83 mgEC/gE and 203.66 mgEC/gE for the hydro-ethanolic and aqueous extracts, respectively.

The extraction yields obtained from the leaves of *Microdesmis keayana* were 25.44% for the aqueous extract and 28.24% for the hydro-ethanolic extract. These results indicate that the yield varies depending on the extraction solvent used.

According to Dah-Nouvlessounon *et al.*, (2015), the extraction capacity of a solvent depends, on one hand, on the solvent's affinity for the phytocompounds and, on the other hand, on the polarity of this solvent. Ojatula *et al.*, (2020) reported a yield of 3.41% from the roots of *Microdesmis keayana* using ethanol as a solvent, while Zamblé *et al.*, (2006b) had a yield of 1.13% from the aqueous extract of *Microdesmis keayana* roots.

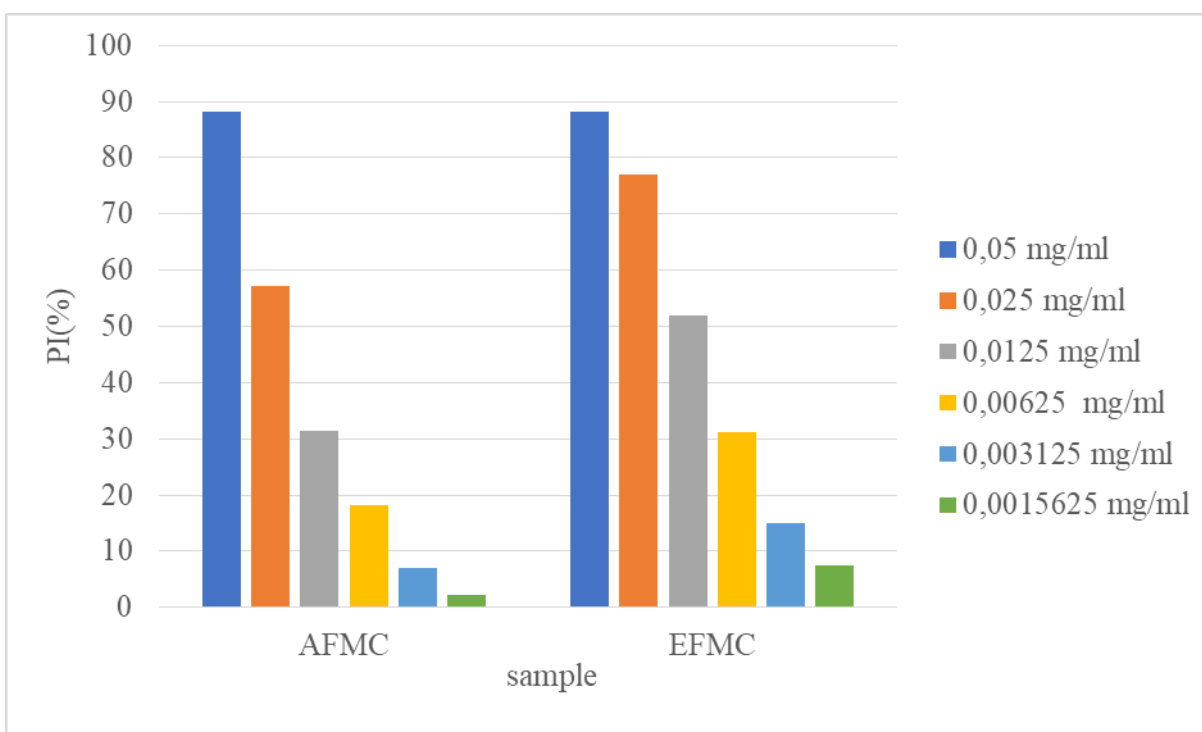
The antioxidant power of crude plant extracts from the studied plants was evaluated using the DPPH method. The antioxidant tests carried out on the two selected plants showed that they both have antioxidant properties.

The hydro-ethanolic and aqueous crude extracts of *Microdesmis keayana* showed very strong antioxidant activities, with IC<sub>50</sub> values of 0.014 mg/ml and 0.025 mg/ml, respectively.

**Figure.1** Twig of *Microdesmis keayana*



**Figure.2** Percentage of inhibition of the extracts based on concentrations



*AFMC*= Aqueous extract of leaves of *Microdesmis keayana*; *EFMC*= ethanolic extract of leaves of *Microdesmis keayana*

**Table.1** Yields of Different Crude Extracts of *Microdesmis keayana*

| Plant Species              | Type of Extract | Powder Weight (g) | Extract Weight (g) | yield(%) |
|----------------------------|-----------------|-------------------|--------------------|----------|
| <i>Microdesmis Keayana</i> | Aqueous         | 25                | 6.36               | 25.44    |
|                            | Hydro-ethanolic | 25                | 7.06               | 28.24    |

**Table.2** Phytocompounds detected in the aqueous and hydro-ethanolic extracts of *Microdesmis keayana* by TLC

| Reagents  |      | Ferric chloride 10 % |       | Potassium hydroxide 5 % |        | Aluminum chloride 5 % |         |       |        | Ammoniac 25 % |        |
|-----------|------|----------------------|-------|-------------------------|--------|-----------------------|---------|-------|--------|---------------|--------|
|           |      | Visible              |       | Visible                 |        | Visible               |         | 365nm |        | 365 nm        |        |
| Extracts  |      | Rf                   | Color | Rf                      | color  | Rf                    | couleur | Rf    | color  | Rf            | Color  |
| Mic. kea. | Aq.  | 0.09                 | black | 0.94                    | yellow | 0.95                  | yellow  |       |        | 0.08          | yellow |
|           |      | 0.36                 | black |                         |        |                       |         |       |        | 0.56          | yellow |
|           |      |                      |       |                         |        |                       |         |       |        | 0.94          | yellow |
|           | Hyd. | 0.11                 | black | 0.41                    | yellow | 0.95                  | yellow  | 0.44  | yellow | 0.14          | yellow |
|           |      | 0.46                 | black | 0.94                    | orange |                       |         |       |        | 0.55          | yellow |

Mic. kea.: *Microdesmis keayana*; Aq: Aqueous; Hyd: Hydro-ethanolic

This strong antioxidant activity of *Microdesmis keayana* leaf extracts could be responsible for its pharmacological activity, which supports its use in traditional medicine to treat oxidative stress-related diseases and improve patients' health (Zamblé *et al.*, 2006b).

The biological activities of plants are related to their chemical composition. Thus, phytochemical analysis, by thin-layer chromatography (TLC) of the aqueous and hydro-ethanolic extracts, was carried out to investigate the compounds responsible for the observed antioxidant activity. TLC was used because it appears to be more reliable than characterization tests (Boua *et al.*, 2008). Indeed, it allows a more detailed analysis of the chemical composition of the extracts under study. The results showed that the leaves of both extracts contained numerous phytochemical groups, notably anthrones and anthraquinones, tannins, coumarins, and flavonoids. These secondary metabolites could justify their use in the treatment of several pathologies. It is agreed that these chemical groups are generally involved in the biological activity of medicinal plants (Olasunkanmie *et al.*, 2017).

Besides, many authors have shown a correlation between phenolic compounds and the antioxidant activity of plants (Ouattara *et al.*, 2011). They are chemically structured to capture free radicals and, according to studies, they seem to be involved in free radical scavenging activity. We chose to measure total

polyphenols, flavonoids, and (condensed) tannins because these are the phytochemicals most cited in the literature for their ability to scavenge free radicals and reduce oxidative stress (Sidiq *et al.*, 2018).

Based on the results of thin-layer chromatography (TLC) and the analysis of the extracts, it appears that the strong antioxidant activity detected in the species *Microdesmis keayana* could be linked to the polyphenolic compounds such as flavonoids, tannins, coumarins, and anthrones and anthraquinones, which were found in both the aqueous and hydro-ethanolic extracts. These findings are supported by the work of Odesanmi *et al.*, (2012), who also detected the same compounds. Additionally, the analysis of the extracts showed high levels of total polyphenols (412.03 mg GAE/gE for the aqueous extract and 433.18 mg GAE/gE for the hydro-ethanolic extract), condensed tannins (203.66 mg EC/gE for the aqueous extract and 207.83 mg CE/gE for the hydro-ethanolic extract), and total flavonoids (46.54 mg QE/gE for the aqueous extract and 46.96 mg QE/gE for the hydro-ethanolic extract). This richness in polyphenolic compounds could be responsible for the high antioxidant activity, as shown by Bidié *et al.*, (2011), who noted that the antioxidant activities of many medicinal plants from the Ivorian pharmacopeia were linked to polyphenols, particularly flavonoids. These antioxidant compounds present in both leaf extracts of *Microdesmis keayana* explain its use in traditional plants and medicine tools in Côte d'Ivoire.

This study focused on *Microdesmis keayana*, a plant widely used in traditional medicine in the city of Man to boost the immune system of newborns. Thanks to the chemical tests carried out, we were able to establish a link between the antioxidant activity of the crude aqueous and hydro-ethanolic extracts of *Microdesmis keayana* and the presence of molecules belonging to the polyphenolic compound family. The tests revealed that all the plant extracts have notable antioxidant effects, which supports their use in traditional medicine. *Microdesmis keayana* deserves special attention because its extracts show remarkable DPPH radical scavenging activity (with IC<sub>50</sub> values of 0.014 and 0.025 mg/ml for the hydro-ethanolic and aqueous extracts, respectively) due to high levels of total polyphenols, flavonoids, and condensed tannins.

## Author Contributions

MONYN E. D. Epse KOUAME collected the data, participated in the experimental work, analyzed the results, and helped write and revise the manuscript. She approved the final version of the manuscript. MAHE Josué collected the data, participated in the experimental work, analyzed the results, and helped write and revise the manuscript and approved the final version. KOUASSI KS. took part in the experimental work, performed the statistical analyses, and approved the final version of the manuscript. KONE Mamidou Witabouna designed the study, revised the manuscript, and approved the final version.

## Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

## Declarations

**Ethical Approval** Not applicable.

**Consent to Participate** Not applicable.

**Consent to Publish** Not applicable.

**Conflict of Interest** The authors declare no competing interests.

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