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Evaluation of Total Polyphenol and Flavonoid Contents of Fresh and Dried Pepper (*Piper nigrum*) from the Ivory Coast

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Abstract

The growing interest in the potential biological activity of polyphenols and flavonoids has highlighted necessity to assess their content in fresh and dried pepper from Ivory Coast. The total polyphenol content was determined using the Folin-Ciocalteu test and the total flavonoid content was also measured spectrophotometrically using the aluminum chloride colorimetric test. The results obtained indicate the total polyphenol levels of fresh and dried peppers are higher in the samples collected in Danane (0.8 ± 0.05 mg GAE/100g) and in Azaguie (0.4 ± 0.08 mg GAE/100g). The lowest concentrations were obtained with fresh and dried pepper samples collected in N'douci (0.22 ± 0.02 mg GAE/100g) and in PK 103 (0.22 ± 0.02 mg GAE/100g). The highest content of total flavonoids was revealed in the locality of PK 103 (1.85 ± 0.62 mg EC/100g) followed by the locality of Maféré (1.37 ± 0.25 mg EC/100g) respectively for black pepper and green pepper. Then, the lowest flavonoid contents were obtained in the dried pepper of Lopou (0.57 ± 0.03 mg EC/100g) and the fresh pepper of PK 103 (0.47 ± 0.03 mg EC/100g). The results obtained highlight the importance of these black spices as promising sources of phenolic compounds and could be used in pharmaceutical treatments and in food.

Keywords

Content, Flavonoids, Polyphenols, *Piper nigrum*

1. Introduction

A spice is an organic material of plant origin that generally has a strong and sometimes pungent taste. Thus, it is used to flavor dishes in order to enhance their

taste. They can be derived from bark (cinnamon), leaves (tea, laurel), bulbs (garlic, onion, ginger) or seeds (fennel, coriander), fruits (dill, mustard and pepper).

Also, they have important medicinal and pharmacological properties [1]. Pepper (*Piper nigrum* L.) is one of the most known and widely used spices. It originates from Malabar, precisely in the south of India. It is mainly cultivated in India, Malaysia, Brazil, Indonesia and Sri Lanka.

Indeed, the world production was estimated at 714,296 tons in 2020 and this production was estimated at 22,342 tons in Africa. In Côte d'Ivoire, the pepper harvested quantity was estimated at 63 tons [2]. Depending on the purity degree and the processing method, different types of pepper can be obtained. Firstly, green pepper is obtained after harvesting the ripe berries. Secondly, black pepper is produced from ripe, still green, sun-dried, while white pepper are mature fruits that are stripped of their skin and then sun-dried. Thus, the pepper popularity is due to its flavor and pungent taste [3]. Various species of the genus Piper are important for their medicinal, aromatic and bioactive properties. It is considered as the king of spices due to the highest volume of its international trade [3]. Several studies have pointed out that pepper contains essential oil which is composed of terpene hydrocarbons (89% in total), oxygenated terpenes and aromatic compounds [4]. Polyphenols are secondary metabolites produced by plants. They are characterized by the presence of numerous phenolic rings [4]. The main sources of polyphenols are berries, grapes, olive oil, cocoa, nuts, peanuts and other fruits and vegetables. These fruits and vegetables contain 200 to 300 mg of polyphenols. In addition, products made from these fruits, such as tea, wine or beer, also contain significant amounts of polyphenols [5]. Next, the most important group of polyphenols is the flavonoids. These flavonoids are found in abundance in colored vegetables (spinach) and fruits such as apples, grapes, oranges, strawberries, plums, and in some foods and beverages used in human nutrition [6]. However, few studies have been performed on the chemical characterization of pepper produced in Côte d'Ivoire, especially on its antioxidant content. Thus, this study will focus on chemical characterization by first sampling the different types of pepper produced in Côte d'Ivoire. Then, determination of antioxidants in these different types of pepper will carry out.

2. Material and Methods

2.1. Plant Material

The plant material for this study consists of fresh and dried pepper. The different samples were collected in areas of high pepper production (**Figure 1**).

2.2. Methods

2.2.1. Choice of Sampling Sites

A database analysis of Association Pepper Producers in Côte d'Ivoire currently shows only 38 plantations are in production in Côte d'Ivoire. These plantations essentially are located in Azaguié, Maféré, N'douci, Guibéroua, Danané, Niablé,

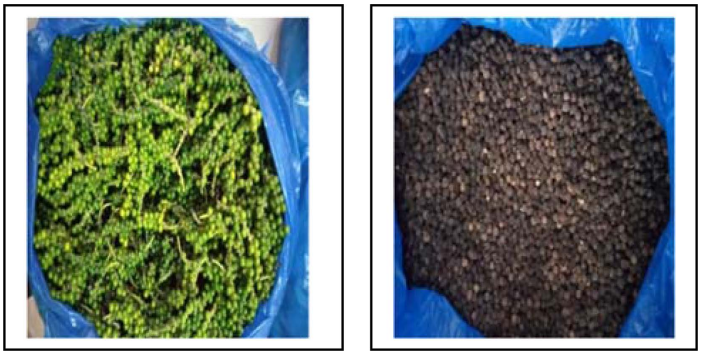

Figure 1. Fresh pepper with weeds and dried pepper without weeds.

Yakassé, Lopou, Assouba and Pk 103. Taking into account the geographical layout and the soils distribution in Côte d'Ivoire, these plantations can be grouped into 10 different entities.

2.2.2. Sampling

The different plantations selected are used for pepper sampling. In each plantation, three (3) fresh pepper samples and three (3) dried pepper samples were collected. The mass of each sample has been 1.5 kg. These three samples were divided into three (3) during the major production season (January-April) and two (2) during the minor season (July-August). The sampling was carried out for two (2) years. Finally, each sample was used to determine total polyphenol and flavonoid contents.

2.2.3. Spectrophotometric Determination of Total Polyphenols

The Wood *et al.* (2002) method was used for determination of total polyphenols. 2.5 mL of diluted (1/10) Folin-ciocalteu reagent was added to 30 µL of pepper extract. The mixture was kept for 2 min in the dark at ambient temperature, and 2 mL of calcium carbonate solution (75 g/L) was added. Then, the mixture was placed in a water bath at 50˚C during 15 min, then rapidly cooled. The absorbance was measured at 760 nm. The tests were performed in triplicate for each pepper sample. A calibration line was performed with gallic acid at different concentrations (1 µg/mL; 2 µg/mL; 4 µg/mL; 6 µg/mL; 8 µg/mL; 10 µg/mL; 12 µg/mL; 15 µg/mL; 20 µg/mL). The concentration of polyphenols was expressed in grams per liter of gallic acid equivalent extract (mg/g, GA Equivalent).

2.2.4. Spectrophotometric Determination of Total Flavonoids

The Marinova *et al.* (2005) method of [7] was used to determine total flavonoids. In a 25 mL flask, 0.75 mL of 5% (w/v) sodium nitrite ($NaNO_2$) was added to 2.5 mL of pepper extract. The pepper extract was obtained after macerating one (1) g of sample in fifty (50 mL) of distilled water. Then, the mixture was added to 0.75 mL of 10% (w/v) aluminum chloride ($AlCl_2$) and incubated for 6 minutes in the dark. After incubation, 5 mL of sodium hydroxide (1 N NaOH) was added and then the volume was made up to 25 mL. The mixture was shaken vigorously before being assayed on a Jasco V-530 UV/VIS UV-visible spectrophotometer,

Japan. The reading was taken at 510 nm. The results were performed in triplicate. The flavonoid content was expressed in grams per liter of querceline equivalent extract.

2.2.5. Statistical Processing

The numerical data obtained were entered with Excel version 2016 and processed by STATISTICA 7.1 software.

Statistical differences in means were tested by analysis of variance (ANOVA). The significance of differences between samples has been determined using Duncan's test. The significance level has been $p < 0.05$.

3. Results

3.1. Total Polyphenol Contents

Figure 2 shows the polyphenol content results of the different fresh and dried pepper samples analyzed. The dried and fresh pepper samples with the highest total polyphenol content were respectively those collected in Danané (0.8 ± 0.05 mg GAE/100g) and Azaguié (0.4 ± 0.08 mg GAE/100g) localities. The lowest total polyphenols levels were found in the dried and fresh pepper samples collected in N'douci (0.22 ± 0.02 mg GAE/100g) and PK 103 (0.22 ± 0.02 mg GAE/100g).

3.2. Total Flavonoids Contents

The flavonoid content of pepper varied significantly from one locality to another

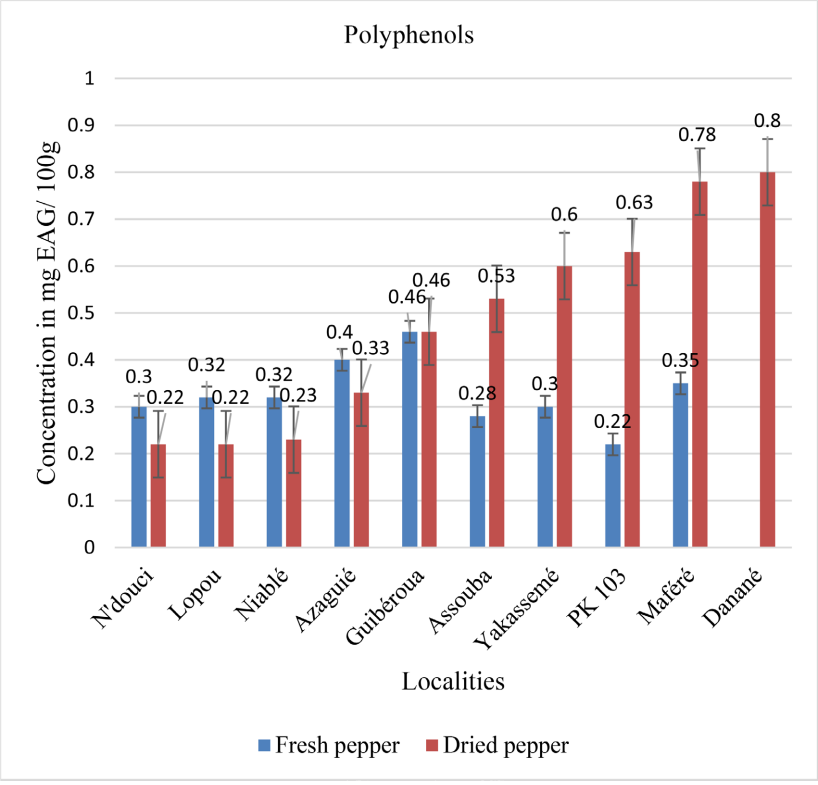

Figure 2. Total polyphenol content of dry and fresh pepper from different localities.

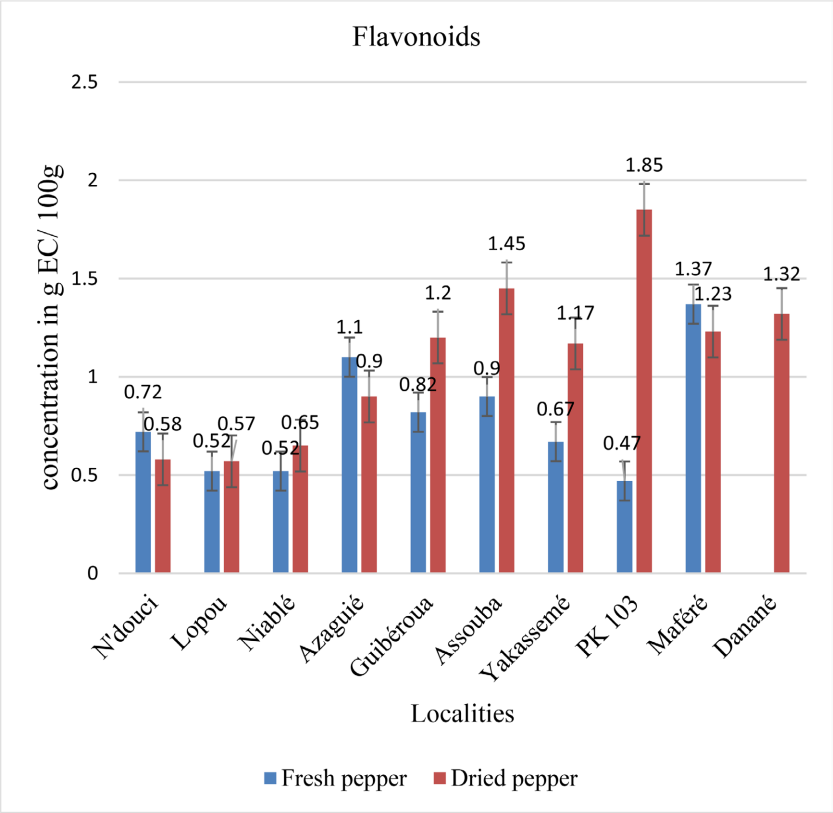

Figure 3. Total flavonoid content of dried and fresh pepper from different localities.

and ranged from (0.47 ± 0.03 g EC/100g) to (1.85 ± 0.62 g EC/100g) for fresh and dried pepper (**Figure 3**). Thus, the highest content of total flavonoids was found in the locality of PK 103 (1.85 ± 0.62 g EC/100g), followed by the locality of Maféré (1.37 ± 0.25 g EC/100g) for dried and fresh pepper respectively. Then, the lowest total flavonoids levels were obtained in the Lopou's locality (0.57 ± 0.03 g EC/100g) for dried pepper and PK 103's locality (0.47 ± 0.03 g EC/100g) for fresh pepper.

4. Discussion

The polyphenols concentrations are lower than those obtained by Ahmad and al. in 2015 [8] in *Piper nigrum* fruit (17.281 ± 0.049 mg/100g). In addition,total polyphenol contents (0.8 ± 0.05 mg/100g) are lower than those described by Nahak and Sahu, 2011 (6.2 mg/100g) [9].

In addition, these differences could be due to the different foods compositions. Like other plants, the pepper analyzed has generally lower levels of total polyphenol than those reported by Fulgencio and Isabel [10] in dried plants (155 ± 20 mg/100g), fruits (538 ± 20 mg/100g), cereals (107 ± 9 mg/100g) or coconuts (890 ± 50 mg/100g). This could be explained by climatic conditions, different agricultural practices, and t seeds maturity during harvest [11]. Secondly, high temperature, sun exposure, drought and soil salinity could stimulate polyphenol biosynthesis [12].

The flavonoid content results reported in the literature are within the range observed by the pepper samples analyzed. Indeed, Wang and al [13] found high values in Citrus spp. skins ranging from 3.27 g/100g to 4.92 g/100g. Then, the low contents as for them were found in the extracts of Amazonian plants with values of 0.07 g/100g to 1.62 g/100g [13]. In addition, Yan *et al.* [14], observed values ranging from 0.21 g/100g to 0.49 g/100g in common herbs in Korea. Then, even lower amounts (0.002 g to 0.082 g/100g) than those obtained in the analyzed pepper were found in date fruit varieties in Iran [15]. Indeed, the flavonoids importance is multiple. First, they act as protectors of other pigments against light and ultraviolet rays. Then, in the leaves for example, some flavonoids (flavonols, flavanones and flavones) absorb the distant ultraviolet rays, dangerous for the nucleic acids and proteins. Flavonoids are involved in resistance reactions to viral infestations, insects and fungi. Finally, in the medical field, they are recognized as having anti-viral, anti-tumor, anti-inflammatory and anti-allergic properties. Given its high content of total flavonoids, pepper is used as a nerve tonic, to relieve pain, atrophic arthritis, apathy, fever and health problems [16].

5. Conclusion

The present work has shown that pepper (*Piper nigrum*) domesticated in Côte d'Ivoire contains a significant amount of polyphenols and total flavonoids. Indeed, the contents of total polyphenols, expressed in milligrams equivalent of gallic acid and those of total flavonoids in equivalent of catechin are equivalent to 0.8 and 0.4 mg EAG/100g and 1.85 and 1.37 mg EC/100g of extract respectively for the dried and fresh pepper. The high flavonoids content observed in the studied pepper could give them properties are beneficial to the health of the consumer. However, this study is a necessary step for further studies on polyphenols and flavonoids in fresh and dried pepper from Ivory Coast.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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