



Study of Antioxidant Activity of Stalk and Fruit of *Solanum melongena* L. (Solanaceae)

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Authors' contributions

This work was carried out in collaboration among all authors. Author KD designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors WD and ADF managed the analyses of the study. Authors SIMD, AIM, AS and CLB performed the statistical analysis. Author ADF managed the literature searches and approved the final corrections. All authors read and approved the final manuscript.

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ABSTRACT

Background : Nowadays with the appearance of diseases such as cancer, atherosclerosis, free radicals are often singled out. What motivates scientific research in natural antioxidants.

Aim/Objective : The aim of this study was to determine the antioxidant activity of the stalks and the fruit of *Solanum melongena* L.

Study Duration : The period of the study was done on 25th July, 2015 at the Department of Pharmacy, Faculty of Medicine, Pharmacy and Odontology, University of Dakar, Senegal.

Methodology : Antioxidant activity was evaluated through two methods (DPPH and FRAP).

Results : For the FRAP test, at the highest concentration (83.3 µg/ml) the aqueous extract of the fruit (0.90±0.08) has a higher reducing power compared to those of ethanol extracts from the fruit (0.77±0.41) and the stalk (0.85±0.004). These results remain inferior to that of tannic acid (0.95±0.0005). The DPPH test reveals that the ethanolic extract of the fruit is more effective in reducing the free radical DPPH with an inhibitory concentration 50 (IC₅₀) equal to 3.37±0.03 µg / ml, followed by the ethanolic extract of the stalks (IC₅₀ = 4.46±0.24 µg / ml) and finally the aqueous extract of the fruit (IC₅₀ = 9.6±0.026 µg / ml).

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Conclusion : These results make it possible to confirm the in vitro activity of the parts studied, but in vivo studies are necessary in order to know the acute and chronic toxicities. Finally, perform a bio-guided fractionation to determine the molecules responsible for the antioxidant activity.

Keywords: Stalks; fruit; *Solanum melongena* L.; antioxidant activity; DPPH; FRAP.

1. INTRODUCTION

Over time, the notion of oxidative stress has evolved especially with the advent of molecular biology which has shown that active oxygen species (OAE) also have an important physiological role [1]. Indeed, permanently produced EOAs in the body are involved in the maintenance of cellular homeostasis (normal cell proliferation, normal metabolism, normal redox state for gene expression). In this perspective, antioxidants are then regulators of the production of OAE, which they prevent the potential deleterious effects. Nowadays, research focuses on natural antioxidants that can reduce the effects of oxygen on the corrosion of metals, but also reduce the oxidation of unsaturated fatty acids and, therefore, their rancidity [2]. Over the past decade, a real awareness of the importance of a diet rich in fruits and vegetables has emerged, particularly in the context of the prevention of metabolic diseases such as cardiovascular diseases, obesity, diabetes, neurodegenerative diseases, cancer, [3,4,5]. High consumption of fruits and vegetables has been associated with reduced risk of these diseases in many epidemiological studies [6]. Multiple constituents and micronutrients of these foods such as fiber, vitamins, minerals and polyphenols potentially play a role in this protective effect [6].

Eggplant, *Solanum melongena* L., is a fruit vegetable that belongs to the family Solanaceae and has a high content of phenolic compounds [7]. It is of economic and traditional importance in the sub-Saharan countries like Senegal where it participates in the culinary preparation. Eggplants also contain phenolic acids [7] such as caffeic acid and chlorogenic acid, and the benefits attributed to chlorogenic acid, which is the major phenolic acid found in eggplant fruits [8], include anticancer properties, antimicrobial, antiviral and inhibition of LDL (bad) cholesterol [9,10].

In this context, the overall objective of this work is to study in vivo the antioxidant activity of aqueous and ethanolic extracts of the fruit and

peduncle of purple eggplant collected in the suburbs of Dakar in Camberene in Senegal.

2. MATERIALS AND METHODS

2.1 Plant Material

The plant material was harvested in the suburbs of Dakar more precisely in Camberène in July 2015 and the fruits and stalks were dried in an oven at 60°C for four days at the Laboratory of Pharmacognosy of the Faculty of Medicine and Pharmacy and Odontology of Dakar. The plant material has been identified by Pr Diatta (Botanist).

2.2 Methodology

2.2.1 Preparation of plant extracts

The extraction of the vegetable powder was carried out by decoction of 100 g of fruit powder, brought to boiling under reflux in 1 L of water for 30 minutes. After filtration, the aqueous extract thus obtained was evaporated with Rotavapor to obtain a dry residue. Finally, 100 g of the fruit powder and 10 g of peduncle powder were extracted successively with 500 ml of ethanol by decoction several times. The extractive solutions were then evaporated to dryness.

2.2.2 DPPH test

The antioxidant capacity was evaluated according to the method described by Molyneux [11]. The extract was tested at different concentrations (12.5, 25, 50 and 100 µg / ml) with DDPH in the proportions of volumes extracted / DDP (1/4). Tannic acid was used as a reference antioxidant and tested at the same concentrations. Absorbance measurement was performed at 517 nm spectrophotometer after incubation for 30 minutes (T30) using ethanol as a blank. Three trials were conducted for each concentration of the test portion (n = 3). The results are first expressed in percent inhibition (PI) according to the formula:

$$PI = 100(A0 - A1) / A0$$

A0: absorbance DPPH A1: sample absorbance. The concentration of the sample required to neutralize 50% of the free radicals (IC 50) was obtained using the Statgraphics Plus 5.0 software. Then, the effective concentrations necessary to neutralize 50% of free radicals (EC 50) (Effective concentration) calculated from the following formula:

$$EC_{50} = IC_{50} / M \text{ DPPH}$$

M DPPH = molarité de la solution de DPPH

IC₅₀ divided by the molecular mass of DPPH and PA (antiradical power) obtained from the formula below :

$$PA = 1 / EC_{50}$$

2.2.3 FRAP test

The reductive capacity of the peduncle and fruit extracts was evaluated according to the Oyaizu method [12] adopted by Bassene [13]. Briefly, different concentrations of each extract (5.21, 20.82, 41.65 and 83.3 µg / ml) were diluted half in distilled water and then mixed with 2.5 ml of the phosphate buffer solution (0.2 M, pH 6.6) and 2.5 ml of potassium ferricyanide [K₃ Fe (CN)₆] at 1%. The mixtures obtained are incubated at 50°C. for 30 minutes and then 2.5 ml of trichloroacetic acid (10%) are added. After centrifugation at 3000 rpm for 10 min, 2.5 ml of the supernatant of each concentration was mixed with 2.5 ml of distilled water and 0.5 ml of FeCl₃ (0.1%). The absorbance is measured at 700 nm using a spectrophotometer (BTS 350,

biosystems). And the reducing powers obtained from this formula:

$$PR = 100(Aa - Ab) / Aa$$

Aa: absorbance of the extract Ab: absorbance of the white

2.3 Statistical Analysis

Variances analysis was performed using the Fisher test at a significance level of 0.05 using Statview software. Statgraphics 5.0 software was used to generate inhibitory concentrations.

3. RESULTS AND DISCUSSION

3.1 DPPH Test

The hydroethanolic extracts of the fruit and peduncles significantly inhibited the DPPH radical in a dose-dependent manner (p <0.05 versus negative control) at all concentrations tested. The ethanolic extract of the fruit (EAF) showed a higher activity than the other extracts. At the highest concentration (100 µg / ml) the ethanolic extract of the peduncle (EEP) has a percentage inhibition (PI) equal to 92.01±0.15%, EEP (80.51±0.0%) and finally, the aqueous extract of the fruit (73.66±7.07%). At the lowest concentration (12.5 µg / ml) the EEP has a very low PI of 2.89±0.40%, followed by EAF (12.55±2.23%) and EEF (38.24±0.93%). The tannic acid used as reference inhibited the radical DPPH by 87.14±2.21%. at the highest concentration and from 3.73±0.44% to the lowest (Fig. 1).

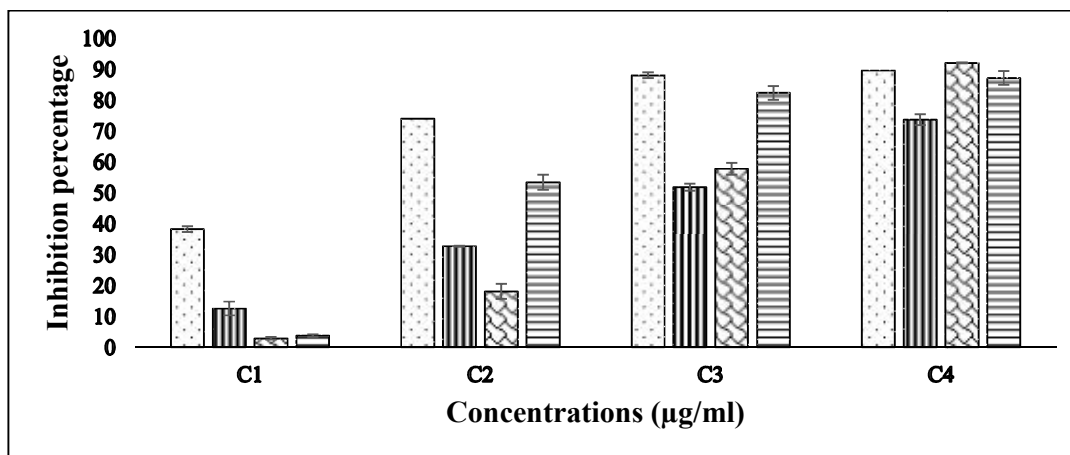


Fig. 1. Inhibitory action of extracts and tannic acid on DPPH

C1 = 12.5 µg / ml ; C2 = 25 µg / ml ; C3 = 50 µg / ml ; C4 = 100 µg / ml ; EEF = Ethanolic fruit extract ; EAF = aqueous fruit extract ; EEP : Ethanol extract stalk and tannic acid

The activities of the different plant extracts tested were compared through calculations of IC₅₀, EC₅₀ and PA. Thus, the aqueous extract of the fruit has an IC₅₀ greater than that of the other extracts (confers Table 1). Tannic acid showed an IC₅₀ equal to 4.95±0.21 µg / ml as shown in Table 1. These observations are also similar for the other parameters (EC₅₀ and PA).

3.2 FRAP Test

The determination of the reducing power of the extracts showed a better activity of the aqueous extract of the fruit compared to those of the others as shown in Fig. 2. Indeed, the aqueous extract of the fruit, at concentrations of 5.21; 20.82; 41.65 and 83.3 gave respective reducing powers of 36.05±0.08%; 52.5±2.06%; 79.42±2.27% and 89.26±2.61%. At the same concentrations, respective reducing powers of 30.28±0.03%; 60.45±0.17%; 74.79±2.28% and 77.25±2.22% for the ethanolic extract of the fruit. The ethanolic extract of the peduncle has the lowest reducing power of 6.09±1.01%. While the strongest reducing power was with tannic acid (95.86±0.06%). The results of the determination of the reducing power of the extracts and tannic acid by the FRAP method expressed as reducing power are shown in Fig. 2.

3.3 Discussion

This study aimed to search for antioxidant potentials in the peduncles and fruits of eggplant (*Solanum melongena* L.). The use of aqueous and ethanolic solvents has made it possible to extract polar compounds such as polyphenols from the peduncle and fruits that are among the main components of antioxidant-active plants [14,15,16,17]. This notion of polarity seems to be confirmed by the phytochemical screening which reveals the presence of phenolic compounds [18]. Regarding the antioxidant activity, the results obtained showed that the ethanolic extract of the fruit has the best IC₅₀ (3.37±0.03 µg / ml). These results could not be correlated with the work of Somawathi et al. [19] on the

antioxidant activity of the total phenols contained in *Solanum melongena*, whose IC₅₀ values range from 0.351±0.62 to 0.487±1.47 µg / ml, three times higher than our values. This difference could be due to the solvent also used to the presence of high content of phenolic compounds and different varieties Somawathi et al. [19]. When the IC₅₀ is higher the antioxidant activity of the compound is very important (Vice versa) [20]. These results are similar to those obtained by Ouerghemmi et al. [21] who showed an IC₅₀ of 3.96 µg / ml on *Solanum sodomaeum*. This could be due to phenolic compounds in the parts studied. Indeed, Cherifa [22] during this work on the study of the antioxidant activity of polyphenols extracted *Solanum melongena* L. by electrochemical techniques, showed a high content of polyphenols in the ethanolic fraction of whole fruit ranging from 91, 42±15.48 mg / g to 21.71±4.41 mg / g depending on the variety. This high content of polyphenols confirms our results obtained with the IC₅₀ of the order of 3.37±0.03 µg / ml. These results obtained, although significant remain slightly low compared to the reference (tannic acid 4.95±0.21 µg / ml). This difference could be due to the nature of the extraction solvent but to the presence of several phenolic compounds in our extracts, which is not the case for tannic acid. These results are comparable with the report by Uchendu, et al. [23] on in vitro antioxidant and antimicrobial activity of fruit pulp on *Solanum melongena*. The results obtained show a much greater activity for the aqueous extract of the fruit than for the ethanolic extract of the fruit and peduncle. The aqueous extract of the fruit has a strong ability to reduce ferric iron; with reducing powers ranging from 36.05±0.08% to 89.26±5.08 for concentrations of 5.21 µg / ml and 83.3 µg / ml. These results could be correlated with the work of Namrata et al. [24] on the antioxidant activity of flavonoids in *S. melongena* Linn fruit by HPTLC ; whose reducing capacity of the fraction of ethyl acetate of the fruit at 50 µg / ml is 0.4569 ±0.005, which is higher than the raw extract (0.4224±0.016). This difference can be explained by the nature of the solvent, since the solvents

Table 1. Concentrations of inhibitions and effective at 50% (IC₅₀ and EC₅₀) and the anti-radical power (AP) of the extracts and the tannic acid

	EE fruit	EA fruit	EE stalks	Tannic acid
IC ₅₀ µg/ml	3.37±0.03*	9.6±0.26*	4.46±0.24*	4.95±0.21*
EC ₅₀ g/mol	33.7±0.003*	96±0.026*	44.6±0.024*	49.5±0.024*
AP	29.6.10 ⁻³ ±0.1*	10.4.10 ⁻³ ±0.11*	22.42.10 ⁻³ ±0.12*	20.2.10 ⁻³ ±0.39*

EEF = Ethanolic fruit extract ; EAF = aqueous fruit extract; EEP: Ethanol extract stalk and tannic acid
* : significant difference versus negative control (DPPH solution) ; n = 3 for each concentration tested

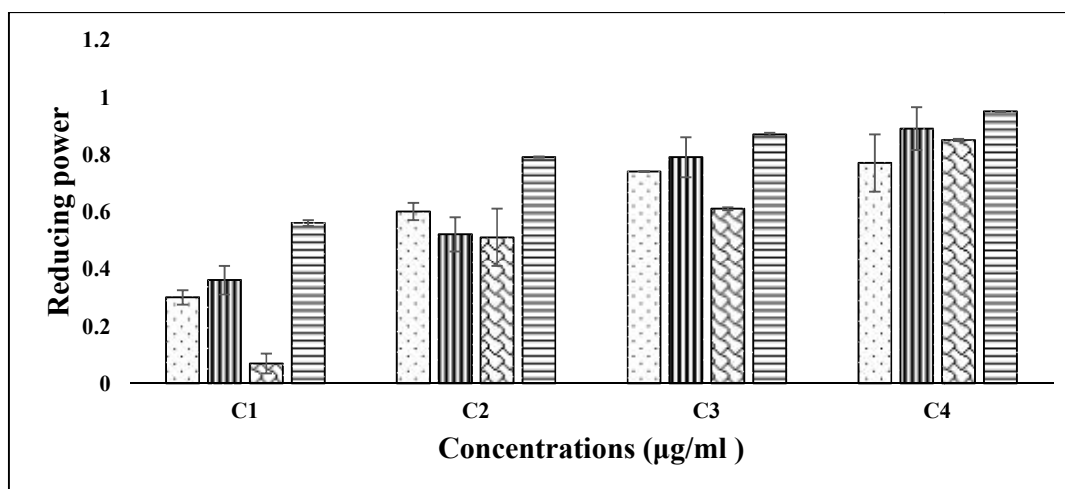


Fig. 2. Reducing power extracts and tannic acid

C1 = 5.21 µg / ml ; C2 = 20.82 µg / ml ; C3 = 41.65 µg / ml ; C4 = 83.3 µg / ml ; EEF = ethanolic fruit extract; EAF = aqueous fruit extract; EEP: Ethanol extract peduncle and tannic acid

used (water and ethanol) during the extraction are much more polar than the solvent of ethyl acetate ; but also this strong reducing power can be explained by the polyphenol richness of the fruit of *S. melongena*. The reducing powers of our extracts (ethanol extract of the fruit 0.60 ± 0.22 , aqueous extract of the fruit 0.64 ± 0.25 , ethanolic extract of the peduncle 0.51 ± 0.33) remain low compared to the reference (Tannic acid 0.79 ± 0.17).

4. CONCLUSION

The results of this work showed that peduncles and fruits have a very important antioxidant power. This is even confirmed by its use of the populations to cure various affections but also to feed. Prospective studies could guide the isolation and identification of antioxidant molecules by the bio-guided method but also the determination of acute and subacute toxicity.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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