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Investigating the Bioactive Compounds in Leaves' Extracts of *Gossypium barbadense* Species using High-Performance Liquid Chromatography

Adeola Abiola*, Abiodun Ojo, Amos Onasanya, Ezekiel Adewole
Chemical Sciences Department, Afe Babalola University, Ado-Ekiti, Nigeria

Abstract
Gossypium barbadense leaves have been used traditionally in the treatment of several ailments such as asthma, pyrexia and yellow fever, with no much scientific scrutiny of its acclaimed efficacy. This study investigated the bioactive components of hybrid and traditional Gossypium barbadense leaves using high-performance liquid chromatography (HPLC). Extraction of samples were carried out by maceration using 95% ethanol. Bioactive compounds were determined using agilent technology; high-performance liquid chromatography. HPLC analysis revealed presence of bioactive compounds in the hybrid leaf, such as: eugenol 1.522 mg/g. camphor 0.128 mg/g, alpha-tumerone 0.095 mg/g, epicatechin 0.118 mg/g, alpha-pinene 0.105 mg/g. bioactive compounds in the tradition leaf include: ellagic acid 1.531 mg/g, Zeaxanthine 0.927 mg/g, P-coumaric acid 0.409 mg/g, catechin 0.279 mg/g. This study revealed that leaves of hybrid and traditional Gossypium barbadense have different bioactive compounds. Quercetin and epicatechin were bioactive compound they have in common and the traditional plant is richer in bioactive compounds. Difference in the compositions of the plant species is influenced by genetic modification of the hybrid specie. Hybrid specie is much useful for production of textile materials, traditional species with more bioactive compounds is considered for treatment of diseases.
Keyword: *Gossypium banbardense, leaves, extracts, bioactive compounds, concentrations.*

INTRODUCTION

Cotton, also referred to as *Gossypium* is native to tropical and sub-tropical regions of the world (Viot and Wendel, 2023), where it has widespread use as herbal medicine for myriads of diseases including fever (Kaleson and Neksumi, 2022). Extracts of the plants' leaves, seed and bark have been utilized traditionally for the treatment of various diseases such as fever, ulcer, inflammation, microorganism infection (Yilma *et al.,* 2023), without much scientific scrutiny as to its efficacy and the chemical compositions responsible for this traditional medical acclaimed efficacy. The study aims to elucidate the potential of *Gossypium barbadense* extracts as functional ingredients in nutraceutical and pharmaceutical applications. Furthermore, understanding the chemical differences between hybrid and traditional plants could inform cultivation strategies for enhanced bioactive compound yield.

MATERIALS AND METHODS

Materials

Leaves of hybrid *Gossypium barbadense* were harvested freshly in the morning in a cellophane bag from Oyo, Oyo state Nigeria and traditional specie from Ado-Ekiti. Plant samples were deposited at the university herbarium of Federal University Oye-Ekiti with voucher numbers 250 and 251 respectively. Samples were washed, drained and air dried in the laboratory (Kishor *et al.,* 2021).

Methods

Leaves of traditional and hybrid *Gossypium barbadense* were weighed separately into 2.5 L jar with extraction solvent (95% ethyl alcohol); 100 g/500 ml (Teshome *et al.,* 2021). Mixtures were allowed to macerate for 72 hours (Chibuye *et al.,* 2023). Concentrated extracts were weighed in a conical flask and mixed with methanol; 100 mg/5 ml. Mixtures were filtered and analyzed by reversed phase HPLC (Alkhamaisah *et al.,* 2019).

Concentrations of compounds were determined using; (Deem and Hein, 2023).

$$\text{Concentration (mg/g)} = \frac{\text{peak area of compound} \times \text{concentration of standard}}{\textit{peak area of standard}}$$

RESULTS

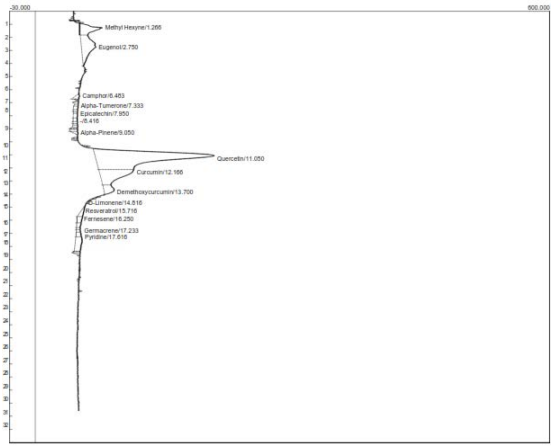

Fig.1: High-Performance Liquid Chromatography (HPLC) Chromatogram of hybrid leaf

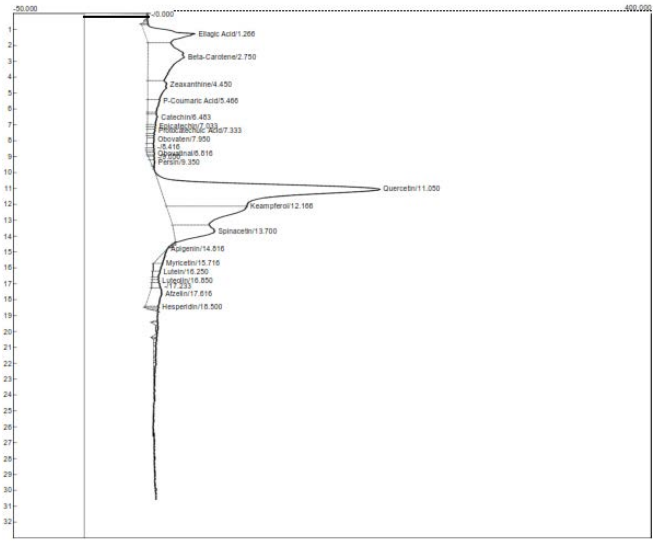

Fig.2: High-Performance Liquid Chromatography (HPLC) Chromatogram of traditional leaf

DISCUSSION

Figure 1 and 2 above, showed bioactive compounds in *Gossypium barbadense* hybrid leaf and traditional leaf respectively. Hybrid leaf contain fourteen compounds at methyl hexyne 0.893 mg/g, eugenol 1.522 mg/g. camphor 0.128 mg/g, alpha-tumerone 0.095 mg/g, epicatechin 0.118 mg/g, alpha-pinene 0.105 mg/g, quercetin 8.210 mg/g, curcumin 2.050 mg/g, demethoxycurcumin 0.483 mg/g, limonene 0.203 mg/g, resveratrol 0.191 mg/g, fernesene 0.119 mg/g, d-germacrene 0.138 mg/g and pyridine 0.558 mg/g. Nineteen compounds found in the tradition leaf were: ellagic acid 1.531 mg/g, Zeaxanthine 0.927 mg/g, P-coumaric acid 0.409 mg/g, catechin 0.279 mg/g, obovaten 0.118 mg/g, obovatinal 0.059 mg/g, persin 0.062 mg/g, lutein 0.119 mg/g, afzelin 0.674 mg/g, kaempferol 3.381 mg/g, epicatechin 0.059 mg/g, quercetin 10.115 mg/g, spinacetin 1.549 mg/g, apigenin 0.2033 mg/g, myricetin 0.191 mg/g, hesperidin 0.067 mg/g, beta-carotene 3.073 mg/g, protocatechuic acid 0.095 mg/g and luteolin 0.060 mg/g. common compounds between the extracts were epicatechin; 0.118 - 0.059 mg/g with the hybrid having higher concentration while traditional leaf had higher concentration of quercetin 8.210 - 10.115 mg/g. Bioactive chemicals are important for normal functioning of the plant and are also required for therapeutic applications for human (Refaz *et al.,* 2023). Bioactive compounds possess biological activities beneficial to the plants and humans such as D-germacrene and farnesene provide potential for prevention of insect pest from damaging the crop (Khalid *et al.,* 2023). Zeaxanthine along-side lutein are important in prevention or treatment of ocular diseases such as cataracts and age-related macular degeneration (Tudor and Pintea 2020). Keampferol possesses ameliorating effect on ARDs (age-related diseases) such as dementia, depression, diabetes (Hussain *et al.,* 2024) (Jaul and barron, 2017). Apigenin is used in the management of inflammatory-related diseases such as cancer, diabetes, obesity, depression etc. (Mushtaq, 2023). Mycetin activities include; anticancer, antidiabetic antihypertensive neuroprotection on Alzheimer, Parkinson (Taheri *et al.,* 2020). Spinacetin anti-inflammatory activity include inhibiting the release of histamine and production of inflammatory mediators such as leukotriene C4 (LTC4) and interlukin-6 (IL-6) (Ji *et al.,* 2018). Hesperidin plays critical role in attenuation of oxidative stress-related toxicities, prevention of membrane integrity loss (Famurewa *et al.,* 2022). Beta-carotene is important for embryonic development, correct growth and sight it also protect the skin lesions against oxidation and exposure to UV radiation (Ludmila and Joanna, 2018). Afzelin biological activities include anti-human lung cancer, anti-cholinesterase and anti-glucosidase (Cao *et al.,* 2021). Protocatechuic acid reduces the risks of cardiovascular disease, due to its anti-inflammatory properties (Semaming *et al.,* 2015). P-coumaric acid possesses scavenging properties important in prevention of oxidative stress related diseases (Shuhadeep *et al.,* 2021). Luteolin is effective in the treatment of inflammatory disorder, hypertension and cancer (Prasher *et al.,* 2022). Persin is effective in the prevention and treatment of cancer, cardiovascular and microbial diseases due to its antioxidant properties (Bhuyan *et al.,* 2019). Obovatinal and obovaten prevent cytotoxicity (Li *et al* 2018). Ellagic acid has antioxidant and anti-inflammatory properties effective for use as antiallergic, hepatoprotective, nephroprotective and neuroproptective activities (Sharifi-Rad *et al.,* 2022). Eugenol has long been used in cosmetology, medicine and pharmacology due to its properties such as antibacterial, antiviral and antifungal (Ulanowska and Olas, 2021). Camphor possesses a strong aroma hence,

it is used as flavor in cooking and repellent in medicine (Ponomarev and Howard, 2016). Alpha-tumerone is an antioxidant known to provide protection against oxidative degradation (Singh *et al.,* 2010). Alpha pinene was effective in the alleviation of nociception caused by formalin, according to (Kaveh *et al.,* 2023). Quercetin is a bioflavonoid known for activities such as anti-inflammatory, antihypertensive anti-obesity activities (Alexander *et al.,* 2016).

CONCLUSION

This study revealed leaves of *Gossypium barbadense* species have different compositions of bioactive compounds. epicatechin and quercetin are the two compounds they share in common as revealed by high-performance liquid chromatography assay. Hybrid specie are cultivated for more volume of cotton for textile purposes, traditional specie contained more bioactive compounds, therefore, should be utilized for the treatment of diseases. Individual specie might be used for it intended purpose as spices and herbs if understudied, combination of both leaves will increase their biological activities.

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LIST OF ABBREVIATIONS

HPLC high-performance liquid chromatography
mg/g milligram per gram

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