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Full Length Article

Phytochemical screening, antioxidant, anti-inflammatory and antiangiogenic activities of *Lophira procera* A. Chev. (Ochnaceae) medicinal plant from Gabon

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ABSTRACT

The objective of this study was to perform phytochemical screening, to determine the content of phenolic compound, to evaluate antioxidant, anti-inflammatory and antiangiogenic activities of ethanol, water-ethanol and water extracts of *Lophira procera*. Antioxidant activity was determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) and phosphomolybdenum assay, anti-inflammatory activity by proteins denaturation inhibition and membranes stabilization test and antiangiogenic activity by chicken chorioallantoic membrane (CAM) method. The results showed that this plant is rich in saponins, polyphenols, tannins, total flavonoids, proanthocyanidins and coumarins. Extracts presented a strong antioxidant activity (IC_{50} values of 5.452 ± 0.119 µg/mL and 6.346 ± 0.544 µg/mL and respective AAI of 9.173 ± 0.203 and 7.919 ± 0.711). Excellent anti-inflammatory activity was also observed (IC_{50} = 16.952 ± 1.897 and IC_{50} = 23.172 ± 0.066 µg/mL for inhibition of protein denaturation and membrane stabilization respectively). Finally, extracts manifested a very good anti-angiogenic activity (with inhibitions ranging from 57.142 ± 0.124% to 100%). These biological activities are certainly due to high content of phenolic compound. This is the first study to report the phytochemical screening, the content of phenolic compound, the antioxidant, anti-inflammatory and antiangiogenic activities of extract derived from *Lophira procera*. The use of this plant in traditional medicine against ulcers, breast cancer, kidney and dental pain is therefore justified and its potential as a candidate for bioactive therapeutic molecule.
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1. Introduction

Lophira procera is a plant of family ochnaceaes. its vernacular name (Fang) is *Akoga* and its trade names are *Azobe* and *Bongossi*. It is a giant tree of the damp forest, one of the largest in the African virgin forest, easily recognizable by its slender barrel, fairly light brown and its large oblong, erect leaves and tufts at the ends of the branches at the top. This tree is very widespread in Gabon [1] (Fig. 1). In traditional Gabonese medicine, this plant is used in the treatment of several pathologies. The decoction of barks from this plant is used in lotions against the evil of the kidneys; by an anal route against rheumatism and lumbago, this decoction is also used against chronic gonorrhea, sterility, sexual asthenia, ulcers, rheumatoid arthritis and breast cancers. traditional therapists and phytotherapists define cancer as an accumulation of hard clods in the body and plants that reduce these clods are considered anticancer.

Cancers are among the leading causes of morbidity and mortality in the world. In 2012, there were approximately 14 million new cases and 8.2 million deaths related to the disease. The number of new cases is expected to increase by about 70% over the next two decades [2]. Cancer, or malignant tumor, is characterized by a rapid proliferation of abnormal cells which, beyond their usual delimitation, can invade adjacent parts of the organism and then swarm into other organs. It was recognized in 1941 that cancers develop from "subliminal neoplastic states" caused by viral or chemical carcinogenic agents that induce somatic changes [3,4]. Today, these states correspond to "initiation", involve DNA alterations,

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Fig. 1. *Lophira procera* A. Chev. (Ochnaceae). Synonym *Lophira alata* Banks ex CF Gaertn. *Photo taken in a forest of Douala, a village in the town Mitzic/Gabon* (Ngoua-Meye-Misso, 2016).

are irreversible and can persist in a tissue until a second type of stimulation called "promotion" occurs [5]. Several promoters directly or indirectly induce cell proliferation, recruit inflammatory cells, increase the production of reactive oxygen species (ROS) leading to oxidative damage to DNA and reduce DNA repair, resulting in replication of DNA and proliferation of cells that have lost normal growth control [6]. Studies have shown that ROS are activators of pro-angiogenic substances such as vascular endothelial growth factor (VEGF) and matrix metalloproteinases (MMP), the goal of which is to induce angiogenesis [6,7].

Angiogenesis is a process of formation of new vessels from arterial vascularization created by endothelial cells. It is essential for the continuous growth of the tumor because it supplies the tumor with nutrients and oxygen, and eliminates cellular waste, which can be toxic to cancer cells [8]. Inflammatory cells and soluble factors are present in all tumors. Signs of "burning" inflammation that include tissue remodeling, angiogenesis and other wound healing characteristics are usually used by pathologists as morphological indices of invasive cancer. Recent evidence demonstrates that these stromal processes play a fundamental role in the development and progression of cancer and, at least in some cases, can predict the clinical behavior of cancer better than the characteristics of neoplastic cells themselves [9]. There are biomolecules present in plants that can neutralize ROS [10,11], prevent inflammation and inhibit tumor angiogenesis [11,12] to finally kill the tumor cells. Also, the plants have been at the origin of many active molecules having shown their effectiveness in the treatment of different cancers, such as breast, ovary and lung treat taxol (paclitaxel) which comes from the bark of Pacific yew (*Taxus brevifolia*).

The aim of this study is to evaluate the antioxidant, anti-inflammatory and antiangiogenic activities of extracts of this plant after carrying out a phytochemical screening.

2. Materials and methods

2.1. Plant material

The stem barks of *Lophira procera* were collected 23 August 2016 in Mitzic (Woleu-Ntem, Northern of Gabon) (Fig. 1). They were identified at National Herbarium of IPHAMETRA, Libreville (Gabon). Voucher specimen has been deposited in the Herbarium of IPHAMETRA and at Laboratory of Biochemistry Research (LAREBIO), Department of Chemistry-Biochemistry, Faculty of Sciences of USTM in Franceville.

2.2. Preparation of plant extract

Barks were dried at ambient temperature of the Laboratory (20–30 °C) and protected from light for several days. After drying, barks were crushed using a grinder (Laboratory Blender, Torrington, CT. USA). This powder was used for extractions by maceration method. Briefly, 200 g of powder was mixed with 2000 mL of solvent (water, water-ethanol (50/50, v/v) and ethanol). After 72 h, the obtained extract was filtered using Whatman N 1 filter paper. Ethanol and water-ethanol extracts were concentrated under reduced pressure at rotavapor (Büchi, Labortechnik, Switzerland) at 40 and 60 °C respectively. Water extract was lyophilized using a lyophilizer (Alpha 1–2 LDplus, Germany). All crude extracts obtained were stored at 4 °C until analysis.

2.3. Phytochemical screening

Each extract was then tested for the presence of flavonoids, coumarins, tannins, total phenolic, saponosids, cardiac glycosides, reducing sugar, sterols and triterpenes, oses and holosides, anthracenics, anthocyans, alkaloids and anthracenosids as described elsewhere [13].

2.4. Total phenolic content

The Folin-Ciocalteu Method [14] with minor modifications was used to determine the total phenolic contents of the different extracts using gallic acid as standard. The absorbance was measured at 735 nm using a Spectrophotometer (Thermo Scientific, Evolution 60S, USA). Results were expressed as gallic acid equivalent per gram of lyophilized sample (average of the triplicate analysis).

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