

In Vitro Antioxidant Activity of Crude Methanolic Flavonoids from Stem Bark Of *Gmelina Arborea Roxb*

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Abstract

Hydrogen peroxide free radical scavenging assay of fractionated flavonoids of crude methanolic stem bark extract of *Gmelina arborea* was determined according to [35], five Different concentrations, 20 µg/ml, 40 µg/ml, 60 µg/ml, 80 µg/ml and 100 µg/ml of the sample tested showed respective concentration dependent percentage free radical scavenging activity 73%, 69%, 56%, 31% and 23% the highest activity was found at 100 µg/ml and the lowest activity at 20 µg/ml of the crude flavonoids. The result obtained was compared with that of standard antioxidant drug (ascorbic acid) using various concentrations; 20 µg/ml, 40 µg/ml, 60 µg/ml, 80 µg/ml and 100 µg/ml. Which have percentage free radical scavenging activity of 98.55%, 97.2%, 96.81, 95.8% and 94.5% respectively. From the results obtained shows that flavonoids of *Gmelina arborea* stem bark is a good natural antioxidant agent, which could be used for treatment of diabetics and cardio vascular diseases. It is recommended that the crude flavonoids should be screened to isolate the bioactive compounds with antioxidant activity.

Keywords: Antioxidant activity of crude methanolic flavonoids from stem bark of *Gmelina arborea roxb*

1. INTRODUCTION

Antioxidant is a chemical compound that terminates the activity of free radical, such as antioxidant compound are, thiols, carotenoid, vitamin C etc.

A free radical is any atom (e.g oxygen, nitrogen) with at least one unpaired electrons in its outermost shell, and is capable of independent existence. A free radical is easily formed when a covalent bond between entities is homiletically broken and one electron remains with each newly formed atom. [27].

Free radical is continuously produced and neutralized in our body so as to maintain the constant internal environment. Those reactive free radicals are generated due to either endogenous sources for example by products of normal metabolic processes for ATP production or exogenous sources like air pollution, cigarette smoking, UV radiation, high polyunsaturated fatty acid diet, trace metals in diet, absence of exercise etc. (Ebadi, 2001).

Reactive free radicals include reactive oxygen species (ROS) like superoxide anion radical (O_2^-), hydroxyl free radical (OH), peroxy radical (ROO) or reactive nitrogen species (RNS).

The ROS or RNS cause oxidative damage of biological macromolecules such as lipid, protein and nucleic acid which plays pivotal role in the pathogenesis of various degenerative disease like diabetes, Alzheimer's Parkinson's, cardiovascular diseases and cell death. [9].

Antioxidants are molecules stable enough to donate electrons to rampaging free radicals and neutralize them, thus reducing their capacity to damage. All human cells can protect themselves through the antioxidant defence systems which include enzymatic antioxidants. Such as superoxide dismutase (SOD), catalase and glutathione peroxide as well as nonenzymatic including glutathione, vitamin C and E by scavenging the oxidants [36].

Recently, there has been increasing in the direction of use and development of ethno medicine having strong ant-oxidative effect with low or no toxicities, Gmelina arborea is one of the reputed tropical used. (Sing et al., 2012).

2. MATERIALS AND METHODS

- Filter paper
- Volumetric flask
- Weighing balance
- Beakers. (250cm³)
- UV Spectrophometer
- Wash bottle
- Graduated measuring cylinder
- Test tubes
- Water bath

- Mechanical shaker
- Crucible
- Stirrer
- Oven
- Separating funnel

REAGENT AND CHEMICAL

- Hydrogen peroxide
- Phosphate buffer
- Distilled water
- Methanol
- n- butanol
- Potassium hydroxide
- Hydrochloric acid

COLLECTION OF THE PLANT MATERIAL AND IDENTIFICATION

The stem bark of *Gmelina arborea roxb* sample was collected from two different locations in Zaria local government, Barewa college and Nuhu Bamalli polytechnic Zaria staff quarters. Also *Gmelina arborea roxb* (Verbenacea) was also identified by its family name (Verbenacea) and authenticated by Mal. Namadi Sunusi, head of herbarium unit, at the department of botany Ahmadu Bello University Zaria (voucher no2712)

DETERMINATION OF ANTIOXDANT ACTIVITY (HYDROGEN PEROXIDE SCAVENGING ASSAY)

The ability of the plant extracts crude stem bark flavonoid of *Gmelina arborea* Roxb to scavenge hydrogen peroxide was determined according to the method of [35]. A solution of hydrogen peroxide (0.04m) was prepared in phosphate buffer (7.4PH). Extract (100ml) in distilled water were added to a hydrogen peroxide solution (0.6ml, 0.04m).

Absorbance of hydrogen peroxide at 230nm was determined, 10 minutes later against a blank solution containing the phosphate buffer without hydrogen peroxide. The percentage of the hydrogen peroxide scavenging of both extracts and standard compound were calculated.

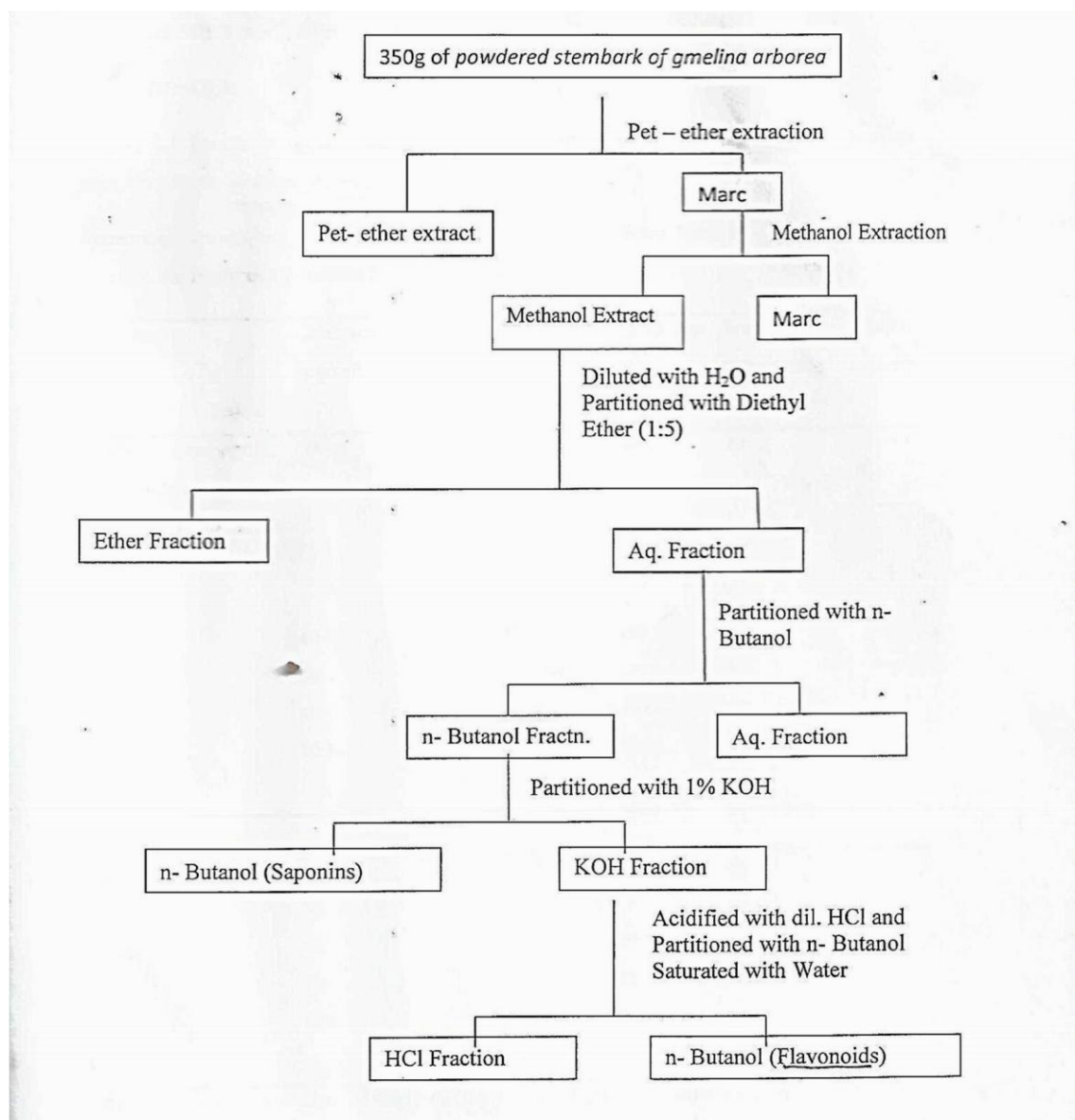
$$\% \text{ scavenged (H}_2\text{O}_2) = \frac{(\text{Abs}_{\text{sample}} - \text{Abs}_{\text{standard}}) \times 100}{\text{Abs}_{\text{sample}}}$$

Where:

Abs_{sample} is the absorbance of the sample

Abs_{standard} is the absorbance of standard

EXTRACTION PROCEDURE :



A schematic chart for fractionation of flavonoids from *Gmelina arborea* roxb stem bark (woo et al.,) 1999

3. RESULT

Result of spectrophotometric analysis of crude methanolic flavonoids from stem bark from *Gmelina arborea roxb.*

Percentage scavenging activity of crude flavonoids from stem bark of *Gmelina arborea roxb* in the hydrogen peroxide scavenging assay.

Samples	Concentration (µg/ml)	Absorbance 230 nm (mean ± SEM)	Free scavenging (%)	Radical activity
Crude Methanolic Flavonoids Extract of <i>Gmelina arborea roxb</i> Stem Bark	20	0.1986 ± 0.0002	23	
	40	0.1778 ± 0.0047	31	
	60	0.1140 ± 0.0003	56	
	80	0.07863 ± 0.0006	69	
	100	0.0691 ± 0.0253	73	
Control		0.2577		
Standard				
Ascorbic acid	20	0.22 ± 0.0031	94.50	
	40	0.22 ± 0.0248	95.80	
	60	0.17 ± 0.0259	96.80	
	80	0.13 ± 0.0012	97.20	
	100	0.11 ± 0.0115	98.55	
Control		4.0970		

Absorbance values are mean of triplicate analysis ± S.E.M (standard error mean)

4. DISCUSSION

The determination of antioxidant active of crude flavonoid from *Gmelina arborea roxb* stem bark methanolic extract revealed that the flavonoid present in the stem bark of the plant is very rich in antioxidant activity which indicate health promoting effects.

The table shows that the activity was the concentration dependent that is the higher the concentration, the higher the scavenging activity and the lower the absorbance the highest activity (98.55%) was

found at 100g/ml while the lowest activity (94.80%) was found are 20 ug/ml of the crude flavonoids comparing the activities obtained 23%, 31%, 56%, 69% and 73%.

Many researchers including [8] reported the in vitro antioxidant activity of different part of *Gmelina arborea* roxb (stem bark, leaves and fruit).

The finding of this research work is in agreement with that of [10] Conclusively, flavonoids from the stem bark *Gmelina arborea* roxb widely used ethno medicinally in many African and Asian countries collect several health benefit on this population through the improvement and the protection against acute and several oxidative stresses that could bring about disease like hypertension and diabetes.

5. CONCLUSION

The present study for the determination of antioxidant activity of flavonoids from *Gmelina arborea* roxb plant considerable free radical scavenging activity.

In this study, it is concluded that the flavonoids, present *Gmelina arborea* roxb stem bark methanolic extract have good antioxidant properly and could be further isolated to determine the bioactive compounds.

The result of this study shows that methanolic extract of flavonoid can be of used as an easily accessible source of natural antioxidant and as a possible food supplement or in pharmaceutical industry.

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