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Full Length Article

# Ethanol extract of *Tetrapleura tetraptera* fruit peels: Chemical characterization, and antioxidant potentials against free radicals and lipid peroxidation in hepatic tissues

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## Abstract

The chemical and antioxidant properties of the ethanolic extract of *Tetrapleura tetraptera* fruit peels were investigated. Dried peels of *T. tetraptera* fruits were extracted with ethanol. The extract was subjected to preliminary phytochemical screening using standard procedures. GC–MS was used in identifying the secondary metabolites. The antioxidant properties of the extract were determined by its ferric reducing activity, 2,2'-diphenyl-1-picrylhydrazyl (DPPH) and nitric oxide (NO) radicals scavenging activities, and the inhibition of lipid peroxidation in hepatic tissues of albino male rats. Preliminary phytochemical screening revealed the presence of flavonoids, phenols, tannins, saponins, terpenoids and phlebotannin. GC–MS analysis revealed the presence of D-fructose, piperazine, octodrine, glycidol, glyceraldehydes, 6-octadecenoic acid and 9,12-octadecenoic acid, with D-fructose being the most predominant compound. The extract exhibited high antioxidant activities both in vitro and ex vivo, as indicated by its ability to scavenge DPPH and nitric oxide as well as inhibition of lipid peroxidation. This is further portrayed by its ferric reducing activity. These results suggest an antioxidant protective effect of the extract against oxidative hepatic damage and can be attributed to a synergetic action of the identified bioactive compounds.

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**Keywords:** Antioxidant; Lipid peroxidation; Phytochemicals; Secondary metabolites

## 1. Introduction

Oxidative stress and free radical mediated reactions have been implicated in the pathogenesis and progression of many degenerative diseases which are major causes of death globally [1]. Reactive oxygen species

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(ROS) and reactive nitrogen species (RNS) are mostly generated from cellular metabolic reactions and could be deleterious to cells. Although low concentrations of ROS and RNS are very important in physiological processes such as cellular signaling and initiation of mitogenic response, overproduction of these reactive molecules mediates oxidative damage to cellular structures and components such as proteins, lipids and DNA [2,3]. Previous *in vivo* and *in vitro* studies have revealed that overproduction of ROS and RNS is associated with inflammation of the liver, chronic hepatic cell damage and fibrosis [4]. Increased levels of nitric oxide (NO) radicals generated from the NO synthase pathway have been implicated in the induction of nitrosative stress [5]. NO• tend to affect lipid metabolism and inhibit protein synthesis. It also reacts with superoxides to form high reactive peroxynitrite which can induce hepatic and necrotic cell death [6].

One of the major defensive mechanisms against radical induced oxidative and nitrosative stress is non-enzymatic antioxidant defense mechanism. Recently, there has been a great interest in plant foods with antioxidant properties due to their health benefits and ability to prevent chronic diseases caused by oxidative stress. The antioxidant properties of plant phytochemicals such as flavonoids, carotenoids, phenolic acids, terpenoids have been reported [7,8].

*Tetrapleura tetraptera* is a flowering medicinal plant of the Mimosaceae family, commonly known as *aridan* and *yanayan* amongst the Yorubas and Urhobos respectively in Nigeria. It has diverse medicinal purposes which include its use in the management of jaundice, inflammation, convulsion, fever, epilepsy and leprosy [7]. It is also a good spice used for cooking soups in the Eastern and Southern parts of Nigeria [8]. Preliminary investigation of the phytochemical properties of ethanolic extract of the fruit peels has been reported [9]. However, to the best of our knowledge there is dearth of information on its bioactive compounds. This present study investigates the antioxidant potentials of *T. tetraptera* fruit peel ethanolic extract against lipid peroxidation in hepatic tissues of albino rat models, as well as report its secondary metabolites.

## 2. Materials and methods

### 2.1. Plant material

*T. tetraptera* fruits were purchased from a local herb seller in Benin – City, Nigeria. The peels were removed, air dried, and blended to fine powder. The blended sam-

ple was stored in an air tight container until further analysis.

### 2.2. Experimental animals

Male wister albino rats (120–160 g) consisting of three animals were housed in standard cages at the animal house, Department of Biochemistry, University of Lagos, Lagos, Nigeria. They were acclimatized on normal pelletized mouse chow, with water given *ad libitum* at room temperature with a 12-h light and dark cycle for 7 days. At the end of the week, the rats were fasted overnight and sacrificed by cervical dislocation.

The hepatic organs were removed, rinsed in the ice-cold 1.15% KCl solution to wash off excess blood and weighed. Parts of the organs were homogenized in 0.1 M Tris/HCl buffer (pH 7.4) and centrifuged at 10,000 rpm for 20 min. The supernatants (post-mitochondrial fraction) were decanted and stored at  $\leq 4^{\circ}\text{C}$  for subsequent analysis. Each time the supernatants were outside the freezer, they were kept in ice bags.

The animals used in the present study were maintained in accordance with the animal ethical committee, University of Lagos, Lagos, Nigeria.

### 2.3. Gas chromatography–mass spectrometry (GC–MS) instrumentation

#### 2.3.1. GC

Shimadzu GC–MS QP2010 ULTRA with column: Optima 5MS (5% diphenyl 95% dimethylpolysiloxane); length: 30 m; thickness: 0.25  $\mu\text{m}$ ; ID: 0.25 mm; column flow: 1.34 mL/min Gas: helium. Column flow was set at 1.34 mL/min and temperature of column maintained at  $60^{\circ}\text{C}$  for 2 min and then raised to  $200^{\circ}\text{C}$  ( $15^{\circ}\text{C}/\text{min}$ ) followed by 9 min at  $280^{\circ}\text{C}$  ( $5^{\circ}\text{C}/\text{min}$ ).

#### 2.3.2. GC–MS

The National Institute of Standards and Technology (NIST) mass spectral program 2011 was used for mass spectral survey. The concentrations of the identified compounds were calculated using area normalization over flame ionization detector response method.

#### 2.3.3. Extraction

The blended samples were extracted by percolation with ethanol and concentrated over a rotary vacuum evaporator.

#### 2.3.4. Phytochemical analysis

The preliminary qualitative and quantitative phytochemical properties of the extract were determined using

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