UNIVERSITY OF KWAZULU-NATAL

A COMPARATIVE CHEMISTRY OF COA® HERBAL MEDICINE AND HERBAL EXTRACTS OF VERNONIA AMYGDALINA (BITTER LEAF) AND PERSEA AMERICANA (AVOCADO)

By

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PREFACE

The experimental work described in this thesis was conducted at the Medicinal Chemistry Laboratory at the School of Pharmaceutical Sciences, University of KwaZulu- Natal, Westville, South Africa, and the Mass Spectrometry Laboratory, School of Chemistry, University of KwaZulu- Natal, Pietermaritzburg, South Africa, from March 2018 to November 2018, under the supervision of Dr. Manimbulu Nlooto and co-supervision of Dr. Rajshekhar Karpoormath.

This work has not been submitted in any form for any degree or diploma to any tertiary institution~~,~~. Where use has been made of the work of others, it is duly acknowledged in the text.

Akwasi Boadu ———————————— Date ————————————

As the candidates supervisor I agree to the submission of the thesis:

Dr. Manimbulu Nlooto———————————— Date————————————

Dr. Rajshekhar Karpoormath———————————— Date ————————————

DECLARATION

I, **Akwasi Boadu**, declare that

i. The research reported in this dissertation, except where otherwise indicated, is my original work.

ii. This dissertation has not been submitted for any degree or examination at any other university.

iii. This dissertation does not contain other persons' data, pictures, graphs or other information, unless specifically acknowledged as being sourced from other persons.

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v. Where other written sources have been quoted, then:

their words have been re-written but the general information attributed to them has been referenced.

Signed: _____ Date: _____

List of manuscripts

1. Boadu, A., Singh, S., Karpoormath, R., Nlooto, M. 2018. Review on ethnomedicinal uses, phytochemical constituents and pharmacological evidence on leaf extract of Persea americana and Vernonia amygdalina of the African continent, Indian Drugs, submitted for review.
2. Boadu, A., Nwabuife, J., Singh, S., Bodede, O., Karpoormath, R., Nlooto, M. 2018. A comparison of phytochemical composition of COA®herbal medicine and crude leaf extracts of *Vernonia amygdalina* (bitter leaf) and *Persea americana (*Avocado*)* collected from Cape Coast, Ghana and Durban, South Africa.

Dedicated

To

Jehovah,

My Wife, Son, and Daughters for

their unconditional love and

encouragement.

My Late Father and Brother

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List of abbreviations

AIDS	**Acquired Immunodeficiency Syndrome**
CAM	Complementary and Alternative Medicine
COA®	Center of Awareness
DCM	Dichloromethane
EtOAc	Ethyl Acetate
EtOH	Ethanol
FDA	Food and Drugs Authority
GC-MS	Gas Chromatography- Mass Spectrometry
GH	Ghana
Hex	Hexane
HIV	Human Immunodeficiency Virus
NA	Not Applicable
PA	*Persea americana*
R_t	Retention time
SA	South Africa
TAM	Traditional African Medicines
TM	Traditional Medicine
TMPs	Traditional Medical Practitioners
UKZN	University of KwaZulu Natal
VA	*Vernonia amygdalina*
WHO	The World Health Organization

ABSTRACT

The aim of this study is to investigate the phytochemical compounds present in standard COA®, dichloromethane (DCM), ethanol (EtOH), hexane (HEX), and ethyl acetate (EtOAc) , extracts of COA® compared to leaf extracts of *Vernonia amygdalina* and *Persea americana* collected from Cape Coast (Ghana) and Durban (South Africa) by phytochemical screening techniques and gas chromatography-mass spectrometry (GC-MS) analysis.

Findings from this study revealed that leaf extracts of *P. americana and V. amygdalina* have been used in many local African communities for management of various diseases. Ethnomedicinal use and pharmacological properties of leaf extracts of *P. americana* and*V. amygdalina* may justify polyherbal formulation involving the two plants in the treatment of diseases such as diabetes, hypertension and other diseases.

Outcomes of the preliminary phytochemicals screening showed the presence of alkaloids, anthraquinones, saponins, flavonoids, tannins, terpenoids and glucosides in ethanolic leaf extracts and standard COA®.

GC-MS study of standard COA®, COA® extract and leaf extracts of *P. americana and V. amygdalina* collected from Ghana and South Africa had 60y phytochemical compounds identified in the hexane extract of COA® extract, 47 in DCM, 37 in ethyl acetate, 18in ethanol and 11 in standard COA® herbal medicine. The identification of phytochemical compounds and pharmacological actions was based on the name of the chemical compound, retention time and molecular formula from GC-MS analysis. The major phytochemicals common to COA® extract and leaf extracts of *P. americana and V. amygdalina* were heneicosane, phytyl acetate, pyrene, octadecanoic acid, eicosane, 2-methyltetracosane, pentadecanoic acid, hexadecanamide, and octadecanamide. Most of these major phytochemicals are present in ethanolic extracts of both COA® and leaf extracts of *P. americana and V. amygdalina*

Leaf extracts of *P. americana and V. amygdalina* collected from Ghana have more phytochemicals compared to that of South Africa.

The finding, of this study, confirm the presence of *P. americana and V. amygdalina* leaf extracts in COA® herbal medicine. It also confirms the profound variations in phytochemicals of *P. americana and V. amygdalina* leaf extracts due to the effects of environmental factors and geographical locations.

Keywords: COA® herbal medicine; *Vernonia amygdalina; Persea americana;* phytochemicals; GCMS analysis.

CHAPTER 1: INTRODUCTION

CHAPTER 1: INTRODUCTION

1.1. Background

Traditional medicine (TM), identified as native healing, folk medicine, ethnomedicine, or complementary and alternative medicine (CAM) is an ancient and culture-bound approach of treatment that humanity has utilized to manage and cope with numerous disorders that have endangered their lifestyle and survival.[1] In previous years, there has been a rapid expansion in the industry of plant-based medication. It is acquiring popularity in developing and developed nations around the world due to its natural origin and lower side effects.[2]

Most users of TM such as Ayurvedic, among others and Traditional African Medicines (TAM)~~;~~ probably the most diversified, assume that since TMs have been used for decades, they are safer with low toxicity and side effects and allopathic medicines are often less ineffective.[3] There are challenges in the formulations of herbal medicines, due to lack of standardization, identification, lack of quality control and pharmacopoeial standards.[4] A number of factors affect the quality of herbal medicines, such as quality of raw materials, the source of plant material, harvesting, storage processing methods, and other contaminants. Stakeholders seek to build quality parameters for collection, handling, processing, production and safety of herbal medicines.[5]

Published guidelines of standards such as the Ayurvedic Pharmacopoeia, for herbal medicines, provides standards for the evaluation of herbal medicines such as external appearance[6], ash values[7], foreign organic matter[8], macroscopical and microscopical.[9] Moisture content indicates the quality and firmness of herbal medicines against deterioration of materials, while extractive values and crude fiber are part of the standards for herbal medicines.[10,11] In the absence of such information on the various plant extracts, it is likely impossible to isolate or standardize the active components responsible for the activity.[5] Qualitative chemical evaluation includes but not limited to, extraction with appropriate solvents, phytochemical screening, isolation and characterization of active compounds by gas chromatography-mass spectrometry (GC-MS), biological examinations and purification of active compounds for their pharmacological importance[12] and finds amounts of major classes of phyto-constituents[13] and their toxicity.[14]

The greater part of the African population (80%) use TM for their main health care demands. A lot of people on the African continent use TMs, although others combine it with conventional drugs.[15] Traditional medical practitioners (TMPs) are an essential feature of the health-care delivery procedure. The African continent is well known to have a wealthy natural

and ethnical diverseness with the noticeable localized difference in therapeutic practices and is regarded as the support of humankind. Several local communities of Africa have used plants such as *Vernonia amygdalina* (VA) and *Persea americana* (PA) in TM for the prevention and treatment of diseases such as skin rashes among Human Immunodeficiency Virus (HIV) infected persons.[16]

COA® herbal medicine (COA®) has been most frequently and widely used by people of South African continent for the management of several ailments, such as cancer, diabetes, hypertension, and immune support, other diseases and general well-being and approved by local food and drugs board authority (FDA) (FDA/DRID/HMD/HMU/16/0981~~,~~). COA® contains *Vernonia amygdalina, Persea americana, Azadirachta indica, Carica papaya, Spondias mombin* and *Ocimum viride.*

Although COA® is FDA approved in Ghana and widely available in the clinical market, there is no written validatory documentation on phytoconstituents of COA® herbal medicine. So, there is a need to validate and proof the phytoconstituents present in the marketed COA® that are responsible for the pharmacological activities found in leaf extracts of VA and PA.

1.2 A brief overview of the literature

A comprehensive literature review has been presented in Chapter Two.

Two plants *V. amygdalina* and *P. americana* were selected out of the six plants present in COA® formulation reason being that other candidates were investigating other plants from the same medication~~,~~ and were collected from the different location in Cape Coast (Ghana) and Durban (South Africa). Voucher specimens were deposited in the University of KwaZulu-Natal Herbarium for botanical authentication if required in future.

Description of *Vernonia amygdalina* (bitter leaf)

Vernonia amygdalina (bitter leaf) has a bitter taste. The bitterness can, however, be abated by boiling or by soaking the leaves in several changes of water. The bitter taste is due to anti-nutritional factors such as alkaloids, saponins, tannins, and glycosides.[17] It is a shrub with classification as Kingdom; Plantae, Order; Asterales and a member of the Asteraceae family, Genus; *Vernonia* and Species; *amygdalina*. It is a short cycled crop, which can be harvested at least twice a month depending on environmental conditions.[18] On the African continent, bitter leaf largely grows in tropical regions such as Ghana, Nigeria and semi-tropical regions such as Zimbabwe, Lesotho, and parts of KwaZulu-Natal in South Africa.

Figure 1. The aerial part of *Vernonia amygdalina.*

Description of the.*Persea americana*

Persea americana is commonly known as Aligator pear, Avocado, butter, Avokado (Afrikaans), fruit, mparachichi (Swahili), Maluma (South Africa), Paya (Ghana-Twi), and Pia (Yoruba). It is classified scientifically as belonging to the Kindom; Plantae, Order; Laurales and a member of the Lauraceae family, Genus; *Persea* and Species; *americana*. It is commonly cultivated in both tropical and subtropical regions in the world. The plant leaf is glossy and green, with a bell-shaped fruit (green/brownish).[19]

Figure 2. The aerial part of *Persea americana.*

1.3 Problem statement

The African continent has historical traditions of conventional treatments that require scientific exploration with systemic strategies towards detection of chemical substances and their bioactive principles to cure disorders. The plant leaf extracts found in COA® have been used by indigenous people in African and countries in other continents for the management of diseases.

There is a need for the provision of a scientific rationale in a form of research to establish the phytochemicals responsible for the pharmacological activities of herbal medicines, their effectiveness, safety, and toxicity. The methods of analysis of herbal medicines, such as phytochemical screening and GC-MS analysis are used in the identification of chemical compounds from different solvent extracts of COA® and a standard COA®.

The geographical area of the leaf extracts used in the preparation of the COA® may have differences or similarities in their phytochemicals responsible for the pharmacological activities of the COA®. esearch by Inbathamizh *et al*. (2013), suggested the profound effect of geographical properties on the phytochemical composition which further influenced the antioxidant potential of *Moringa oleifera* flowers.[19] It is not well known whether the medicinal plants in the Cape Coast of Ghana have similarities/and or differences with those plants found in Durban.

This study provides a scientific rationale, on the class of phytochemicals, in standard COA®, ethanolic leaf extracts of *V. amygdalina* and *P. americana* collected from Cape Coast (Ghana) and Durban (South Africa) using phytochemical screening methods. GC-MS analysis of the chemical compounds from standard COA®, leaf extracts of *V. amygdalina* and *P. americana* (Ghana and South Africa) and extracts of COA® in four solvents of extractions, namely dichloromethane, hexane, ethanol, and ethyl acetate. [20]

1.4 Research questions

The general question of the proposed study is: "What class of phytochemicals and chemical compounds are found in standard COA®, COA® extracts in four solvents and leaf extracts in four solvents of *V. amygdalina* and *P. americana* collected from Cape Coast (Ghana) and Durban
(South Africa) using phytochemical screening methods and GC-MS?" Specific questions to answer the general question are as follows:

1. What class of phytochemicals are found in standard COA® herbal medicine and ethanolic leaf extracts of *V. amygdalina* and *P. americana* using phytochemical screening methods?
2. What chemical compounds/ phytochemicals are found in GC-MS analysis of standard COA® herbal medicine compared to four extracts of COA® in DCM, hexane, ethanol and ethyl acetate?
3. What similar chemical compounds/ phytochemicals from GC-MS analysis are contained in DCM, hexane, ethanol and ethyl acetate extracts of COA® and leaf extracts of *V. amygdalina* and *P. americana* from the same four solvents of extractions, collected from Cape Coast (Ghana) and Durban (South Africa)?

4. What phytochemicals are similar or different from GC-MS analysis of leaf extracts of the four solvents of extraction of *V. amygdalina* and *P. americana* found in Cape Coast (Ghana) and Durban (South Africa)?

1.5 Aim and specific objectives of this study

The aim of this study is to compare the different phytochemicals and their chemistry found in both COA® herbal medicine and crude extracts from *V. amygdalina* and *P. americana* found in Cape Coast (Ghana) and Durban (South Africa) using phytochemical screening methods and GCMS analysis.

The objectives are:

1. To identify and classify the chemical composition of the COA® herbal medicine using phytochemical screening methods and GC-MS analysis.
2. To establish the chemical composition of the leaf extract of *V. amygdalina* and *P. americana* found in Cape Coast (Ghana) and Durban (South Africa) using GC-MS analysis.
3. To compare the similarities and/or differences of the phytochemicals in the leaf extracts of *V. amygdalina* and *P. americana* found in Cape Coast (Ghana) and Durban (South Africa) and COA® herbal medicine.

1.6 General methodology

1.6.1 **Procedure**

This study was done through the classification of phytochemicals, identification of chemical compounds and the comparative study of the effect of geographical locations on type and number of chemical compounds in the leaf extracts of the medicinal plants using preliminary phytochemical screening and choosing the appropriate solvent of extraction for GC-MS analysis.

1.6.2 Study design

This study is experimental based on the phytochemical analysis of COA® herbal medicine and leaf extracts of *V. amygdalina* and *P. americana* collected from Cape Coast (Ghana) and Durban (South Africa), using phytochemical screening methods in the Medicinal Chemistry laboratory at the School of Pharmaceutical Sciences, University of KwaZulu Natal (Westville) and GC-MS, at the Mass Spectrometry Laboratory, School of Chemistry University of KwaZulu Natal – Pietermaritzburg.

1.6.3 Study setting

Samples of the COA® herbal medicine and the leaves of *V. amygdalina* and *P. americana* found in Cape Coast (Ghana) and Durban (South Africa).

1.7 Layout/ structure of the thesis

The thesis entitled "**A comparative chemistry of COA® herbal medicine and leaf extracts of *Vernonia amygdalina* (bitter leaf) and *Persea americana* (avocado)"** that comprises of the work done by the author in the Medicinal Chemistry Laboratory at the School of Pharmaceutical Sciences, University of KwaZulu -Natal (Westville) and GC-MS, at the Mass Spectrometry Laboratory, School of Chemistry University of KwaZulu Natal – Pietermaritzburg.

This thesis has been organized into **Four Chapters**

Chapter I: Chapter one of this thesis deals with **Introduction** along with background and a brief overview of the literature of the proposed studies. This section gives an overview of the statement of the problem, research questions, the aim and objectives of the study. The brief methodology has been discussed to achieve specific aim and objectives.

Chapter II: Chapter two is a **Review Paper**, which has been prepared and submitted following the guidelines of the Journal **Indian Drugs** titled, **"Review on ethnomedicinal uses, phytochemical constituents and pharmacological evidence on leaf extract of *Persea americana* and *Vernonia amygdalina* of the African continent".**

Chapter III: Chapter three deals with an **Experimental** research manuscript. This manuscript has been prepared according to South African Journal of Chemistry submission guidelines, entitled **"A Comparative Chemistry of COA® Herbal Medicine and Leaf Extracts of *Vernonia amygdalina* (bitter leaf) and *Persea americana* (Avocado) collected from Ghana and South Africa".**

Chapter IV: Chapter Four deals with **Synthesis and Discussion** of the findings. It summarizes the comparative chemistry of COA® herbal medicine and leaf extracts of *Vernonia amygdalina* (bitter leaf) and *Persea americana* (Avocado) collected from Ghana and South Africa in four solvents of extraction (DCM, hexane, ethanol and ethyl acetate). It also discusses the impact of geographical location on the phytoconstituents of the plants. The finding of the phytoconstituents in herbal formulation motivate researchers to work in traditional medicines.

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CHAPTER 2: MANUSCRIPT 1

"Review on ethnomedicinal uses, phytochemical constituents and pharmacological evidence on leaf extract of *Persea americana* and *Vernonia amygdalina* of the African continent"

Published: Indian Drugs (Manuscript NO. 11623)

After the introduction chapter, a full review paper on *Vernonia amygdalina* and *Persea americana* title **"Review on ethnomedicinal uses, phytochemical constituents and pharmacological evidence on leaf extract of *Persea americana* and *Vernonia amygdalina* of the African continent"** was prepared. The paper presented findings of nutrients and phytochemicals and the use of these plants in the treatment of ailments as well. This paper was prepared and submitted following the guidelines of the journal '**Indian Drugs'.** The manuscript has been accepted for publication by the Journal.

Review on ethnomedicinal uses, phytochemical constituents and pharmacological evidence on leaf extract of *Persea americana* and *Vernonia amygdalina* of the African continent

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Abstract:

People on the African continent frequently consume *Persea americana (P. americana*) *and Vernonia amygdalina* (*V. amygdalina*) for food and medicinal purposes. The aforementioned review is an effort to compile potential ethnomedicinal, phytochemical and pharmacological knowledge of the above two plants from the year 2000-2018. A search was carried out using a combination of keywords: *Persea americana, Vernonia amygdalina*, Traditional medicine, ethnomedicine, Avocado, Bitter leaf, and Integrated approaches. Published papers in the English language were retrieved from the online bibliographical databases: Web of Science, PubMed, Google Scholar, Scopus and Cochrane library electronic databases. Findings from this review revealed that leaf extract of *P. americana* and *V. amygdalina* have been used in local African communities for the management of various diseases. Ethnomedicinal uses, and pharmacological properties of leaf extracts of *P. americana* and *V. amygdalina* may justify polyherbal formulation involving the two plants in the treatment of diseases such as diabetes, hypertension and other diseases.

Keywords: *Persea americana Vermonia amygdalina*, Traditional medicine, ethnomedicine, Avocado, Bitter leaf and Integrated approaches.

Introduction

Most modern medicines or drugs have been generated or synthesized from natural products, specifically medicinal plants[1]. The African continent and particularly Southern Africa is a heart of medicinal plants with abundant varieties. Statistically, near around 25% of the entire quantity of essential plants in the earth is located in Africa and Sahara[2]. The indigenous communities use these medicinal plants as part of their ethnomedicinal practice on the African continent. Ethnomedicine is defined as "*those beliefs and practices relating to disease, which are the products of indigenous cultural developments and are not explicitly derived from the conceptual framework of modern medicine*"[3]. The World Health Organization (WHO) defines traditional medicines as" "*the sum total of the knowledge, skill, and practices based on the theories, believes and experiences indigenous to different cultures, whether explicable or not, used in the maintenance of health as well as in the prevention, diagnosis, improvements or treatment of physical and mental illness*"[4]. The fifty-sixth world health assembly of WHO on traditional medicines indicated that in Ghana, Mali, Nigeria, and Zambia, herbal medicines have been the first line of treatment of malaria for more than 60% of children with high fever[5]. The Africa continent is enormous, bestride by the equator and endowed with more than 50,000 distinct known species in the sub-Saharan African region alone[6]. Several countries have used plant resources in traditional medicine for the prevention and treatment of diseases. African traditional medicine is the oldest, and perhaps the most assorted, of all therapeutic systems[7]. In the African continent, people have used or continue to use traditional medicines for the management and /or treatment of ailments in both humans and livestock[8,9]. The most preferred part of medicinal plants for treatments in Traditional African Medicines (TAM) is the young succulent, green and fresh leaves. Although other parts such as the roots, stems, and flowers are used. In Traditional African Medicines, medicines made from the leaves of plants are largely water-based organic decoction extract used as tonic drinks[10].

Majority of the people of Ghana and Africa still give preferences to traditional medicine for their health needs[11]. In South Africa, a great proportion of the population relies on plant-derived remedies to treat diseases[12]. A large proportion of the South African population in the Eastern Cape still depend on traditional herbal medicines to manage numerous diseases such as skin rashes among Human Immunodeficiency Virus (HIV) infected persons[13].

*Persea americana (*avocado), belonging to the Lauraceae family and cultivated in both tropical and subtropical regions in the world is used by indigenous African communities for the treatment of numerous disease regimes[14]. The plant has a simple glossy, green leaf with fruits in bell shape (largely greenish or brownish)[15].

Ethnomedicinal study of *P. americana* conducted in South Africa[16], Uganda[17] and Cameroon[18] respectively reported the use of root, seed and leaf extracts in the treatment of diabetes. The use of leaf extracts of *P. americana* in the treatment of malaria has been reported in Ghana[19] and Nigeria[20, 21]. Other ethnomedicinal treatments include but are not limited to hypertension and typhoid.

The consumption of the fruit as food and the use of different parts of the plant in the treatment of diseases by local communities around the world have been reported. It has been reported the use of the fruit in the preparation of baby food in most African communities[22]. Leaves of *P. americana* are used by the people of Bukavu city, Democratic Republic of Congo, in the treatment of ailments such as diarrhea, hypertension and cough conditions[23]. A study conducted in Nigeria reported the use of dried leaves in the treatment of hypertension[24].

Amaechina along with his co-workers reported the pharmacological use of aqueous seed extracts of *P. Americana* in *the* reduction of blood pressure of normotensive rabbits and rats in Nigeria[25]. Analgesic and anti-inflammatory effects of aqueous leaf extract have been reported in mice. The extract was able to raise the pain threshold to heat in the animals[26]. Leaf Extract of *P. americana* has been used as a natural supplementary remedy in essential hypertension and certain cases of cardiac dysfunctions in rural Africa communities [27].

Natural bioactive components (phytochemicals) in *P. americana* serve as nutrients to protect the plant against diseases[28]. Major chemical components found in different parts of the plant reported to date include alkaloids, flavonoids, saponins, phenols, terpenoid, glycosides, furan ring-containing derivatives, and coumarin[29-33]. Alkaloids, flavonoids, saponins, and phenols are responsible for analgesic and antispasmodic, anti-oxidant, antibiotic and antidiabetic activities respectively[34,35]. Isorhamnetin, luteolin, rutin, quercetin, and apigenin have been isolated phytochemicals from leaves of dried avocado[36]. Among these isorhamnetin and quercetin showed anti-oxidant activities[37].

Another medicinal plant of interest for this review is ~~*V.*~~ *Vernonia amygdalina* (bitter leaf). It belongs to the Asteraceae family and grown or cultivated on the African continent in tropical regions such as Ghana, Nigeria, Cameroon and semi-tropical regions such as Zimbabwe, Lesotho, Mozambique and certain parts of KwaZulu-Natal in South Africa[38].

In ethnomedicine, leaf extracts of *V. amygdalina* have been used in Ghana and Nigeria for their immune stimulating activity in individuals with HIV or immunocompromised condition such as cancer or diabetes. Decoctions of leaves of *V. amygdalina* have been used in treatments and prevention of ailments including but not limited to piles, diabetes, kidney diseases, malaria,

hiccups, fever, high blood pressure, and stomach discomfort[9,39]. A review paper has been published which confirmed that *V. amygdalina* has ~~a~~ low or no toxicity[40].

Researchers have isolated numerous phytochemicals with potent bioactivity from leaves of *V. amygdalina* including sesquiterpene edotides, lactones, flavonoids: Luteolin-7-*O*-glucuronide, Luteolin 7-*O*-glucoside, and glycosides[38,41]. Luteolin and glycosides have been reported to have anti-microbial, antioxidant activities and in vivo biological activity as a cancer chemo preventive agent. [42].

Methods

Search strategy

Databases and specific keywords

Web of Science, Google Scholar, Pub-Chem, Pub-Med and Cochrane library electronic databases were explored for the period 2000-2018. Specific keywords used in carrying out the search included the following: use of *Persea americana* and *Vernonia amygdalina* in disease management, avocado, bitter leaf, ethnomedicinal use of *Persea americana* and *Vernonia amygdalina*, phytochemical constituents and pharmacological activities of *Persea americana* and *Vernonia amygdalina*.

Inclusion criteria applied to studies performed on the African continent that reported ethnomedicinal uses, phytochemical constituents and pharmacological activities of *Persea americana* and *Vernonia amygdalina.*

Exclusion criteria included publications in a language other than English, not related to our study and duplicates.

Selection of potential research papers

About three hundred eligible papers were produced according to the search benchmarks implemented via online out of which 142 made the list as presented in Figure 1 among which 92 articles/papers were used in the review findings, while the rest were used in the introduction and discussion.

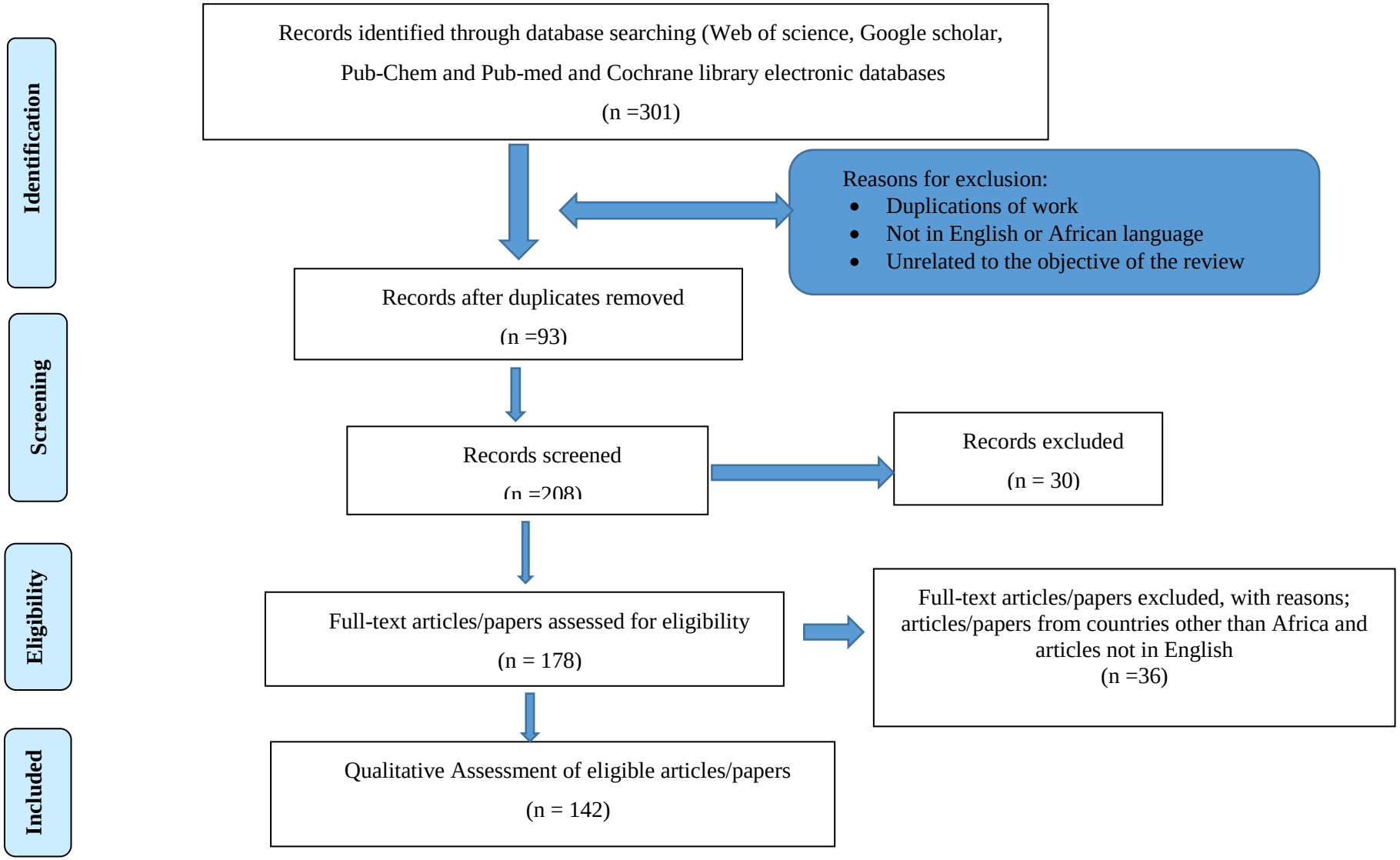

Fig. 1. Flow diagram showing the selection process of studies included in the literature

Analysis of data extracted from search criteria

Selected papers taken into consideration for this review were analyzed based on the following: Country, year of study, study design, number of participants, research period, ethnomedicinal use, treatment modalities/part used and/ or side effects of using *Persea americana* and *Vernonia amygdalina* therapy. Moreover, pharmacological action and phytochemical chemical constituents of the plant reported in various studies (Table 1, 2, 3, 4, 5, and 6) were taken into consideration.

Review findings

Ethnomedicinal uses for *Persea americana* and *Vernonia amygdalina*

Table 1 presents the findings from this review about leaf extracts of *Persea americana* which were commonly used in ethnomedical practices by local communities in African countries for the management of diabetes, hypertension, malaria and other diseases, without no documented side effects except for dizziness in the case of in the treatment of malaria in Ivory Coast[43].

Table 1. Ethnomedicinal use of *Persea americana* in the treatment of diseases on the African continent.

Research Time Frame	Country	Study Design	Sample Size	Local Name	Plants Part Used	Disease(s) reportedly treated in different local communities	References
NA	Ghana	Ethnomedicinal survey	NA	Paya ahaban	Leaves	Hypertension	44
April 2004	Kenya	Ethnobotanical survey	30	NA	Leaves	Diabetes	45
NA	Cameroon	Survey, questionnaire	137	NA	Leaves	Diabetes	18
NA	Cameroon	Cross-sectional, Self-structured questionnaire	200	NA	Seed and bark	Toothache	46
NA	Ivory Coast	Ethnobotanical survey	NA	Awuca, Pya, Afouca	Leaves	Malaria	43
April-May 2013	Uganda	Ethnobotanical survey	338	Ovakedo	Seeds	Diabetes	17
Sept.-Dec.2013	Ghana	Semi-structured interviews	40	Paya	Leaves	Malaria	19
Feb-June 2012	Nigeria	Ethnobotanical survey	50	Apoka	Leaves, root	Typhoid	47
Feb 2011-Dec 2012	Congo DR	Ethnobotanical survey	NA	Ivoka	Leaves	Diarrhea, Hypertension, Cough	23
NA	South Africa	Ethnobotanical survey	54	NA	Seed	Skin Diseases	13
May-Sept. 2012	Nigeria	Ethnobotanical survey	250	Pia	Leaves	Malaria	21
June 2009 to March 2010	Congo	Leaves and fruit Wines	47	Ivoka (mashi)avokati (Swahili)	Leaves and fruits	Constipation, Kidney, Various Pains	48
NA	Nigeria	Ethnobotanical survey	151	Igba, apoka	Leaves	Malaria	49
NA	South Africa	Ethnobotanical survey	52	Moafokhathe	Root	Diabetes	16
May-Sept 2007	Burkina Faso	Interviews	41	Avoka-tiiya	Leaves	Sedative	50
NA	Nigeria	Questionnaire, interviews	120	Pia, apoka	Leaves	Blood Pressure	51
March-June 2008	Nigeria	Semi structured questionnaires	104	NA	Stem bark and leaves	Malaria	20
Dec. 2005-July 2006	Cameroon	Ethnobotanical survey	412	NA	Bark	Syphilis	52
NA	Ivory Coast	Ethnobotanical survey	32	NA	Seed	Diabetes	53
Jan. 2007-April 2008	Nigeria	Questionnaire	NA	NA	Leaves	Hypertension	54
NA	Nigeria	Ethnobotanical survey	NA	NA	Dried leaves	Hypertension	24
Feb. 2002-Dec 2005	Guinea	Ethnobotanical survey	NA	Piya	Leaves	Anti-Bacterial	55

Table 2 presents the findings from this review about the ethnobotanical surveys and ethnomedicinal uses of *Vernonia amygdalina* in different local communities on the African continent. Leaf extracts of this plant have been used for the managements of various diseases such as malaria.

Table 2. Ethnomedicinal use of *Vernonia amygdalina* in the treatment of diseases on the African continent.

Research Time Frame	**Country**	**Study Design**	**Sample Size**	**Local Name Of *Vernonia amygdalina***	**Plants Part Used**	**Disease(s)**	**References**
September 2011-February 2012	Ethiopia	Ethnobotanical survey	189	Hecho	Leaves	Antimalarial	56
NA	Cameroon	Survey questionnaire	200	Bekantsu	Leaves	Toothache	46
March-September 2104	Nigeria	Ethnobotanical survey	NA	Ewuro/aromagbo	Leaves	Asthma	57
September 2011-March 2012	Nigeria	Ethnobotanical survey	10	Shiwaka	Leaves	Chickenpox, Hepatitis, Measles, Rabies	58
NA	Nigeria	Ethnobotanical survey	NA	Shiwaka	Stem/Root	Pile, diarrhea, High-Blood Pressure	59
NA	Nigeria	Ethnobotanical survey	NA	Ebe oboyiwo	Leaves	Diabetes	60
NA	Nigeria	Ethnomedical Survey	110	Ewuro	Leaves	Anti-aging	61
October 2010 - May 2011	Cameroon	Ethnobotanical Survey	108	Bekantsu	Leaves	Menstrual cramps	62
NA	Nigeria	Survey questionnaire	208	Ewuro	Leaves	Measles, Jaundice, Chickenpox	63
NA	Nigeria	Survey questionnaire	10	Shuwaka	Leaves	Yellow Fever, Hypertension, and Malaria	64

In vivo and In vitro studies on *Persea americana* and *Vernonia amygdalina* with pharmacological actions

Table 3 presents the findings of this review about the pharmacological activities of leaf extracts of *Persea americana in* various diseases. Most of the studies reported anti-microbial, anti-diabetic, antihypertensive and other pharmacological properties.

Table 4 presents the findings of this review about the pharmacological activities of leaf extracts of *Vernonia amygdalina* such as its anti-oxidant, anti-microbial and anti-diabetic properties.

Table 3. Pharmacological action of *Persea americana* on cell lines, animal models and human subjects.

Country	Study Design	Cell Lines, Animal Models And Human Subjects	Treatment Modalities	Pharmacological Action	Outcomes	References
Nigeria	In vitro	Clinical isolates (Bacillus cereus, Bacillus subtilis, Pseudomonas aeruginosa, Salmonella typhi, Staphylococcus aureus, Shigella flexneri, Escherichia coli, Candida albicans)	Leaf extracts	Anti-microbial	Inhibition of growth isolates	65
Nigeria	In vitro	alpha-amylase and alpha-glucosidase	Aqueous extract seeds	Anti-diabetic	Inhibition of key enzymes for carbohydrate digestion	66
Nigeria	In vivo	Mice	Ethanolic extract	Neuropharmacological	Depressant effect on CNS	67
Nigeria	In vivo	Wistar rats	Ethanol extract of seed	hepatoprotective and hemopoietic activity	The decrease in liver enzymes and an increase in hematological parameters	68
Ghana	In vitro	*Salmonella*	Leaf extract	Anti-microbial	Inhibition in the agar diffusion assay	69
Nigeria	In vivo	Albino Mice	Aqueous leaf extract	Activated partial thromboplastin	Increase in prothrombin time	15
Ghana	In vitro	Chloroquine-sensitive 3D7 *Plasmodium falciparum* and chloroquine-resistant W2 *Plasmodium falciparum* strains	Aqueous extract of leaves	Antimalarial	Antiplasmodial activity against the chloroquine-sensitive 3d7 ꝑ. *Plasmodium falciparum*	70
Nigeria	In vitro	Bacteria isolates	Methanolic leaf and bark extract	Antibacterial infections	Inhibition of growth of bacteria	71
Nigeria	In vivo	CCl4 induced damage male albino rats	Aqueous.extract	Hepatoprotective activity	Bradychardia, vasorelaxation, and hypotension	72
Cameroon	In vivo	Rats	Aqueous leaf extract of *P. americana*	Anti-hypertensive	Protection against liver and kidney damage as a result of increment of oxidative status by the extract	73
Nigeria	In vivo	Nonpregnant rat uterus	Aqueous extract of *P. americana* seed	Pre term labor	Effect on contractility of uterine smooth muscles	74
Nigeria	Clinical trials	Human	Liquid extract of leaf	Anti-hypertensive	Reduction in cholesterol levels in patients	75
Nigeria	In vitro	alpha-amylase and alpha-glucosidase	Phenolic leaf and fruit extracts	Antidiabetic	Inhibition of enzymes linked to type 2 diabetes	76
Nigeria	In vitro	Sorghum bicolor seed germinating rapidly	The methanol extract of leave and seed	Anti-tumor	Inhibition of germination of seed	77
Nigeria	In vitro	Pancreatic rats(Fe^{2+} induced lipid peroxidation)	Phenolic extract of leaf	The inhibitory effect on Fe^{2+} induced lipid peroxidation	Protect the pancreas from Fe^{2+} induced lipid peroxidation in vitro	76

Nigeria	In vivo	Wistar rats	Chloroform-methanol extracts of the seeds	Antispasmodic effect	Decreases in the gastro-intestinal motility	78
Nigeria	In vivo	Wistar rats	Fruit extracts	Cardio vascular disease inhibitor	Decreased total cholesterol	79
Nigeria	In vitro	MCF-7 cell line	Stem bark	Antiproliferative and pro-apoptotic activities	Lacks antiproliferative and apoptotic effects in mcf-7 cell line	80
Nigeria	In vivo	Diabetic Rats	Aqueous seed extract.	Anti-diabetic		81
Nigeria	In vitro	Cancer cells	Methanolic extracts	Cell proliferative activity	Anti proliferative estrogen receptor blood cancer cells	82
Nigeria	In vitro	HBAs and HbSS blood samples	Alkaline and alcoholic extracts	Anti-sickling	Reduction in the number of sickle cells	83
Nigeria	In vivo	Rats	Aqueous seed extract	Hypotensive	Reduced bp in normotensive beat rates	84
Nigeria	In vitro	*Escherichia coli, Klebsiella pneumonia, Bacillus subtilis, Streptococcus pyogenes,*	Seed extracts.	Anti-microbial	Inhibition of growth of microbes	85
South Africa	In vivo	STZ induced diabetic rats	Ethanolic leaf extracts	Anti-diabetic	Reduced urine flow and electrolyte excretion rate	86
Nigeria	In vivo	Rats	Leaf extracts	Anti-diabetic and hypocholesterolemic	Lower plasma glucose and influence lipid metabolism	87
South Africa	In vivo	Guinea pig(*Cavia porcellus*)	Aqueous leaf extract	Hypotensive	Caused bradychardia and vasorelaxation	88
South Africa	In vivo	Mice	Aqueous leaf extract	Anti-convulsant	Enhance gaba neurotransmission on the brain	89
South Africa	In vitro	Male Balb C mice (*Mus domesticus*)	Intraperitoneal injections aqueous leaf extracts	Anticonvulsant effect	Graded doses in mice gave an ld50 value of 1336±103mg/kg indicating non-toxicity in mice.	89
Nigeria	In vivo	Isolated thoracic rat aorta	Aqueous leaf extracts	Vasorelaxant	Vasoconstriction due to inhibition of ca^{2+}	36
Nigeria	In vivo	Albino Rats	Aqueous leaf extracts	Anti-ulcer	Anti-ulcer activity against experimentally induced gastric lesions	90

Table 4. Pharmacological action of *Vernonia amygdalina* on cell lines, animal models and human subjects.

Country	Study design	Cell lines, animal models and human subjects	Treatment modalities	Pharmacological action	Outcomes/side effects	References
Nigeria	In vitro	*Trichoderma* sp.	Chewing of stem	Anti-microbial Activity	Effective in inhibiting both bacterial and fungal infections.	91
Nigeria	In vitro	*Staphylococcus aureus* and *Pseudomonas aeruginosa* bacteria.	Inhibition by disk diffusion method as described by Kirby-Bauer diffusion technique	Anti-oxidant and antimicrobial activity	Significant antimicrobial activity	92
Nigeria	Prospective randomized, double-blinded, placebo-controlled clinical trial	40 HIV-infected patients on ART regime for at least 1 year with CD4 line 120-212	Oral:2 times daily of leaf extracts for 4 months	Immunological effect	Immune stimulating activity of leaf extracts and its combination with immunace®	93
Nigeria	In vivo	Albino Wister rats (*Rattus novergicus*)	Methanolic leaf extracts	Antioxidant activity	Leaf extracts contain ~~contains~~ phenolics and flavonoids and possess hepatoprotection effect against rat organ damage	94
Nigeria	In vivo	Albino Wistar rats(both sexes)	Gastric intubation in a 12 h cycle for 14 days	Anti-diabetic	Elevated blood glucose decreased compared to both normal control and alloxan treatment after 14 days.	34
Nigeria	In vivo	Beans weevil (*Acanthoscelide. obtectus*)	Seeds sprayed with different concentrations of the ethanol extracts	Insecticidal Properties	The average mortality indicated that 4.00% concentration resulted in higher toxicity of 33.60 in *Acanthoscelide. Obtectus*.	95
Uganda	In Vivo (4-Day Suppression Test) and (Rane Test)	Human patients	1 L of freshly made infusion daily to be taken as 250 mL, 4 times daily for 7 days	Anti-malarial	There was no evidence of significant side-effects or toxicity from the medication.	96
Nigeria	In Vivo	Clinical cross-sectional work comprising sixteen humans (8 female and 8 males)	Oral administration using the squeeze-wash-drink method.	Anti-diabetic	*V. amygdalina* elicited significant reductions ($p<0.05$) in blood glucose levels at most postprandial time points and for area-under-curve (AUC) values.	97

Phytochemical constituents of *Persea americana* and *Vernonia amygdalina* and their pharmacological actions

Table 5 presents the findings of this review about the phytochemical constituents and biological activities of leaf extracts of *Persea americana*. Most of the studies reported the presence of alkaloids, flavonoids, and saponins with largely anti-oxidant activity.

Table 6 presents the findings of this review about the phytochemical constituents and biological activities of leaf extracts of *Vernonia amygdalina* Most of the studies reported the presence of alkaloids, flavonoids, saponins, tannins, terpenoids, anthraquinones, and glycosides.

Table 5. Phytochemical constituents observed in *Persea americana* in African countries.

Country	Part Used	Phytochemicals Present	Outcomes	References
Nigeria	Seed	Tannin, total oxalate, phytate	Good source of nutrients	98
Nigeria	Leaves	Alkaloids, tannins, cardiac glycosides, anthraquinones, flavonoids, proanthocyanidins, phytates and saponins	Antioxidant activity	99
Nigeria	Seed	Alkaloids, flavonoids, tannins, saponins, Oxalates and phytates	Unsaturated fat good for heart	100
Nigeria	Seed	Saponins, phenols, tannins, flavonoids, alkaloids, steroids and glycosides	Antioxidant activity	101
Nigeria	Aqueous and chloroform leaf extracts.	Saponins, flavonoids, tannins and cardiac glycosides	Effect on tumor producing cells	102
Nigeria	Seed	Phenols, flavonoids and pro-anthocyanidin	Antioxidant activity	103
Nigeria	Leaves, seed and bark	**Leaves:** moderately high amounts of flavonoids, proteins, trace amounts of alkaloids, terpenes and tannins **Bark:** moderately high concentrations of alkaloids, flavonoids, saponins and trace amounts of proteins **Seed:** moderately high concentration of flavonoids, proteins, alkaloids and in trace amounts carbohydrate	Protease inhibitor	104
Nigeria	Leaves	Alkaloids, terpenoids, flavonoids, saponins, tannins phytates, oxalate.	Antioxidant activity	105
Nigeria	Leaf extracts	Alkaloids, tannins, flavonoids, cardiac glycosides, saponins, terpenes and steroids	Inhibitory activity responsible for anti-tumor related complications	106
Nigeria	Leaf extracts	Sterols, tannin, saponin, flavonoids, alkaloids, phenols, phlobatannins, anthraquinones, triterpenes, cardiac glycosides, phytates, oxalate	Antioxidant activity	107
Nigeria	Seed	Alkaloids, saponins, tannins, flavonoids, cyanogenic glycosides	Na	108

Table 6. Phytochemical constituents observed in *Vernonia amygdalina* in African countries.

Country	Part used	Phytochemicals present	Outcomes	References
Nigeria	Stem (ethanol extract)	Anthraquinones, tannins, flavonoids, terpenoids, saponins alkaloids	Ethanol extract showed higher inhibitory effects on most of the test organisms as compared with chloroform and ethyl acetate extracts	91
Nigeria	Leaves	Flavonoids, saponins, alkaloids, beta-carotene, ascorbic acid	Possible supplement in animal nutrition and it justifies their use in traditional medicines for the treatment of different diseases	109
Nigeria	Leaves	Tannins, phlobatanin, flavonoids, alkaloids, glycosides, cyanogenic, glycosides, anthraquinone, terpenoids, saponin polyphenols, cardenolide	Might be a source of active antimicrobial agents for the development of drugs for the treatment of these microorganisms.	92
Nigeria	Stem and root bark Extracts	Alkaloids, tannins, flavonoids, saponins, and hydrocyanide	The extracts serve a source of pharmacologically active.	110
Nigeria	Stem and root bark Extracts	Saponins, alkaloids, flavonoids, hydrocyanide and trace amounts of tannins	Source of pharmacologically active phytochemicals	110
Nigeria	Root extract	Alkaloids, glycosides Saponins, flavanoids Reducing sugar, steroids	Elevated blood glucose ($p<0.01$), compared to both normal control and pre-alloxan treatment value, Decreased ($p<0.01$)	34
Nigeria	Stem extract	Alkaloids, glycosides, flavonoids, Reducing sugar, polyphenols steroids	Elevated blood glucose ($p<0.01$), compared to both normal control and pre-alloxan treatment value, Decreased ($p<0.01$),	34
Nigeria	Stem extract	Alkaloids, glycosides Saponins, polyphenols, reducing sugar, steroids	Elevated blood glucose ($p<0.01$), compared to both normal control and pre-alloxan treatment value, Decreased ($p<0.01$),	34
Nigeria	Leaf extract	Alkaloids, glycosides, flavanoids Reducing sugar, polyphenols steroids, tannins hydroxymethyl anthraquinones	Elevated blood glucose ($p<0.01$), compared to both normal control and pre-alloxan treatment value, Decreased ($p<0.01$),	34
Nigeria	Leaf extract	Alkaloids, glycosides, flavanoids Reducing sugar, polyphenols steroids,	Elevated blood glucose ($p<0.01$), compared to both normal control and pre-alloxan treatment value, Decreased ($p<0.01$),	34
Nigeria	Leaf	Alkaloids, flavonoids, saponins, tannins Phlobatannins, terpenoids, cardiac glycosides	Average mortality showed that 4.00% concentration resulted in higher toxicity of 33.60 in *A. obtectus* while 3.00% showed toxicity of 29.93,	34

Discussions

Although progress and advances are made for the treatment of diseases with orthodox medicines, largely African patients resort to the use of ethnomedicine for solutions to treatments such as HIV/AIDS, cancer and diabetes[111,112]. In ethnomedicine on the African continent, medicines made from the leaves of plants are largely water-based organic decoction extract used as tonic drink[39,9].

This review found that *Persea americana* has been used in ethnomedicine in the African continent. In Nigeria, the use of leaves of *Persea americana* as a therapy for malaria by a population in Ogun state[20]. It has been reported the use of leaf extracts of *Persea americana* in the treatment of diabetes by a population in Nyeri county[45], while Tsabang *et al*., indicated the treatment of this same disease by people in Cameroon with leaves as a therapy[18]. This is in agreement with a research carried out in Latin American countries ~~in~~ which indicated the use of leaves by a population[113]. It is substantial to point out that there were not much reported side effects because of the use of *Persea americana* (whole or part) in the treatment of diseases across the African continent. This is an affirmation of the reason why the indigenous population in Africa resort to alternative medicine in the management of diseases. Limited side effects of ethnomedicine are one of the reasons for the greater use of herbal medicines among African ethnic communities. [114]~~.~~ An ethnobotanical survey conducted in Cameroon[16] and Kenya[45] revealed the use of leaves of *Persea americana* in the treatment of diabetes by patients.

In Trinidad and Tobago, an ethnobotanical interviews conducted from 1996–2000 with thirty male and female respondents concluded that *Persea americana* among other medicinal plants have been used for hypertension and diabetic treatment[115]. In Guatemala indigenous population consumption of the fruit as food and the use of different parts of the plant in the treatment of diseases such as diarrhoea[116]. *Persea americana leaves* are used in the treatment of diabetes in Latin America[113].

Vernonia amygdalina had a lot of health-promoting benefits for human use as well as the poultry industry. Leaf extracts of *V. amygdalina* had immune stimulating activity in individuals with HIV or immunocompromised conditions such as cancer or diabetes[40]. Leaf decoctions of *V. amygdalina* have been used in ethno medicine in the treatments of ailments such as diabetes, malaria, hypertension, fever, high blood pressure, kidney diseases, immunological effect, insecticidal properties and syphilis within the African community[9,39,117,] Leaf extracts of *V. amygdalina* in Nigeria[34] are used in ethnomedicines for treatment of diabetes; this is in agreement with Okolie *et al*.[97]. An ethnobotanical survey in Nigeria found that leaf extracts are used to treat the same disease[60]. This is confirmed by an ethnobotanical survey in Nigeria for

diabetic treatment with *Vernonia amygdalina* leaf extracts[118]. Aqueous leaf extracts of *V. amygdalina* are used in the treatment of malaria in Ghana. [119] This use is in agreement with report observations by Asnake *et al*, in Ethiopia[56] and Nigeria[120].

Pharmacological research carried out in Ghana indicated the use of leaves of *Persea americana* and as an antimalarial agent[19]. In vivo and in vitro results in Table 3 justify the reason why indigenous folks in African communities lean on a specific part of *Persea americana* in the treatment of certain kind of disease. Hypoglycemic effect of *P. americana* leaf extracts in rats. Observed data revealed after this same study indicated the reduction of plasma glucose in the animals. In a similar study conducted in South Africa, ethanolic leaf extract indicated a hypoglycemic effect on Streptozotocin (STZ) induced diabetic rats.[121]. A reduction in urine flow and electrolyte excretion rates with the animals were observed[122]. An in vitro study in Mexico on antioxidant activity against 2,2′-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)(ABTS) radical of peel, pulp and seed ethyl acetate, acetone, and methanolic extracts of *Persea americana* , concluded that there are natural source phenolics with high antioxidant and antimicrobial potential[123].

Interestingly, there is a linkage between ethnomedicinal use of leaves of *P. americana* and pharmacological actions reported in Tables 1 and 3 that reveal their use in the treatment of diabetes in African communities.

In pharmacological activities, reported antioxidant activity of *V. amygdalina* leaf extracts in vitro with *Staphylococcus aureus* and *Pseudomonas aeruginosa* bacteria[35,92]. Observed data revealed indicated the reduction of plasma glucose in the animals by the use of *V. amygdalina* leaf extracts[34]. Clinical cross-sectional work on humans[97] as well as in vivo in albino Wistar rats[34] (both sexes) in Nigeria indicated the treatment of diabetes using leaf extracts of *V. amygdalina.* These results are in accord with Osinubi, (2007) who reported that aqueous leaf extracts of *V. amygdalina* indicate a substantial blood-glucose-lowering potential in both normoglycaemic and alloxan-induced diabetic male Sprague-Dawley rats comparable to that in chlorpropamide-treated rats[124]for its anti-diabetic activity[124]. An in vitro study in Nigeria using *Staphylococcus aureus* and *Pseudomonas aeruginosa* bacteria reported anti- oxidant activity of leaf extracts of *V. amygdalina*[92]. The anti-oxidant activity of aqueous and methanolic leaf extracts of *V. amygdalina* as reported in an in vitro assay[125] is in agreement with Johnson *et al.*[92] In Nigeria in an in vivo study, involving albino rats (*Rattus novergicus*) orally administered with methanolic leaf extracts showed anti- oxidant activity[94]. The result of in vitro *Trichoderma sp* assay in Nigeria indicated that leaf extracts and the chewing of the stem of *V. amygdalina* have anti-microbial activity, which is effective in inhibiting both bacterial and fungal

infections[91,126]. An in vitro study using *Ascaris suum* conducted in Uganda among pastoralist communities showed that ethanolic and water leaf extracts of *V. amygdalina* A study on an in *vitro* study against *Ascaris suum* that ethanolic and water extracts of *V. amygdalina* have anthelmintic activity in the cattle.[127] In Malaysia presented an in vivo study on Nulliparous and non-pregnant female SD rats (Rattus norvegicus), with a single oral doses of *V. amygdalina* aqueous extract at progressive dose of 175 mg/kg, 550 mg/kg, 2000 mg/kg and 5000 mg/kg of Body Weight (BW) showed antioxidant activity[35]. They further indicated that the presence of flavonoids have a potent antioxidant together with terpenoids may have contributed to the lack of direct organ toxicity by free radical scavenging activities. *V. amygdalina* have antihyperglycemic effect; polyphenols in the extract may be the candidates that are responsible for the above-mentioned biological activities[128]. A review paper indicated a polyherbal formulation in Ghana containing only *P. americana* and *V. amygdalina* is under in-vivo evaluation for anti-hypertensive[129]. The reported phytochemicals present in *P. americana* are in agreement with what other researchers reported in different parts of the globe[31,33,130,131]. The abundance of these phytochemicals in *P. americana* contribute to biological activities when applied in humans. The presence of two glucosylated abscisic acid derivate in *P. americana* seed could help improve symptoms of type 2 diabetes[32]. The potential of triterpenoid of *P. americana* seed as an anticancer agent is being investigated[14]. The growth inhibitory effects exhibited by leaves *of P. americana* examined on the radicle length of guinea corn bicolor seed make it a potential antitumor agent and have appreciable phytochemical constituents as indicated in Nigeria but exhibited low levels of toxicity[105]. The presence of two glucosylated abscisic acid derivate in *P. americana* seed could help improve symptoms of type 2 diabetes[32]. The potential of triterpenoid of *P. americana* seed as an anticancer agent is being investigated. The growth inhibitory effects exhibited by leaves *of P. americana* examined on the radicle length of guinea corn bicolor seed make it a potential antitumor agent[106]. It has been further reported that *P. americana* has a rich mineral content and recommended it to be part of human and animal foods formulations[105]. However, veterinarians must not recommend leaves of *P. americana* as a feed ration for horses. Natural instances of avocado poisoning were reported in Australia when horses were fed on fruits and leaves of the plant. Mastitis was the condition observed in lactating Arabian mores after the incident[132].

.*Vernonia amygdalina* contain medicinally important bioactive compounds that show inert potentials for use as a possible supplement in animal nutrition and it justifies their use in traditional medicines for the treatment of different diseases[109]. A large number of researchers have isolated numerous phytochemicals with potent bioactivity from leaves of *V. amygdalina* including sesquiterpene edotides and lactones, flavonoids: Luteolin-7-*O*-glucuronide, Luteolin

7-*O*-glucoside, and glycosides[38,41]. Phytochemicals have been isolated from leaves of dried avocado that yielded isorhamnetin, luteolin, rutin, quercetin, and apigenin[133]. The previous studies had demonstrated that vernodalin from *V. amygdalina* leaf have antitumor activity Luteolin-7-*O*-glucuronide (the most abundant compound) have antioxidant activity[40].

A preliminary phytochemical screening conducted in Nigeria of ethanolic stem extracts of *Vernonia amygdalin* yielded anthraquinones, tannins, flavonoids, terpenoids, saponins.[91] Th is in contrast with investigations conducted by Atangwho *et al*. (2010), that indicated the presence of alkaloids, glycosides, flavonoids, reducing sugar, polyphenols, and steroids. Although aqueous stem extracts have alkaloids, glycosides, saponins, polyphenols, reducing sugar, steroids[34]. Stems and root barks extract analysis found alkaloids, tannins, flavonoids, saponins, and hydrocyanide present[110]. This result obtained is in line with Atangwho *et al*. (2010) in the ethanolic and aqueous root extracts of *V. amygdalin* obtaining alkaloids, glycosides saponins, flavonoids, reducing sugar and steroids[34]. The same researchers reported that ethanolic leaf extracts of *V. amygdalina* had alkaloids, glycosides, flavonoids reducing sugar, polyphenols, steroids, tannins hydroxymethyl anthraquinones, while the aqueous leaf extracts of the same plant produced alkaloids, glycosides, flavonoids, reducing sugar, polyphenols, and steroids[34]. Alkaloids exhibit analgesic and antispasmodic activities while tannins are able to eliminate protein deficiency syndrome known as "kwashiorkor''. [105] Further to some of these activities, it has previously been established that tannins have wound-healing property in humans. [134]. Flavonoids have good antioxidant potential that helps with the reduction of oxidative stress[135]. Saponins serve as natural antibiotics for the treatment of fungal infections[100]. Phenols and proanthocyanidin are such a good source of harnessing antioxidants[103].

The differences in the types of phytochemicals may be due to the geographical location of the plants. Geographical factors may influence the presence of primary and secondary metabolites found in herbal medicines[136]. Sampaio *et al*. (2016), further argues that variations are very useful in the chemical characterization of plants of the same species collected from different regions. Due to environmental factors, species of the same plant but growing in different environmental conditions may have substantial differences and/ or similarities in the production and accumulation of primary and secondary metabolites caused by chemical interactions between plants and the environment. Processes of variations involved long term acclimation, seasonal differences related to phenology or environmental changes in the biotic and abiotic factors, geographical differences involving different populations (genetic differences within a plant species), or different environmental conditions of the growth location of the species individuals[136]. Environmental factors had an influence on the types and contents of

phytochemicals in plants[137]. Liu *et al.* (2016) indicated that altitude and annual mean temperatures were significantly and negatively correlated to the tannin content and the content of total phenolics respectively. While annual sunshine duration and altitude were significantly and positively correlated to the flavonoids, rutin content, and antioxidant activity. In addition, altitude was significantly and positively correlated to the content of total phenolic [137].

Implications of findings for future research into *Persea americana* and *Vernonia amygdalina*

The use of *Persea americana* and *Vernonia amygdalina* as food and in the treatment of diseases by people on the African continent emphasize the significance of these plants. However, standardization of the whole plant is essential to evaluate the safety and efficacy of using the plant and phytochemical evaluation is key towards achieving that goal[138].

Limitations of this review

This review is about research-based studies on *Persea americana* and *Vernonia amygdalina* within the African continent reported in the 21st century. Studies included in this narrative review were centered on recognized quality of valuation since this was not a systematic review, which solely relies on expert judgment. Identified articles retrieved from diverse kinds of journals included both quantitative and qualitative work. A bias towards articles published in English was taken into consideration. In terms of publications on *Vernonia amygdalina* more recent publishers (between 2010 and 2018) were used due to the reason that over hundreds of papers were retrieved and most had similar outcomes and researched in Nigeria.

Conclusions

Various drugs have been produced and used in the national and international market by following the traditional system and its ethnopharmacological application. The traditional system of medicine remains the most ancient yet living traditions for the treatment of various diseases. The increased popularity of alternative medicine can be attributed to the emergence of a number of factors. Primarily, most people have resorted to the use of alternative medicine in the belief that they stand to accrue benefits that are not available in conventional medicine and related therapeutic interventions.

Findings from this review revealed that *P. americana* and *V. amygdalina* are rich in nutrients and phytochemicals and therefore not only good for human consumption but for treatment of ailments as well. Traditional healers have an unmistakable and crucial role to play in building the health system in South Africa and strengthening and supporting the national response to different disease conditions. Ethnomedicinal uses, and pharmacological properties of leaf

extracts of *P. americana* and *V. amygdalina* may justify polyherbal formulation involving the two plants in the treatment of diseases such as diabetes and HIV/AIDS.

Further studies are needed to determine the chemistry of polyherbal mixtures claiming to contain leaf extracts of *Persea americana, Vernonia amygdalina* and /or other medicinal plants. In addition, the chemical composition of these plants found in different geographical areas needs to be investigated.

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CHAPTER 3: MANUSCRIPT 2

"A Comparative Chemistry of COA®Herbal Medicine and Leaf Extracts of *Vernonia amygdalina* (bitter leaf) and *Persea americana (*Avocado*)* collected from Ghana and South Africa"

Prepared according to the South African Journal of Chemistry

This chapter, following the full review paper is presented on the phytochemical compositions of COA® herbal medicine and crude leaf extracts of *Vernonia amygdalina* (bitter leaf) and *Persea americana (*Avocado*)* titled **"A comparison of phytochemical composition of COA®herbal medicine and crude leaf extracts of *Vernonia amygdalina* (bitter leaf*)* and *Persea americana (*Avocado*)* collected from Cape Coast, Ghana and Durban, South Africa"** was prepared, submitted and under review following the guidelines of the **South African Journal of Chemistry.** The paper indicated that COA® herbal medicine contains leaf extracts of *V. amygdalina* and *P. americana* and that geographical and environmental factors have profound effect on the phytochemicals present in leaf extracts of the two plant species from Ghana and South Africa.

A comparison of the phytochemical composition of COA®herbal medicine and crude leaf extracts of *Vernonia amygdalina* (bitter leaf) and *Persea americana (*Avocado*)* collected from Cape Coast, Ghana, and Durban, South Africa

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ABSTRACT

Centre of awareness (COA®) herbal medicine is made from six different leaf extracts collected from two different locations, namely Ghana (GH) and South Africa (SA). Out of these leaf extracts, two were selected, i.e., *Vernonia amygdalina* (VA) and *Persea americana* (PA). The aim of this study is to investigate the phytochemicals in four different solvents of extracts of COA® compared to leaf extracts of VA and PA collected from GH and SA by phytochemical screening techniques and Gas Chromatography-Mass Spectrometry (GC-MS).

Outcomes of the preliminary phytochemical screening showed a positive result for alkaloids, anthraquinones, saponins, flavonoids, tannins, terpenoids as well as cardiac glucosides in ethanolic leaf extracts and standard COA®. GC-MS study was carried out in four solvents (dichloromethane, hexane, ethanol and ethyl acetate) and standard COA®. The finding, of this study, confirms the presence of VA and PA leaf extracts in COA®. The major phytochemicals common to COA® extract and leaf extracts of VA and PA were heneicosane, phytyl acetate, pyrene, octadecanoic acid, eicosane, 2-methyltetracosane, pentadecanoic acid, hexadecanamide, and octadecanamide. Most of these major phytochemicals are present in ethanolic extracts of both COA® and leaf extracts of VA and PA. Leaf extracts of medicinal plants collected from Ghana have more phytochemicals compared to that of South Africa. The variation in phytoconstituents can be ascribed to the plants' geographical area and environmental factors.

Keywords: COA® herbal medicine; *Vernonia amygdalina*; *Persea americana*; phytochemicals; GC-MS analysis.

1. Introduction

The Africa continent is enormous, bestride by the equator and endowed with more than 50,000 distinct known plant species in the sub-Saharan African region alone.[1] The continent of Africa has lavish cultures and traditions, which are valuable and protected with pride and to preserve the cultural heritage in a swiftly changing society. Herbal medicine practices are parts of the traditions of the people on the African continent. Traditional African Medicine (TAM) is probably the oldest, and most assorted, of all traditional medicine systems (TMS).[2] On the African continent, people have used or continue to use traditional medicines for the management or treatment of diseases in both humans and livestock. The most preferred part of medicinal plants for treatments in TAM is the young succulent, green and fresh leaves, although other parts such as the roots, stem, and flowers are used in TAM. Medicines made from the leaves of plants are largely water-based organic decoction extracts used as tonic drinks. [3,4]

Traditional preparations of the *Vernonia amygdalina* and *Persea americana* are decoctions and infusions of leaves, stems, twigs and/ or roots. Yeap *et al*. (2010) reported that *V. amygdalina* had a lot of health-promoting benefits for human use as well as the poultry industry.[5].[5] Root decoctions of *P. americana* have been reported to be used in treating diabetes in South Africa[6]. In Cameroon and Kenya, the management of diabetes makes use of leaf decoctions. [7,8]. Leaf infusions of the same plant were used in treating diarrhea, hypertension and cough in the Democratic Republic of Congo[9] as well as malaria in Ghana[10], Ivory Coast[11], and Nigeria[12-14].

The Centre of Awareness (COA), a non-governmental organization, based in Cape Coast, formulated the COA® (Centre of Awareness) herbal medicine, which contains *Vernonia amygdalina* (VA) (Bitter leaf), *Persea americana* (PA) (Avocado), *Azadirachta indica Carica papaya. Spondias mombin* and *Ocimum viride* (FDA/DRID/HMD/HMU/16/0981, 2016) amongst which VA and PA were used for the present study. The COA® has been ordered online by several patients as immune boosters against human immunodeficiency virus/Acquired Immune Deficiency Syndrome (HIV/AIDS) and as treatments in diabetes mellitus, cancer and high blood pressure (https://www.coadrugs.org).

In spite of advancements in treating diseases with orthodox medicines, numerous African patients resort into the utilization of TM for basic health care.[15,16] For decades, plants that make the composition of COA® have been used by local communities in Africa for the management of diseases.[5,6] Numerous phytochemicals with potent bioactivities have been isolated from leaves of *V. amygdalina* including sesquiterpenes, edotides, flavonoids: luteolin-7-*O*-glucuronide, luteolin 7-*O*-glucoside, and glycosides. [17, 18] Owolabi *et al*. (2005) isolated phytochemicals from leaf extracts of *P. americana* that yielded isorhamnetin, luteolin, rutin,

quercetin, and apigenin. Identified chemical compounds of *V. amygdalina* extracts, examined by gas chromatography-mass spectrometry (GC-MS) includes aliphatic acidic, esters, terpenes, diterpene alkanols and phytostanol in aqueous, methanol and petroleum ether extracts.[19] Phytol was the principal diterpene alcohol~~s~~ in methanolic and petroleum ether extracts.[20] Hydrocarbons (heneicosane, octadecane, eicosane, docosane, tricosane, tetracosane, and squalene), with sterols such as sitosterol and stigmasterol derivatives, were identified in fruit and seeds extracts of *P. americana*.[21] *P. americana* is rich in βeta-sitosterol and it decreases cholesterol.[22] *P. americana* fruit extract contains vitamin E. Vitamin E is needed in the body to the maintenance of an overall health.[22]

Alkaloids are heterocyclic nitrogen compounds. Morphine is an opiate alkaloid isolated from *Papaver somniferum*, whereas, the popular codeine and heroin are derivatives of morphine. Diterpene alkaloids have antimicrobial properties while other alkaloids might be valuable against HIV and intestinal contaminations related ~~with~~ to AIDS.[23] Alkaloids have antidiarrheal effects. The antidiarrheal activity could be the effect of travel time in the small digestive tract. Berberine is an aromatic member of the alkaloids with a planar orientation that has antidiarrheal activity-linked to ~~its~~ deoxyribonucleic acid (DNA) intercalating ability.[23]

Due to diverse medicinal importance of the COA®, there is a need to identify different phytochemicals in both COA® and crude extracts of *V. amygdalina* and *P. americana* found in Cape Coast (Ghana) and Durban (South Africa) using phytochemical screening methods and GCMS analysis. It has been reported in the literature that *V. amygdalina* and *P. americana* contains tannins, polyphenols, flavonol, terpenoids, and alkaloids. Cowan *et al*.(1999), in a clinical microbiology reviews on plant products as antimicrobial agents, reported that it is advisable to use ethanol as a solvent to carry out the preliminary phytochemical screening due to its nontoxicity, low evaporation temperature and also act as a preservative in medicinal plant extracts suspected to contain tannins, polyphenols, flavonol (flavonoids), terpenoids and alkaloids. [23, 24]

The least complex bioactive phytochemicals are phenols and phenolic acids consisting of a phenolic ring. Polyphenols are largely effective against viruses, bacteria, and fungi. [23] A few authors suggested that phenolic toxicity is directly proportional to the sum total of hydroxyl groups present. Therefore, increase~~d~~ in the sum total of hydroxyl groups, increases the inhibitory properties of phenols. Flavonoids are hydroxylated derivatives of phenolic compounds and have antimicrobial properties. Polymeric phenolic compounds found in almost every part of medicinal plants are tannins. Tannins are antimicrobial and are derivatives of flavan through condensation polymerization of quinone units. Another important phytochemical whose

mechanism of action is not well explained is a class of compound with a general chemical structure of $C_{10}H_{16}$ called terpenes. Terpenes occur as hemiterpenes, sesquiterpenes, diterpenes, triterpenes, and tetraterpenes (n = ½, 1½, 2, 3, 4. where n = number of carbons in the general structure = C_{10} as C_5, $C_{15,}$ C_{20}, C_{30}, and C_{40}). [23] Terpenes that have oxygen and /or other elements as additional elements are known as terpenoids. Terpenoids are antibacterial, antifungal and antiviral and are largely synthesized from acetate units originating from fatty acids but differ from fatty acids due to the extensive branching and cyclization of the chains. [23]

The geographical area of the leaf extracts used in the preparation of the COA® may have differences and/or similarities in their phytochemicals responsible for the pharmacological activities of the COA®. The aim of this study is to investigate the phytochemicals in four different solvent extracts of COA® compared to leaf extracts of *V. amygdalina* and *P. americana* collected from Cape Coast (Ghana) and Durban (South Africa) by phytochemical screening techniques and gas chromatography-mass spectrometry (GC-MS) analysis.

2. Experimental

2.1. Collection of Plant Materials, Reagents and Instrumentations

Twenty-four bottles of COA® (6000 mL) were purchased on the Durban market. Medicinal plants, *V. amygdalina* (bitter leaf) and *P. americana* (avocado) were collected from Pinetown (coordinates:-29.8224146, 30.8453970), Port Shepstone (coordinates:-30.7373037, 30.4387322) Durban-South Coast in South Africa respectively (03/03/2018 at 10:30 Central African Time, average temperature of 22 °C, average, humidity of 79% and average pressure of 1014 mbar). Medicinal plants from Cape Coast (Coordinates: 5.114467,-1.287032) Central Region of Ghana were collected on 17/08/2018 at 10:30 am Central African Time with an average temperature of 25 °C, average humidity of 80% and the average pressure of 1009 mbar. The plants were identified and authenticated at the Herbarium in the School of Life Sciences, at University of KwaZulu-Natal - Westville Campus.

All chemicals and solvents used in this research work were of analytical reagent grade (AR grade) and purchased from Sigma Aldrich and Merck Millipore, South Africa. Hexane and ethyl acetate were purchased from Associated Chemical Enterprises. Dichloromethane and ethanol were purchased from Sigma Aldrich, South Africa. Whatman filter paper was purchased from Sigma Aldrich, South Africa. Different instruments such as Rotary evaporator (Büchi-Germany), Analytical balance (Lasec, South Africa) and Magnetic Stirrer (Scientech ultrasonic, China) were used.

2.2. Methodology

2.2.1. Study Design

Experimental procedures in this research were carried out after the collection of materials in two phases, namely preliminary phytochemical screening and GC-MS analysis. Preliminary phytochemical screening was carried out using the methods as reported by Iqbal *et al.* (2015). GCMS analysis described by Alara *et al.* (2018) was used.[25,26] These experiments were conducted on COA® extract and leaf extracts of *V. amygdalina* and *P. americana* collected from Cape Coast, Ghana (Coordinates:5.114467,-1.287032) and Durban, South Africa (coordinates:-30.7373037, 30.4387322). This study was conducted in the Medicinal Chemistry laboratory at the School of Pharmaceutical Sciences, UKZN, Westville and the Mass Spectrometry Laboratory, School of Chemistry, UKZN, Pietermaritzburg.

2.2.2. Drying, Size Reduction and Extraction Procedure

The fresh leaves of *V. amygdalina* (bitter leaf) and *P. americana (*~~A~~vocado*)* were cleaned from dirt and unwanted pathogens separately with distilled water. The leaves were air-dried, at room temperature and pulverized to a coarse-to-fine powder. Leaf powder (30g) was weighed using an analytical balance for each medicinal plant and placed in a 250 mL conical flask that was subsequently sealed. Serial exhaustive extraction method involving successive extractions with solvents of different polarities namely hexane, dichloromethane, ethyl acetate and ethanol at gas chromatography grade (GC grade) were used. Each leaf powder was successively extracted with 250 mL solvent twice. A total volume of solvent used for each plant leaf powder was 500 mL. This was to ensure that a range of compounds with varying polarities was extracted.[27] Each solvent extract was filtered using a Whatman filter paper, concentrated and dried using a rotary evaporator and preserved in the refrigerator for future use in phytochemical screening and GCMS.[28]

2.2.3. Solvent (liquid-liquid) Extraction

Non-volatile components of a liquid can be extracted using solvent extraction. [29] The COA® herbal medicine was prepared in an aqueous medium, by fermentation of all medicinal plant leaves and distilled by selective boiling and condensation. The COA® extract was partitioned between less polar (hexane, dichloromethane) and more polar (ethyl acetate, ethanol) organic solvents. This was to allow a considerable separation of the non-polar chemical constituents of COA® from the polar ones. In terms of the more polar solvents used for extraction, ethanol was the preferred polar solvent to aqueous because water is too polar and will not extract non-polar substances. Although both methanol and ethanol are polar molecules, the high electronegative oxygen in

ethanol permits hydrogen bonding to take place with other molecules, thereby, attracting polar and ionic molecules. The non-polar property of the ethyl (C_2H_5) group in ethanol permits the attraction of non-polar molecules. Thereby, ethanol that is least toxic of the alcohols can attract both polar and non-polar molecules.[30]

2.2.4. Phytochemical Screening

For the successful determination of biologically active chemical components, there is a need for proper use of solvents in the extraction procedure. Ncube *et al*. (2008)[31] suggested that some characteristics of a good solvent for plant extractions may include toxicity, rapid evaporation at low temperatures, promotion of increased physiologic absorption of the extract, preservative properties and the ability of solvent not to cause the extract to complex or dissociate.[31]

Tests were carried out (in triplicates) according to standard procedures as reported by Iqbal *et al*. (2015) [25] and Sengar *et al*. (2015).[32] Qualitative analysis to determine the presence of alkaloids, steroids, flavonoids, saponins, terpenoids/triterpenoids, tannins, anthraquinone derivatives, and cardiac glycosides were carried out following standard procedures on standard COA® herbal medicine and ethanolic leaf extracts of *V. amygdalina* and *P. americana* . One milliliter each of COA® herbal medicine and ethanolic leaf extracts of *V. amygdalina* and *P. americana* collected from Ghana and South Africa were measured into 5 different test tubes that were used as controls.

2.2.4.1. Test for Saponins (Frothing test)

One milliliter each of COA® herbal medicine extract and ethanolic leaf extract of *V. amygdalina* and *P. americana* (Ghana and South Africa) were taken into 10 different test tubes out of which 5 test tubes ~~act~~ acted as controls. To each 1.0 mL of extract,-10 mL of distilled water was added. The mixture was agitated for 15 min. Formation of a layer of foam indicated the presence of saponins.

2.2.4.2. Test for Tannins (Ferric chloride test)

One milliliter each of standard COA® herbal medicine extract and ethanolic leaf extracts of *Vernonia amygdalina* and *Persea americana* were measured into 5 separate test tubes and 1 mL of ferric chloride (5% $FeCl_3$ in 95% ethanol) was added and the mixture was shaken. Formation of a blue-black precipitate indicated a positive result for tannins.

2.2.4.3. Test for Anthraquinones Derivatives (Borntrager's test)

One milliliter each of standard COA® herbal medicine extract and ethanolic leaf extracts of *V. amygdalina* and *P. americana* were measured into 5 different test tubes. One milliliter of 32% ammonia was added with agitation for a few seconds. The appearance of pink-red color in the lower ammoniacal layer indicated the presence of anthracene derivative.

2.2.4.4. Test for Cardiac Glycosides (Keller Killian test)

The two-milliliter volume of Standard COA® extract and 2.0 mL ethanolic leaf extracts of *V. amygdalina* and *P. americana* were measured into 5 separate test tubes. To this 2.0 mL of chloroform, 1.0 mL acetic acid and 1.0 mL ferric chloride solution was added~~,~~ Dropwise 1.0 mL concentrated H_2SO_4 was added on the inner walls of the test tube. Appearance of a brown ring at interphase with an upper green-blue coloration shows the presence of glycosides.

2.2.4.5. Test for Alkaloids

(a) Meyer Test

Standard COA® extract of volume 1.0 mL and 1.0 mL ethanolic leaf extracts of *V. amygdalina* and *P. americana* (Ghana and South Africa) were measured into 5 different test tubes. To these test tubes, 2 mL of Meyer's reagent was added and agitated slightly. Extracts in five other test tubes were used as controls. A light yellowish precipitate confirmed the presence of alkaloids.

(b) Wagner Test

To 1.0 mL each of standard COA® extract and ethanolic leaf extracts of *V. amygdalina* and *P. americana* were measured in 5 separate test tubes. Two milliliters of Wagner Reagent (1.271 g iodine + 2.001 g potassium iodide) was added and shaken. Formation of a brown precipitate indicated a positive result for alkaloids.

(c) Acid Tests (Litmus Paper)

This was a confirmatory test for alkaloids in standard COA® herbal medicine extract as the Meyer test had very light yellowish precipitate. A Universal litmus paper was dipped in 1.0 mL standard COA® extract. The litmus paper showed acidity of the COA® extract. This confirms that the extract contains alkaloids.

2.2.4.6. Test for Terpenoids (Liebermann-Burchard test)

Standard COA® extract of volume 1.0 mL and 1.0 mL ethanolic leaf extracts of *V. amygdalina* and *P. americana* was measured into 5 separate test tubes. To these 3 drops of acetic anhydride were added. Dropwise, 1.0 mL concentrated H_2SO_4 were added dropped onto the inner walls of

the test tube slowly and the mixture left to stand for a few minutes. A brown ring at the interface and a green color indicated the presence of terpenoids.

2.2.4.7. Test for Flavonoids (Sulfuric acid tests)

To 1.0 mL of standard COA® extract and ethanolic leaf, extracts of *V. amygdalina* and *P. americana* were measured into five separate test tubes. 1.0 mL of concentrated H_2SO_4 was added and shaken vigorously. A deep yellowish to reddish color after a few minutes indicated the positive result of flavonoids.

2.3. Characterization of the Various Phytochemicals in COA® Herbal Mixture and the Plant Leaf Extracts

Gas chromatography-mass spectrometry (GC-MS) technique was used for the characterization of the various volatile and non-volatile compounds present in the solvent leaf extracts of *V. amygdalina* and *P. americana*.

The chemical compound identification and retention times of *V. amygdalina* and *P. americana* leaf extracts from Cape Coast-Ghana and Durban- South Africa were evaluated using GC-MS analysis. GC-MS analysis was conducted according to procedures as described by Alara *et al*. (2018). [26]

Approximately 30 g powder of each of *V. amygdalina* and *P. americana* were extracted successively for 6 hours per session with GC grade solvents of dichloromethane (DCM), ethanol (EtOH), hexane (HEX), and ethyl acetate (EtOAc) and filtered using Whatman filter paper. Standard COA®, COA® extract from the four different solvents and leaf extracts from the four different solvents of *V. amygdalina* and *P. americana* (Ghana and South Africa) extracts were concentrated using a Rotary evaporator. Each sample was loaded in GC-MS compatible vials and analyzed.

Specifications: Analysis by GC–MS using Perkin (Emler) Gas Chromatography (Clarus 580) equipped with MSD mass spectrometer (Clarus SQ8S) instrument with the built-in auto sampler.

Column: Elite-5MS (30 m x 0.25 mm id x 0.25 μm). The oven temperature programmed from 37 to 320°C at a rate of 18-25°C/min and held for 0.5 and 1.85 mins at 18 and 320 °C respectively. The injector temperature: 250°C and MS Ion Source temperature: 280°C with full scan and solvent delay of 0–2.30 min. MS Scan Range was m/z 35 – 500 in 0.10 sec. One microlitre of the samples injected in Helium carrier gas at a split flow of 20 mL/min.

2.4. Data Analysis

All the experimental analysis were carried out according to the standard experimental procedures to ensure the validity and accuracy of the experimental work. Results from GC-MS of the chemical compounds obtained were checked for duplications for phytochemical structures and /or names and contaminants from the GC-MS columns were identified and eliminated from the study. Data are categoried into similar and /or different phytochemical structures and /or names between COA® and leaf extracts of plants collected around Cape Coast-Ghana and Durban- South Africa. In addition to the identification of similar and /or different phytochemicals by compound names, retention time of the phytochemicals were used in the analysis of similarities and /or differences between standard COA®, COA® in the four solvents and leaf extracts in all four solvents of plants collected around Cape Coast-Ghana and Durban- South Africa.

3. Results and Discussions

3.1. Preliminary Phytochemical Screening of Ethanolic Leaf Extracts (*V. amygdalina* and *P. americana*) and standard COA® Herbal Medicine

Phase one consisted of a qualitative chemical examination (done in triplicate but since results were similar it is recorded as one) of different ethanolic extracts of *V. amygdalina* and *P. americana* to determine the presence of alkaloids, anthraquinone derivatives, saponins, flavonoids, tannins, terpenoids and cardiac glucosides as demonstrated in **Table 1.**

In this investigation, the class of phytochemical detected in the ethanol leaf extracts of *V. amygdalina* and *P. americana* were alkaloids, anthraquinone derivatives, saponins, flavonoids, tannins, terpenoids, and cardiac glucosides. Result of phytochemical screening and insecticidal activity of ethanolic extracts of *V. amygdalina* revealed the presence of alkaloids, tannins, phlobatannins, flavonoids, terpenoids, cardiac glycosides, and saponins, which is in line with the results obtained in this investigation.[33] Findings from fruit extract of *P. americana* indicated the presence of alkaloids, flavonoids, terpenoids, and cardiac glycosides but tannins and saponins were absent.[34] An ethanolic leaf extract of *P. americana* in a study by Kumala *et al.* (2013) indicated the presence of flavonoids, coumarin, and saponins but the absence of alkaloids and tannins.[35]

Table 1 Preliminary phytochemical screening of COA® herbal medicine extract and different ethanolic extracts *P. americana* and *V. amygdalina* collected from Ghana and South Africa.

Class of phytochemicals	**Tests performed**	**Ethanolic leaf extracts**				**COA® (Standard)**
		***P. americana*- Ghana**	***P. americana*- South Africa**	***V. amygdalina* Ghana**	***V. amygdalina* South Africa**	**Solution**
Alkaloids (n=3)	Meyer	+++	+++	+++	+++	+++
	Wagner	+++	+++	+++	+++	+++
	Acid tests	NA	NA	NA	NA	+++
Anthraquinones Derivatives (n=3)	Borntrager's	+++	+++	+++	+++	+++
Saponins (n=3)	Frothing	+++	+++	+++	+++	+++
Flavonoids (n=3)	Sulfuric acid	+++	+++	+++	+++	+++
Tannins (n=3)	Ferric chloride	+++	+++	+++	+++	+++
Terpenoids (n=3)	Liebermann-Burchard test	+++	+++	+++	+++	+++
Cardiac glucosides (n=3)	Keller Killian	+++	+++	+++	+++	+++

Legend: (+) present; (-) absent; (NA) not applicable, (n=3) experiment done in triplicate (+++)

3.2 Gas Chromatographic and Mass Spectrometry (GC-MS)

3.2.1 GC-MS analysis of COA® herbal medicine extract in DCM, Hex, EtOH, and EtOAc

Phase two consisted of the identification and characterization of chemical compounds present in COA® herbal medicine (standard) extract and COA® extract using GC-MS analysis in DCM, Hex, EtOH, and EtOAc.

Table 2 presents chemical compounds in extracts of COA® (standard) and in the four different solvents. The names of the compounds in the standard and the four solvents are presented with chemical compound presented by names, chemical formula and retention time. Among the four types of solvents used in this study and the standard COA®, COA® hexane extracts had 60 chemical compounds, the highest, DCM ~~at~~ had 47, ethyl acetate had 37 ethanol ~~at~~ had 18 with COA® standard having the least number at 10 chemical compounds. Previously, Hashmi *et al*. (2013) also carried out the GC-MS analysis of different organic crude extracts from *Thymus vulgaris* L. and reported that hexane, as a solvent of extraction presented major chemical

compounds. Hexane which is a non-polar solvent, extracted more chemical compounds COA® extract more than the other solvents of extraction..[36]

The major active chemical constituents present are 2-methyltetracosane, Phytol acetate, 2,2'-Methylenebis(6-tert-butyl-p-cresol), 11-decyl-Tetracosane, gamma-Sitosterol, Stigmasterol, Testosterone Valerate, 2(3H)-Furanone, Furyl hydroxymethyl ketone, 2H-Pyran-3-ol, Epiglobulol and Cholesta-4,6-dien-3-ol.[37]

The various active constituents present in these extracts are responsible for different biological and pharmacological activities. For example 2-methyltetracosanehas been reported to have free radical scavenging activity[38] while phytol acetate a derivative of phytol, has antimicrobial, antiinflammatory, anticancer and antidiuretic properties.[39] Cantrell *et al*. (2001) reported antimycobacterial activity of phytol acetate against *Mycobacterium tuberculosis*, in vitro.[40] Phenol, 2,2'-methylenebis(6-tert-butyl-p-cresol) have been reported to have antimicrobial and anti oxidant effects.[41] gamma –Sitosterol is a phytosterol present in plants, used in traditional medicines and have strong antifungal, antibacterial and anti-angiogenic activity.[42] Previous studies have reported that stigmasterol has a chemopreventive effect on 7,12-dimethylbenz[a]anthracene (DMBA)induced skin cancer investigated in Swiss albino mice.[43] 2(3H)-Furanone has been reported to have antimicrobial activities against *S. aureus, Bacillus subtilis E. coli* and *Pseudomonas aeuroginosa*.[44] Furyl hydroxymethyl ketone exhibits moderate inhibition against the two Gram-positive bacterial strains (*S. aureus* KCTC 209 and *S. aureus* KCTC 503).[45] 2(*H)*-Pyran-3-ol has been reported to have antibacterial and antifungal properties [46] while Lourens *et al*.(2004), reported antioxidant activity against diphenyl-picrylhydrazine (DPPH assay), antimicrobial and anti-inflammatory activity on *Helichrysum dasyanthum, Helichrysum, felinum, Helichrysum excisum* and *Helichrysum petiolare* of Epiglobulol.[47] Cholesta-4,6-dien-3-ol is a steroid with antimicrobial and free radical scavenging activities.[48]

The presence of various bioactive compounds present in different solvents system justifies the medicinal applications of COA® in the treatment of various disorders or disease conditions.

Table 2: Phytochemicals in dichloromethane (DCM), hexane, ethanol, and ethyl acetate extract of COA® herbal medicine and standard COA® herbal medicine

	COA® Herbal Medicine dichloromethane **extract**		**COA® Herbal Medicine-Hexane extract**		**COA® Herbal Medicine-Ethanol extract**		**COA® Herbal Medicine Ethyl acetate extract**		**COA®Herbal Medicine (standard) extract**	
	Chemical Constituents	**R_t min**	**Chemical Constituents**	**R_t Min**	**Chemical Constituents**	**R_t min**	**Chemical Constituents**	**R_t min**	**Chemical Constituents**	**R_t min**
1	2-Penten-1-ol	3.684	N,N-Dimethyl-formamide	2.663	Triethylamine	2.53	Toluene	3.314	Carbonic acid, methyl pentyl ester	2.99
2	Butanoic acid	4.652	4-Hydroxy- butanoic acid	3.625	N,N-dimethyl-Formamide	2.906	Spiro[2,4]hepta-4,6-diene	10.566	Dimethylsulfoxonium formylmethylide	3.015
3	4-Hexen-1-ol	4.797	3-hydroxy-Benzonitrile	4.047	chloro-Benzene	3.099	Toluene	10.597	Acetic acid,	3.348
4	(E)-2-Hexen-1-ol	4.883	Aniline	4.209	4-hydroxy-Butanoic acid	3.752	1,3-Octanediol	10.706	Acetic acid	3.419
5	Formic acid hexyl ester	4.929	(E)-3-Hexenoic acid	4.268	isocyanato-Benzene	4.088	Furfural	10.819	3-Hexen-1-ol	3.475
6	Pentanoic acid	5.108	2H-Pyran-2, 6 (3H)-dione	4.324	Aniline	4.284	Chlorobenzene	11.304	Formic acid, hexyl ester	3.615
7	3-Pentanol	5.176	(E)-2-Hexenoic acid	4.479	Phytol, acetate	10.59	Hexyl chloroformate	11.958	Hexanoic acid	4.579
8	o-Xylene	5.249	Dehydromevalonic lactone	4.623	Phytol; 3,7,11,15-Tetramethyl-2-hexadecen-1-ol	10.728	5-Methyl-2-furaldehyde	12.101	Heptanoic acid	4.645
9	Ethanol, 2-butoxy-	5.367	1-Octadecyne	5.718	Pentadecanoic acid	11.297	2-Hexenoic acid	12.266	4-Octene	4.705
10	Butanoic acid	5.471	Dodecane	6.106	Pyrene	12.108	α.-Calacorene	12.357	2-Hexenoic acid	4.818
11	Cyclopentane	5.561	5-Methyl- tridecane	7.324	7-Tetradecenal	12.272	4,6-Heptadien-2-one	12.603	Methyl salicylate	6.232
12	Heptan-2-yl acetate	5.645	Tetradecane	7.685	Octadecanoic acid	12.375	Di-epi-.alpha.-cedrene-(I)	12.99	NA	NA

13	4-Methylhept-1-ene	5.956	4-Methyl- tetradecane	8.408	Hexadecanamide	12.5	1-Naphthalenol	13.339	NA	NA
14	2*H*-Pyran-2,6(3H)-dione	6.599	Butylated Hydroxytoluene	8.48	9-Octadecenamide	13.4	2-Naphthalenemethanol	13.951	NA	NA
15	Hexanoic acid	6.787	5-methyl- tetradecane	8.765	Octadecanamide	13.506	Naphthalene	14.039	NA	NA
16	3-Hexenoic acid	6.946	Hexadecane	9.098	Diisooctyl phthalate	14.15	Benzene	14.392	NA	NA
17	Methoxyethylamine Acrylo Nitryl	7.037	8-Hexyl- pentadecane	9.646	Cholesta-4,6-dien-3-ol	16.543	9-Undecenal	14.818	NA	NA
18	Benzyl alcohol	7.194	3-Methyl-heptadecane	9.956	Stigmast-5-en-3-ol, oleate	16.642	Neoclovene oxide	14.862	NA	NA
19	2(3H)-Furanone	7.427	5-Methyl-tetradecane	10.062	NA	NA	1-Cyclohexanone	15.185	NA	NA
20	2-Hexenoic acid	7.68	2-Methylhexadec-1-ene	10.157	NA	NA	4-(2-Acetyl-5,5-dimethylcyclopent-2-enyliden	15.215	NA	NA
21	Furyl hydroxymethyl ketone	7.824	Heptadecane	10.366	NA	NA	Pentafluoropropionic acid	15.304	NA	NA
22	α-Methyl-α-[4-methyl-3-pentenyl]oxiranemethanol	7.914	2-methyltetracosane	10.448	NA	NA	2-Naphthalenemethanol	15.621	NA	NA
23	1-Cyclohexyl-2-nitropropane-1,3-diol	7.978	Phytyl acetate	10.59	NA	NA	7-Oxocholesteryl isocaproate	15.72	NA	NA
24	Maltol	8.314	5-Methyl-tetradecane	10.949	NA	NA	(1S,2E,4S,5R,7E,11E)-Cembra-2,7,11-trien-4,5-diol	16.012	NA	NA
25	4-Butylcyclohexene	8.713	l-(+)-Ascorbic acid 2,6-dihexadecanoate	11.287	NA	NA	3,3,5,5-Tetramethyl-4,5-dihydro-3H-benzo[c] azepine,2-oxide	16.205	NA	NA
26	2(H)-Pyran-3-ol	9.06	Methyl 2-Bromo-5-methoxybenzoate	11.637	NA	NA	Phthalic acid	16.302	NA	NA

27	Methyl salicylate	9.314	Trifluoroacetoxy hexadecane	11.988	NA	NA	4,6,10,10-Tetramethyl-5-oxatricyclo[4.4.0.0(1,4)]dec-2-en-7-ol	16.45	NA	NA
28	Cyclopentan-1-al	9.655	Pyrene	12.102	NA	NA	3-Heptyn-2-one	16.486	NA	NA
29	1-Methyl-3-butenyl 3-methyl-3-hydroxybutyl	9.75	(Z)-9-Octadecenal	12.281	NA	NA	Pentadecanoic acid	16.546	NA	NA
30	2-Carene	9.79	Octadecanoic acid	12.366	NA	NA	Metolachlor	17.183	NA	NA
31	7-Octen-3-ol	10.26	Eicosane	12.564	NA	NA	3-Buten-2-one	17.459	NA	NA
32	Pentadecane	10.531	2-Octyl-1-Dodecanol	12.599	NA	NA	n-Nonadecanol-1	17.528	NA	NA
33	p-Ethoxybenzyl alcohol	10.76	Hexadecanoic acid	12.782	NA	NA	Tricyclo[3.3.1.1(3,7)]decane	17.594	NA	NA
34	2,2'-Bioxepane	10.89	8-Hexyl-pentadecane	12.874	NA	NA	Benzene	17.83	NA	NA
35	2,5-Dimethyltetrahydrofuran	11.089	2-methyltetracosane	12.957	NA	NA	cis-9-Hexadecenal	18.216	NA	NA
36	1,5-Heptadien-4-ol	11.2	11-Decyl-tetracosane	13.056	NA	NA	Octadecanoic acid	18.402	NA	NA
37	Tetradecane	11.676	2,4-dimethyl-Docosane,	13.292	NA	NA	1,4-Ethenoanthracene	18.477	NA	NA
38	2(4H)-Benzofuranone	13.257	Phenol	13.632	NA	NA	NA	NA	NA	NA
39	Z-5-Nonadecene	13.716	Trifluoroacetoxy hexadecane	11.988	NA	NA	NA	NA	NA	NA
40	Epiglobulol	14.2	2-Methylhexacosane	13.983	NA	NA	NA	NA	NA	NA
41	1-Naphthalenol	14.548	2-Methyloctacosane	14.119	NA	NA	NA	NA	NA	NA
42	Oleyl alcohol, trifluoroacetate	15.852	7-Hexyl-docosane	14.199	NA	NA	NA	NA	NA	NA
43	1,1,6-trimethyl-3-methylene-2-(3,6,10,13,14-	16.023	Hexatriacontane	14.412	NA	NA	NA	NA	NA	NA

	pentamethyl-3-ethenyl-pentadec-4-enye)cyclohexane									
44	2-Methyl-5-(2,6,6-trimethyl-cyclohex-1-enyl)-pentane-2,3-diol	16.461	Tetratetracontane	14.834	NA	NA	NA	NA	NA	NA
45	Heneicosane	17.45	3,3,13,13-Tetraethylpentadecane	14.965	NA	NA	NA	NA	NA	NA
46	n-Tetracosanol-1	19.342	11-Decyl-tetracosane	15.042	NA	NA	NA	NA	NA	NA
47	n-Nonadecanol-1	21.846	1-iodo-Hexadecane	15.757	NA	NA	NA	NA	NA	NA
48	NA	NA	Octadecyl 2-Propyl ester	16.373	NA	NA	NA	NA	NA	NA
49	NA	NA	(3 beta)-Cholesta-4,6-dien-3-ol	16.54	NA	NA	NA	NA	NA	NA
50	NA	NA	5-Methyl-Octadecane	16.692	NA	NA	NA	NA	NA	NA
51	NA	NA	1-Bromo-2-methyl-Decane	17.353	NA	NA	NA	NA	NA	NA
52	NA	NA	Stigmasterol	17.481	NA	NA	NA	NA	NA	NA
53	NA	NA	6,6-Diethylhoctadecane	17.657	NA	NA	NA	NA	NA	NA
54	NA	NA	gamma-Sitosterol	17.859	NA	NA	NA	NA	NA	NA
55	NA	NA	Silane	18.104	NA	NA	NA	NA	NA	NA
56	NA	NA	1-bromo-Triacontane	18.333	NA	NA	NA	NA	NA	NA
57	NA	NA	Testosterone Valerate	18.473	NA	NA	NA	NA	NA	NA
58	NA	NA	Acetic acid	19.717	NA	NA	NA	NA	NA	NA

59	NA	NA	Nonahexacontanoic acid	22.207	NA	NA	NA	NA	NA	NA
60	NA	NA	octadecyl ester hexadecanoic acid	23.233	NA	NA	NA	NA	NA	NA

R_t =Retention time (min)

3.2.2 GC-MS analysis of COA® herbal medicine extract and *V. amygdalina* and *P. americana* (Ghana and South Africa) in DCM

The GC-MS analysis was carried out to identify the components present in the solvent extracts of COA® herbal medicine and the plant species *V. amygdalina* and *P. americana.*

Table 3 presents comparative similarity studies of phytochemical compounds in COA® extract and *V. amygdalina* and *P. americana* (Ghana and South Africa) in DCM. Results showed that out of 50 compounds detected in the GC-MS only Heneicosane is present in both COA® and leaf extract of *P. americana* of Ghana although retention time is different which may suggest that the two are different compounds but the compound is of interest to find similarity. The variation in chemical constituent may be because of diversity in the geographical location.

Falco *et al.* (2013) reported the presence of heneicosane as an essential oil in *Persea americana* (Avocado) Ghana. It indicated antimicrobial activities against numerous gram-positive pathogens and specifically *Bacillus cereus* and *B. subtilis*. The topographical area with the distinctive atmosphere condition showed a critical effect on the compound constituents of the plants.[49] Şanli *et al*., 2017 detailed that few sections of the plant have essential oils in various amount, that can be affected by environmental conditions, such as altitude. Essential oil contents of avocado fruits are increased due to increasing altitude level.[50] Hence, the geographical area of the plant must be considered in relation to the intended use.

Table 3: Comparative similarity studies of phytochemical compounds in COA® herbal medicine extract and *V. amygdalina* and *P. americana* (Ghana and South Africa) in DCM

S. No.	COA® **Herbal Medicine DCM**			***Vernomia amygdalina* (GH) EXTRACT**		***Vernomia amygdalina* (SA) EXTRACT**		***Persea americana* (GH) EXTRACT**		***Persea americana* (SA) EXTRACT**	
	Chemical Constituents	**Chemical Formula**	R_t **min**	R_t**min**	**Status**	R_t **min**	**Status**	R_t **min**	**Status**	R_t **min**	**Status**
1	Heneicosane	$C_{21}H_{44}$	17.45	-	-	-	-	**16.21**	+	-	-

Legend: (+) present; (-) absent.

3.2.3. GC-MS Analysis of COA® Extract and Leaf Extracts of *V. amygdalina* and *P. americana* in Hexane from Ghana and South Africa

Table 4 presents the comparative similarity study of phytochemical compounds in COA® extract and leaf extracts of *V. amygdalina* and *P. americana* in hexane from Ghana and South Africa. Phytyl acetate is present in both *P. americana* (GH) and *V. amygdalina* (GH) of Ghana and *V. amygdalina* (SA) of South Africa and COA® extract with R_t values ranging between 10.56-10.96 but not in *P. americana* (SA). Pyrene is present only in *P. americana* (SA) hexane extract with an R_t value of 12.11. Octadecanoic acid is the only chemical compound present in all the leaf extracts collected from two different locations and COA® extract. It has almost the same R_t value i.e. 12.35-12.26. It is a fatty acid ester having antioxidant activity[51] and an antihypertensive agent. In *P. americana* (SA) leaf extracts eicosane was found with R_t value 13.05. Eicosane has wound healing, antimicrobial and anti-inflammatory properties.[52] A compound, 2-methyltetracosane is present in leaf extracts of *V. amygdalina* (SA) with 16.03 and a free radical scavenging agent.[38] From the results the major chemical compound similar to the COA® extract and leaf extracts from both countries are octadecanoic acid and phytol acetate. There is a lack of phytol acetate in *V. amygdalina* (SA) leaf extract. This variation in chemical constituent suggests the diversity in biotic and abiotic factors in both countries such as geographical location, environmental conditions, and seasonal variation.

Table 4: Comparative similarity studies of phytochemical compounds in COA® herbal medicine extract and *V. amygdalina* and *P. americana* (Ghana and South Africa) in Hexane.

S. No.	**COA Herbal Medicine**			***Vernomia amygdalina* (GH) EXTRACT**		***Vernomia amygdalina* (SA) EXTRACT**		***Persea americana* (GH) EXTRACT**		***Persea americana* (SA) EXTRACT**	
	Chemical Constituents	**Chemical Formula**	R_t **min**	R_t **min**	**Status**	R_t **min**	**Status**	R_t **min**	**Status**	R_t **min**	**Status**
1	Phytyl acetate	$C_{22}H_{42}O_2$	10.59	**10.56**	+	**10.96**	+	**10.56**	+	-	-
2	2-methyltetracosane	$C_{25}H_{52}$	12.95	-	-	**16.03**	+	-	-	-	-
3	Pyrene	$C_{16}H_{10}$	12.10	-	-	-	-	-	-	**12.11**	+
4	Octadecanoic acid	$C_{17}H_{35}CO_2H$	12.36	**12.36**	+	**12.38**	+	**12.35**	+	**12.35**	+
5	Eicosane	$C_{20}H_{42}$	12.56	-	-	-	-	-	-	**13.05**	+

Legend: (+) present; (-) absent.

3.2.4. GC-MS Analysis of COA® Extract and Leaf Extracts of *V. amygdalina* and *P. americana* in Ethanol from Ghana and South Africa

Table 5 presents comparative similarity studies of phytochemical compounds in COA® extract and *V. amygdalina* and *P. americana* (Ghana and South Africa) in ethanol. Chloro-benzene with R_t value of 3.11 is only present in leaf extract of *P. americana* (SA). Phytol acetate and pentadecanoic acid are present in COA® and leaf extracts of the two plants with R_t value of 10.57-10.58 and 11.28-11.38. However, in leaf extracts of *V. amygdalina* (SA) and *P. americana* (SA), Hexadecanamide and octadecanamide are present with R_t value of 12.47-12.50 and 13.49.

Hexadecanamide has been reported to have cytotoxic effect against a human colon adenocarcinoma cell line (HCT-116).[53] On the other hand octadecanamide can be used a biomarker for hyperlipidemic conditions.[54]

Table 5: Comparative similarity studies of phytochemical compounds in COA®herbal medicine extract and *V. amygdalina* and *P. americana* (Ghana and South Africa) in ethanol.

S. No.	**COA Herbal Medicine**			***Vernomia amygdalina* (GH) EXTRACT**		***Vernomia amygdalina* (SA) EXTRACT**		***Persea americana* (GH) EXTRACT**		***Persea americana* (SA) EXTRACT**	
	Chemical Compounds	**Chemical Formula**	R_t **min**	R_t **min**	**Status**	R_t **min**	**Status**	R_t **min**	**Status**	R_t **min**	**Status**
1	Chloro-Benzene	C_6H_5Cl	**3.09**	-	-	-	-	-	-	**3.11**	+
2	Phytol acetate	$C_{22}H_{42}O_2$	**10.59**	**10.57**	+	**10.58**	+	**10.57**	+	**10.58**	+
3	Pentadecanoic acid	$C_{15}H_{30}O_2$	**11.29**	**11.32**	+	**11.28**	+	**11.30**	+	**11.28**	+
4	Hexadecanamide	$C_{16}H_{33}NO$	**12.5**	-	-	**12.47**	+	-	-	**12.47**	+
5	Octadecanamide	$C_{18}H_{37}NO$	**13.50**	-	-	**13.49**	+	-	-	**13.49**	+

Legend: (+) present; (-) absent.

3.2.5 GC-MS Analysis of COA® Extract and Leaf Extracts of *V. amygdalina* and *P. americana* in Ethyl Acetate from Ghana and South Africa

Table 6 present comparative similarity studies of phytochemical compounds in COA® extract and *V. amygdalina* and *P. americana* (Ghana and South Africa) in ethyl acetate. Octadecanoic acid is present in all the leaf extracts collected from two different locations and COA® extract with an R_t value of 12.35 to 12.37. Toluene is present in *V. amygdalina* (SA) with R_t value of 2.60. Toluene is a pollutant in the environment due to its use in a wide variety of household and commercial products (largely from motor vehicle exhausts). [55] It has antipunishment property and acute and chronic effects on the central nervous system and may cause congenital anomalies in humans.[56] In an investigation on phytochemical and pharmacological properties of n-hexane leaf extracts of *Nymphoides indica*, octadecanoic acid (stearic acid) including hexadecanoic acid was isolated with mild antimicrobial activity.[57]

Table 6: Comparative similarity studies of phytochemical compounds in COA® herbal medicine extract and *V. amygdalina* and *P. americana* (Ghana and South Africa) in ethyl acetate.

	COA® extract			***Vernomia amygdalina* (GH) EXTRACT**		***Vernomia amygdalina* (SA) EXTRACT**		***Persea americana* (GH) EXTRACT**		***Persea americana* (SA) EXTRACT**	
	Chemical constituent	**Chemical formula**	**Retention time**	**Retention time**	**Status**	**Retention time**	**Status**	**Retention time**	**Status**	**Retention time**	**Status**
1	Toluene	C_7H_8	2.55	-	-	**2.60**	+	-	-	-	-
2	Octadecanoic acid	$C_{18}H_{36}O_2$	12.36	**12.35**	+	**12.37**	+	**12.35**	+	**12.35**	+

Legend: (+) present; (-) absent.

3.2.6 GC-MS Analysis of Leaf Extracts of *V. amygdalina* in DCM, Hexane, Ethanol and Ethyl Acetate from Ghana and South Africa

Table 7 presents similar and different chemical compounds in DCM, hexane, ethanol and ethyl acetate leaf extracts of *V. amygdalina* (Ghana and South Africa). The outcomes from the four solvents used in this study yielded 180 chemical compounds: 94 chemical compounds (52.22%) were available in VA (Ghana) only, 45 chemical compounds (25%) were available in VA (South Africa) only and 41 chemical compounds (22.78%) occurred in both VA (Ghana and South Africa). These findings may suggest that geographical location and climatic conditions may influence the chemical composition of medicinal plants. One author observed that there was a substantial deviation in the amounts of phenolic compounds and protein-binding capacity of leaf extracts of *Achyranthes aspera* collected from Ciaat, Eritrea and Ukulinga, South Africa. *A. aspera* leaf extracts have antibacterial, antifungal, anthelmintic properties as compared to similar extracts from Ukulinga in South Africa.[58] Another investigation revealed that the choice of solvents of extraction has a profound effect on the phytochemical composition of *Lantana camara* leaf extracts.[59]

Table 7: Comparative study of similar and different chemical compounds in DCM, hexane, ethanol and ethyl acetate leaf extracts of *V. amygdalina (*Ghana and South Africa).)

	COMPOUND NAME	VA(GH)	VA(SA)	VA(GH)	VA(SA)	VA(GH)	VA(SA)	VA(GH)	VA(SA)
		SOLVENT 1 DCM	SOLVENT 1 DCM	SOLVENT 2 HEXANE	SOLVENT 2 HEXANE	SOLVENT 3 ETHANOL	SOLVENT 3 ETHANOL	SOLVENT 4 ETHYL ACETATE	SOLVENT 4 ETHYL ACETATE
		STATUS	STATUS	STATUS	STATUS	STATUS	STATUS	STATUS	STATUS
1	α.-Amyrin	+	-	+	-	+	-	+	-
2	γ.-Tocopherol	+	+	+	+	+	-	+	-
3	12-Methyl-E,E-2,13-octadecadien-1-ol	+	-	-	-	-	-	-	-
4	13-Docosenamide	+	+	-	+	-	+	-	-
5	(22Z)-cholesta-5,7,22-trien-3-ol	+	-	-	-	+	-	-	-
6	1-Heneicosanol	+	-	+	-	+	-	+	-
7	2-(8-Chloro-3,7-dimethyl-octa-2,6-dienyloxy)tetrahydropyran	+	-	-	-	+	-	+	-
8	2-Nonadecanone	+	+	+	-	-	-	-	-
9	Hexadecanal	+	+	+	-	+	-	-	-
10	Phytol	+	-	+	+	+	+	+	+
11	Phytol acetate	+	+	+	+	+	+	+	+
12	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	+	+	+	+	+	+	+	-
13	Heptadecanal	+	-	+	-	-	-	-	-
14	Pentadecanoic acid	+	+	+	+	+	+	+	+
15	9-Octadecen-1-ol	+	+	+	-	+	-	+	+
16	Octadecanoic acid	+	+	+	+	+	-	+	-
17	9,12,15-Octadecatrienoic acid	+	-	+	-	+	-	-	-
18	9-Octadecanone	+	-	-	-	-	-	+	-

19	4,4,6a,6b,8a,11,11,14b-Octamethyl-1,4,4a,5,6,6a,6b,7,8,8a,9,10,11,12,12a,14,14a,14b-octadecahydro-2H-picen-3-one	+	-	+	-	+	-	-	-
20	Tetratetracontane	+	+	+	-	-	-	+	+
21	2-Pentadecanone	-	+	+	-	-	+	+	+
22	Hexadecanoic acid	+	-	+	-	+	-	+	-
23	Chondrillasterol	+	-	+	-	+	-	+	-
24	Ergosterol	-	-	+	-	-	-	+	-
25	Cyclopropanemethanol	+	-	+	-	-	-	-	-
26	Diisooctyl phthalate	+	-	+	-	-	-	-	-
27	E,E,Z-1,3,12-Nonadecatriene-5,14-diol	+	-	+	-	+	-	-	-
28	Squalene	+	-	+	+	+	-	+	-
29	n-Tetracosanol-1	-	+	-	-	+	-	+	+
30	Stigmasterol	+	-	-	-	+	-	+	+
31	Vitamin E	+	-	+	-	+	-	+	-
32	Tetradecanoic acid	+	-	+	-	+	-	-	-
33	Eicosanoic acid	+	-	+	-	+	-	+	-
34	Oxirane	+	-	+	-	-	-	+	-
35	9-Octadecenamide	-	-	+	-	+	+	+	+
36	n-Nonadecanol-1	-	-	+	-	-	-	+	-
37	E-10,13,13-Trimethyl-11-tetradecen-1-ol acetate	+	-	+	-	-	-	-	-
38	Ergost-5-en-3-ol	+	-	-	-	+	-	-	-
39	1,6,10,14,18,22-Tetracosahexaen-3-ol	-	-	+	-	+	-	-	-
40	γ-Tocopherol	-	+	+	+	+	-	+	-
41	9,12-Octadecadienoic acid (Z,Z)-	-	-	+	-	+	-	-	+
42	Preg-4-en-3-one	+	-	-	-	+	-	+	-

43	dl-.alpha.-Tocopherol	-	-	+	-	-	-	+	-
44	Benzene, chloro-	-	-	-	+	-	-	+	+
45	1-Hexacosene	-	+	-	-		+	+	+
46	cis,cis,cis-7,10,13-Hexadecatrienal	-	-	-	-	+	-	-	+
47	Hexadecanamide	-	-	-	-	-	+	-	+
48	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate	+	+	-	-	-	-	-	+
49	Octadecanamide	-		-	-	-	+	-	+
50	11,13-Dimethyl-12-tetradecen-1-ol acetate	+	+	-	-	-	-	-	-
51	2-methyltetracosane	-	+	-	+	-	+	-	-
52	2-methyloctacosane	+	+	-	-	-	-	-	-
53	Stigmast-5-en-3-ol, oleate	+	+	-	+	-	-	-	-
54	Heneicosane	+		-	-	-	-	-	+
55	γ.-Sitosterol	-	+	-	+	-	-	-	+
56	Oxirane	-	+	-	-	-	-	+	-
57	2-methylhexacosane	-	+	-	-	-	-	+	-
58	8-Methylenecyclooctene-3,4-diol	-	-	-	-	-	-	+	-
59	1-Phenanthrenemethanol	-	-	-	-	-	-	+	-
60	o-Xylene	-	-	-	-	-	-	+	-
61	(.+/-.)-Lavandulol, chlorodifluoroacetate	-	-	-	-	-	-	+	-
62	Undecane	-	+	-	-	-	-	-	-
63	2-Hexadecene	-	+	-	-	-	-	-	-
64	Eicosyl heptafluorobutyrate	-	+	-	-	-	-	-	-
65	Tetracosyl trifluoroacetate	-	+	-	-	-	-	-	-
66	Tetracosyl acetate	-	+	-	-	-	-	-	-
67	1,19-Eicosadiene	-	+	-	-	-	-	-	-
68	1-Octadecanol	-	+	-	-	-	-	-	-
69	Triacontyl trifluoroacetate	-	+	-	-	-	-	-	-

70	Oxirane	-	-	-	+	-	-	-	
71	16-Hentriacontanone	-	-	-	-	-	-	-	+
72	2-Tert-Butyl-5-(dimethoxy-phosphoryl)-3-methyl-4-oxoimidazolidine-1-caboxylic acid	-	-	-	+	-	-	-	-
73	3.alpha.,7.beta.-Dihydroxy-5.beta	-	-	-	-	-	-	-	+
74	Cholesta-4,6-dien-3-ol	-	-	-	+	-	-	-	-
75	10-Methyl-E-11-tridece-1-ol acetate	-	-	-	-	-	-	-	+
76	Geranyl acetate	-	-	-	-	-	-	-	+
77	7-Octen-3-ol	-	-	-	-	-	-	-	+
78	17-Acetyl-14-hydroxy-16-methoxy-10,13-dimethyl-hexadecahydrocyclopenta[A]phenanthren-3-on	-	-	-	+	-	-	-	-
79	Hexatriacontane	-	-	-	+	-	-	-	-
80	Pyrene	-	-	-	+	-	-	-	-
81	9-(3,3-Dimethyloxiran-2-yl)-2,7-dimethylnona-2,6-dien-1-ol	-	-	-	+	-	-	-	-
82	cis-9-Hexadecenals	-	-	-	+	-	-	-	-
83	Sulfurous acid, 2-propyl tridecyl ester	-	-	-	+	-	-	-	-
84	Cyclopentanone	-	-	-	-	-	+	-	-
85	2-Ethylnon-1-en-3-ol	-	+	-	-	-	-	-	-
86	Bis(tridecyl) phthalate	-	-	-	+	-	-	-	-
87	Formamide	-	-	-	-	-	-	+	-
88	Pentanoic acid, 4-hexadecyl ester	-	-	-	-	-	+	-	-
89	γ.-Dodecalactone	-	-	-	-	-	+	-	-
90	Eicosane	-	-	-	+	-	-	-	-
91	1,1':3',1"-Tercyclopentane	-	-	-	-	-	+	-	-

92	Isosteviol	-	-	-	+	-	-	-	-
93	13-Octadecenal	-	+	-	-	-	-	-	-
94	1,2-Epoxynonane	-	-	-	+	-	-	-	-
95	Silane	-	-	-	-	-	-	-	+
96	cis-9-Hexadecenoic acid, trimethylsilyl ester	-	-	-	-	-	-	-	+
97	Pentadecanal-	-	+	-	-	-	-	-	-
98	Ergosta-7,22-dien-3-ol	-	-	-	-	-	+	-	-
99	11-Dodecyn-1-ol acetate	-	-	-	-	-	-	-	+
100	Cyclopropa[5,6]stigmast-22-en-3-ol	-	-	-	+	-	-	-	-
101	2,3,4-Trimethyl-5-hexen-3-ol	-	-	-		-	-	-	+
102	3-methyl-4-[(trimethylsilyl)oxy]	-	-	-	+	-	-	-	-
103	Tetradecanamide	-	+	-		-	-	-	-
104	Octadecane	-	-	-	+	-	-	-	-
105	d-Glucitol, 2,5-anhydro-1-O-octyl-	+	-	-	+	-	-	-	-
106	9-Octadecenoic acid	+	-	-	-	-	-	-	-
107	Tetracosyl pentafluoropropionate	+	-	-	-	-	-	-	-
108	Hexadeca-2,6,10,14-tetraen-1-ol	+	-	-	-	-	-	-	-
109	Acetic acid	+	-	-	-	-	-	-	-
110	Docosyl trifluoroacetate	+	-	-	-	-	-	-	-
111	Methanesulfonic acid	+	-	-	-	-	-	-	-
112	Methyl 12-oxo-9-dodecenoate	+	-	-	-	-	-	-	-
113	1-Octadecanesulphonyl chloride	+	-	-	-	-	-	-	-
114	2,2,4-Trimethyl-3-(3,8,12,16-tetramethyl-heptadeca-3,7,11,15-tetraenyl)-cyclohexanol	+	-	-	-	-	-	-	-
115	2,6,8-Trimethylbicyclo[4.2.0]oct-2-ene-1,8-diol	+	-	-	-	-	-	-	-
116	3,4-Dimethyl-2-pentanone	+	-	-	-	-	-	-	-

117	Cholest-22-ene-21-ol	-	-	+	-	-	-	-	-
118	γ.-Ergostenol	-	-	+	-	-	-	-	-
119	9,19-Cyclolanost-23-ene-3,25-diol	-	-	+	-	-	-	-	-
120	2,6,10,14,18-Pentamethyl-2,6,10,14,18-eicosapentaene	-	-	+	-	-	-	-	-
121	1-Hexadecanol	-	-	+	-	-	-	-	-
122	Hexadecanoic acid, octadecyl ester	-	-	+	-	-	-	-	-
123	Lup-20(29)-en-3-ol, acetate	-	-	+	-	-	-	-	-
124	Hexadecanoic acid	-	-	+	-	-	-	-	-
125	Lanosterol	-	-	+	-	-	-	-	-
126	11,13-Dimethyl-12-tetradecen-1-ol acetate	-	-	+	-	-	-	-	-
127	2,3-Butanediol	-	-	-	-	+	-	-	-
128	Octanoic acid	-	-	-	-	+	-	-	-
129	9-Octadecenal	-	-	-	-	+	-	-	-
130	2-Hexadecene	-	-	-	-	+	-	-	-
131	Bicyclo[6.1.0]nonane	-	-	-	-	+	-	-	-
132	d-Glycero-d-tallo-heptose	-	-	-	-	+	-	-	-
133	D-Allose	-	-	-	-	+	-	-	-
134	Docosanoic acid	-	-	-	-	+	-	-	-
135	(E,E,E)-3,7,11,15-Tetramethylhexadeca-1,3,6,10,14-pentaene	-	-	-	-	+	-	-	-
136	Valeric acid	-	-	-	-	+	-	-	-
137	Phosphoric acid, diethyl octyl ester	-	-	-	-	+	-	-	-
138	Butyrolactone	-	-	-	-	+	-	-	-
139	Hexadecane	-	-	-	-	+	-	-	-
140	β.-d-Ribopyranoside	-	-	-	-	+	-	-	-
141	Hexadecane	-	-	-	-	+	-	-	-

142	2-Hexadecene	-	-	-	-	+	-	-	-
143	1-Hexadecen-3-ol	-	-	-	-	+	-	-	-
144	Methanol	-	-	-	-	+	-	-	-
145	2,3-Butanediol	-	-	-	-	+	-	-	-
146	3-Eicosene	-	-	-	-	+	-	-	-
147	Megastigmatrienone	-	-	-	-	+	-	-	-
148	Pentalene, octahydro-2-[(2-octyl)decyl]-	-	-	-	-	+	-	-	-
149	2-Dodecylcyclobutanone	-	-	-	-	+	-	-	-
150	Cyclohexane	-	-	-	-	+	-	-	-
160	3H-Naphtho[2,3-b]furan-2-one	-	-	-	-	+	-	-	-
161	(E,E,E)-3,7,11,15-Tetramethylhexadeca-1,3,6,10,14-pentaene	-	-	-	-	+	-	-	-
162	Lycoxanthin	-	-	-	-	+	-	-	-
163	15,17,19,21-Hexatriacontatetrayne	-	-	-	-	+	-	-	-
164	beta.-D-Glucopyranose	-	-	-	-	+	-	-	-
165	Stigmasta-4,7,22-trien-3.alpha.-ol	-	-	-	-	+	-	-	-
166	Cholest-7-en-3-ol	-	-	-	-	+	-	-	-
167	5,8,11-Eicosatriynoic acid, methyl ester	-	-	-	-	+	-	-	-
168	10,12,14-Nonacosatriynoic acid	-	-	-	-	+	-	-	-
169	2,3-Hexanedione	-	-	-	-	+	-	-	-
170	Dodecanoic acid	-	-	-	-	+	-	-	-
171	cis-13,16-Docasadienoic acid	-	-	-	-	+	-	-	-
172	(E)-1-(2,3,6-trimethylphenyl)buta-1,3-diene	-	-	-	-	+	-	-	-
173	trans-2-[2'-(2"-Methyl-1"-propenyl)cyclopropyl]propan-2-ol	-	-	-	-	+	-	-	-
174	Pentane	-	-	-	-	+	-	-	-
175	Benzeneacetic acid	-	-	-	-	+	-	-	-

176	Methoxyacetic acid, 2-pentyl ester	-	-	-	-	+	-	-	-
177	Erythritol	-	-	-	-	+	-	-	-
178	5-Isopropyl-6-methyl-hepta-3,5-dien-2-ol	-	-	-	-	+	-	-	-
179	1,6,10,14-Hexadecatetraen-3-ol	-	-	-	-	+	-	-	-
180	3-Hexanol	-	-	-	-	+	-	-	-

Compound occur in Ghana (VA) only Compound occur in both Ghana and South Africa (VA) Compound present in South Africa (VA) only

(n = 180; n represents the total number of chemical compounds identified inclusive of the repeated compound).

3.2.7 GC-MS analysis of leaf extracts of *P. americana* in DCM, hexane, ethanol and ethyl acetate from Ghana and South Africa

Table 8 presents the comparative study of similar and different chemical compounds in DCM, hexane, ethanol and ethyl acetate leaf extracts of *P. americana (*Ghana and South Africa)

Table 8 presents' similar and different chemical compounds in DCM, hexane, ethanol and ethyl acetate leaf extracts of *P. americana* (Ghana and South Africa). The outcomes from the four solvents used in this study yielded 167 compounds: 61 compounds (36.53%) were present in PA (Ghana) only, 52 compounds (31.13%) were only present in PA (South Africa) and 54 compounds (32.34%) occurred in both PA (Ghana and South Africa). These findings may suggest the effect of geographical location and climatic conditions on the presence and amount of phytochemicals. The outcome of this investigation is not in line with results reported by Shah 2015 on the phytochemical analysis of plants that have medicinal properties in mountainous areas around Mandi Bahauddin, Pakistan that there is no difference in plants due to their different environment, rather differences in species.[60]

Table 8: Comparative study of similar and different chemical compounds in ethanol leaf extracts of *P. americana* (Ghana –PA (GH) and South Africa-PA (SA)(n = 167, n represents the total number of chemical compounds identified inclusive of the repeated compound)

S/No.	CCOMPOUND NAME	PA (GH)	PA (SA)	PA (GH)	PA (SA)	PA (GH)	PA (SA)	PA (GH)	PA(SA)
		SOLVENT 1 DCM	SOLVENT 1 DCM	SOLVENT 2 HEXANE	SOLVENT 2 HEXANE	SOLVENT 3 ETHANOL	SOLVENT 3 ETHANOL	SOLVENT 4 ETHYL ACETATE	SOLVENT 4 ETHYL ACETATE
		STATUS	STATUS	STATUS	STATUS	STATUS	STATUS	STATUS	STATUS
1	Bicyclo[3.1.0]hexane	+	-	-	-	-	-	+	-
2	Eicosane	-	-	-	+	-	+	-	-
3	1,2,3-Propanetriol, 1-acetate	-	+	-		-	-	-	+
4	Cubedol	-	+	+	+	-	-	+	-
5	4-epi-cubedol	-	-	-	-	+	-	-	-
6	α.-Cubebene	+	-	+	-	-	-	+	-
7	α .-Copaene	-	+	+	-	-	-	+	-
8	Phytolacetate	+	+	+	*	+	+	+	+
9	Caryophyllene	+	+	+	-	+	-	+	+
10	Caryophyllene oxide	-	+	+	+	-	-	+	-
11	β.-copaene	+	-	+	-	-	-	+	-
12	2-Pentadecanone	-	+	-	+	-	+	-	-
13	1-Hexacosene	-	+	-	-	-	+	-	-
14	Humulene	+	-	+	-	-	-	+	-
15	Pentadecanoic acid	+	+	+	+	+	+	+	+
16	1,6-Cyclodecadiene	+	-	+	-	+	-	+	-
17	Aromandendrene	-	-	+	-	-	-	+	-

18	Phytol	+	+	+	+	+	+	+	+
19	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	+	+	+	+	+	-	+	+
20	9,12-Octadecadienoic acid (Z,Z)-	-	-	-	-	+	+	-	-
21	Heptadecanal	+	-	+	-	-	-	+	-
22	γ.-Elemene	-	-	+	-	-	-	+	-
23	cis,cis,cis-7,10,13-Hexadecatrienal	-	-		-	-	+	+	-
24	9-Octadecen-1-ol	+	+	+	-	+	-	+	+
25	2-Hexadecene	+	-	+	-	+	-	+	-
26	Octadecanoic acid	+	-	+	+	+	+	+	+
27	Hexadecanamide	-	-	-	+	-	+	-	-
28	7-Tetradecenal	+	+	-	-	-	-	-	+
29	1,5-Cyclodecadiene	-	-	+	-	-	-	+	-
30	(-)-Spathulenol	-	-	+	-	-	-	+	-
31	7-Octen-3-ol	-	+	+	+	+	+	+	+
32	Stigmasterol	+	+	-	+	+	+	+	+
33	γ.-Sitosterol	+	+	+	+	+	+	+	+
34	13-Docosenamide	-	+	+	-	+	-	+	-
35	Heneicosane	+	-	-	-	-	+	-	-
36	Stigmastanol	-	-	+	-	+	-	+	-
37	Vitamin E	+	+	+	-	+	-	+	+
38	9,17-Octadecadienal	+	+	+	-		-	+	-
39	10-Methyl-E-11-tridece-1-ol acetate	-	-	-	+	+	+	+	+
40	cis-9-Hexadecenoic acid, trimethylsilyl ester	+	+	+	+	-	+	-	+
41	Di(1-decynyl)mercury	+	+	+	-	+	-	+	+
42	Tetradecanoic acid	-	-	+	-	+	-	+	-

43	11-Dodecyn-1-ol acetate	+	+	+	+	+	+	+	+
44	3,7,11,Trimethyl-8,10-dodecedienylacetate	-	-	-	-	+	-	+	-
45	Oxirane	-	+	-	+	-	-	-	-
46	6,11-Undecadiene	+	-	-	-	+	-	-	-
47	2,3,4-Trimethyl-5-hexen-3-ol	+	-	-	-	-	+	-	-
48	2-Hydroxy-(Z)9-pentadecenyl propanoate	+	+	+	-	+	-	+	+
49	11,14-Eicosadienoic acid, methyl ester	+	+	-	-	-	-	-	-
50	Cyclohexane	-	-	+	-	-	-	+	-
51	9-Octadecenamide	-	-	-	+	-	-	-	-
52	(R)-(-)-14-Methyl-8-hexadecyn-1-ol	+	-	+	-	-	-	-	+
53	1-Heneicosanol	-	+	-	-	+	-	-	-
54	Tetratetracontane	+	+	+	+	-	+	+	+
55	2-Nonadecanone, O-methyloxime	-	+	-	-	+	-	-	+
56	Cyclopropaneoctanoic acid	-	-	+	-	+	-	-	-
57	Cyclopropanecarboxylic acid, pentadecyl ester	+	+	-	-	-	-	-	+
58	E-11-Methyl-12-tetradecen-1-ol acetate	+	+	-	-	-	--	-	+
59	16-Hentriacontanone	+	+	+	+	-	+	+	-
60	Acetic acid, 3-ethylpent-3-yl ester	-	+			-	-	-	+
61	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate	+	+	+	+	+	-	+	+
62	3-Methyl-1-dodecyn-3-ol	-	-	+	-	+	-	+	+
63	Hexadecanal	+	+	+	+	-	-	+	+
64	3-Methyl-2-(2-methylene-cyclohexyl)-butan-2-ol	-	+	-	+	-	-	-	+

65	3-Isopropyl-4-methyl-dec-1-en-4-ol	+	+	+	-	-	-	-	-
66	Estran-3-one	+	-	+	-	-	-	+	-
67	(2-Methyl-[1,3]dioxolan-2-yl)-acetic acid	-	-	-	-	+	-	-	+
68	γ- Tocopherol	+	-	+	+	+	-	+	+
69	9-Methyl-10,12-hexadecadien-1-ol acetate	+	+	+	-	+	-	+	+
70	Z-5,17-Octadecadien-1-ol acetate	-	+	+	-	-	-	-	+
71	Ethanol	+	+	+	-	+	-	+	+
72	1-Heptacosanol	+	+	+	+	-	-	+	+
73	9-Methyl-Z,Z-10,12-hexadecadien-1-ol acetate	+	+	-	-	-	-	-	-
74	Octacosyl acetate	-	+	-	+	-	-	+	+
75	Ergost-5-en-3-ol	+	+	+	+	+	-	+	+
76	9-Eicosyne	-	+	-	-	-	-	+	-
77	Heptacosyl acetate	+	+	-	-	-	-	-	+
78	Docosyl trifluoroacetate	+	-	+	-	-	-	+	-
79	9,19-Cyclolanost-23-ene-3,25-diol	-	-	-	+	+	-	+	-
80	9,19-Cyclolanostan-3-ol	-	-	+	+	-	-	-	+
81	Oleic acid, 3-(octadecyloxy)propyl ester	-	+	-	-	-	-	+	+
82	Tetracosyl acetate	-	-	+	-	-	-	+	-
83	α.-Tocopherol	-	-	+	-	+	-	+	+
84	17-Pentatriacontene	+	-	+	-	+	-	-	-
85	2,3-Butanediol	-	-	-	-	+	-	-	-
86	β.-Pinene	+	-	-	-	-	-	-	-
87	2-Pentanol	-	-	+	-	-	-	-	-
88	2-Propenoic acid, oxiranylmethyl ester	-	-	-	-	+	-	-	-
89	Formamide	-	-	-	-	-	+	-	-

90	Benzene	-	-	-	-	-	-	+	-
91	2(4H)-Benzofuranone	-	-	-	+	-	-	-	-
92	Benzene, chloro-	-	-	-	-	-	+	-	-
93	Pentanoic acid, ethyl ester	-	-	-	-	-	-	+	-
94	4-Penten-2-one	-	+	-	-	-	-	-	-
95	1H-Cycloprop[e]azulen-7-ol	-	-	-	+	-	-	-	-
96	Ethyl orthoformate	-	-	-	-	+	-	-	-
97	1-Heptanol	-	-	-	-	-	-	+	-
98	1,6,10-Dodecatrien-3-ol	-	-	-	-	-	-	-	+
99	1H-Cyclopenta[1,3]cyclopropa[1,2]benzene	-	+	-	-	-	-	-	-
100	1H-Cycloprop[e]azulene	-	-	-	+	-	-	-	-
101	(3-Methyl-oxiran-2-yl)-methanol	-	-	-	-	+	-	-	-
102	Eicosanoic acid	-	-	-	+	-	-	-	-
103	4H-Pyran-4-one	-	-	-	-	+	-	-	-
104	2-Butanone	-	+	-	-	-	-	-	-
105	Isosorbide	-	-	-	-	+	-	-	-
106	Cyclohexane	+	-	-	-	-	-	-	-
107	Bicyclo[7.2.0]undec-4-ene	-	+	-	-	-	-	-	-
108	6-Methyl-6-(5-methylfuran-2-yl)heptan-2-one	-	-	-	-	+	-	-	-
109	6,11-Dimethyl-2,6,10-dodecatrien-1-ol	-	-	-	+	-	-	-	-
110	Tetracyclo[6.3.2.0(2,5).0(1,8)]tridecan-9-ol	-	+	-	-	-	-	-	-
111	2-Piperidinone	-	+	-	-	-	-	-	-
112	n-Tetracosanol-1	-	-	-	-	-	+	-	-

113	2-methylhexacosane	+	-	-	-	-	-	-	-
114	2-methyloctacosane	-	-	+	-	-	-	-	-
115	Squalene	-	-	-	-	-	-	-	+
116	Z,Z-8,10-Hexadecadien-1-ol	-	-	-	+	-	-	-	
117	2-Hexanone	-	-	-	-	+	-	-	-
118	Silane	-	-	-	-	-	+	-	-
119	7-Oxabicyclo[4.1.0]heptane	-	+	-	-	-	-	-	-
120	1,3-Benzenediol	-	-	-	-	+	-	-	-
121	3-Buten-2-one	-	+	-	-	-	-	-	-
122	Z,Z-5,16-Octadecadien-1-ol acetate	-	-	-	-	-	-	-	+
123	8-Hexadecanol	+	-	-	-	-	-	-	-
124	30-Norlupan-28-oic acid	-	-	-	+	-	-	-	-
125	5-Isopropyl-6-methyl-hepta-3,5-dien-2-ol	-	-	-	-	+	_	-	-
126	Octadecanamide	-	-	-	-	-	+	-	-
127	Kauran-18-al,	-	-	-	+	-	-	-	-
128	n-Nonadecanol-1	+	-	-	-	-	-	-	-
129	1-Heptanol	-	-	-	+	-	-	-	+
130	Geranyl acetate	-	-	-	-	-	+	-	-
131	α.-Tocopheryl acetate	-	-	-	+	-	-	-	-
132	2-Ethylnon-1-en-3-ol	-	+	-	-	-	-	-	-
133	Tetradecyl trifluoroacetate	-	-	+	-	-	-	-	-
134	3-alpha.,7.beta.-Dihydroxy-5.beta-cholestanate	-	-	-	-	-	+	-	-
135	cis-4,7,10,13,16,19-Docosahexaenoic acid	-	-	-	+	-	-	-	-
136	Silane	-	+	-	-	-	-	-	-
137	2-Octylcyclopropene-1-heptanol	-	-	-	+	-	-	-	-

138	n-Heptadecanol-1	-	-	-	-	-	-	+	-
139	cis, 6-Octadecenoic acid, trimethylsilyl ester	-	+	-	-	-	-	-	-
140	Decalin-8a-ol-7-one	-	+	-	-	-	-	-	-
141	Cyclopropanecarboxylic acid	-	-	-	-	+	-	-	-
142	5-Hexadecanol	-	+	-	-	-	-	-	-
143	Cholest-4-en-3-one	-	-	+	-	-	-	-	-
144	α.-Tocopherol-.beta.-D-mannoside	-	-	-	-	-	-	+	-
145	Oxirane	-	-	-	+	-	-	-	-
146	Z,Z-4,6-Nonadecadien-1-ol acetate	-	-	-	-	+	-	-	-
147	Tetracosanal	-	-	-	-	-	-	-	+
148	2-Isopropenyl-5-methylhex-4-enal	-	-	-	-	-	-	+	-
149	1,2-15,16-Diepoxyhexadecane	-	-	+	-	-	-	-	-
150	10,11-Epoxy-n-undecan-1-ol	-	-	-	-	-	-	+	-
151	Oxirane	-	-	+	-	-	-	-	-
152	1,3-Dioxolane	-	-	+	-	-	-	-	-
153	Cyclopropanecarboxylic acid	-	-	-	-	-	-	+	-
154	Stigmast-4-en-3-one	-	-	-	+	-	-	-	-
155	2,4-Hexadienedioic acid	-		-	-	+	-	-	-
156	Tetracosyl trifluoroacetate	-	+	-	-	-	-	-	-
157	4,22-Stigmastadiene-3-one	-	-	-	-	-	-	-	+
158	Tetracosane	-	-	+	-	-	-	-	-
159	Stigmastane-3,6-dione	-	-	-	+	-	-	-	-
160	Eicosyl pentafluoropropionate	-	-	-	-	+	-	-	-
161	Tetracosyl pentafluoropropionate	-	-	-	-	-	-	-	+
162	Heptacosyl trifluoroacetate	+	-	-	-	-	-	-	-

163	13,15-Octacosadiyne	-	-	-	-	-	-	-	+
164	6,10,14-Trimethyl-pentadecan-2-ol	-	-	+	-	-	-	-	-
165	Triacontyl acetate	-	-	+	-	-	-	-	-
166	Heneicosyl trifluoroacetate	-	-	+		-	-	-	-
167	Oxirane	-	-	+	-	-	-	-	-

Legend: Present in Ghana (PA) only Present in South Africa (PA) only Present in both Ghana and South Africa (PA)

n = 167, n represents the total number of chemical compounds identified inclusive of repeated compounds

4. Conclusions

This comparative investigation examined similarities of chemical compounds found in standard COA®, solvent extracts of COA® and leaf extracts of *V. amygdalina* and *P. americana* (Ghana and South Africa) in four different solvents (DCM, hexane, ethanol and ethyl acetate), to establish a scientific rationale in the pharmacological properties of medicinal plants collected from the two different locations (Ghana and South Africa). It also investigated the effect of geographical location of each species in terms of similar and different chemical compounds in the leaf extracts of *V. amygdalina* and *P. americana*. A qualitative chemical examination of different ethanolic extracts of *V. amygdalina* and *P. americana* showed the presence of alkaloids, anthraquinones derivatives, saponins, flavonoids, tannins, terpenoids, and cardiac glucosides. Following the GC-MS analysis, the major chemical compounds common to all four solvents used for the extraction are octadecanoic acid, phytol acetate, pentadecanoic acid, and heneicosane.

In terms of geographical location, results indicated substantial differences and moderate similarities in chemical compounds in all four solvents of extraction for *V. amygdalina* species from the two geographical areas. *P. americana* showed mild differences in chemical compounds from the two regions.

It can be concluded that COA® herbal medicine contains leaf extracts of *V. amygdalina* and *P. americana* and that geographical and environmental factors have a profound effect on the phytochemical compounds present in leaf extracts of the two plant species from Ghana and South Africa.

Further studies are warranted to establish whether the presence of the identified bioactive components in the four solvent leaf extracts are responsible for the pharmacological and biological properties of the COA® herbal medicine.

Conflict of Interest

The authors declare no conflict of interest.

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CHAPTER 4 - SYNTHESIS CHAPTER

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Synthesis and discussion - the significance of major findings

Ethnomedicinal use and pharmacological properties for *Persea americana* and *Vernonia amygdalina*

Plant extracts have been used in recent years as biological controls in the treatment of diseases with most ideas obtained from ethnomedicinal uses of leaf extracts of *V. amygdalina* and P. *americana.* These plants are used by local communities in almost all African countries for the management of various diseases.[1,2] Ethnomedicine is an alternative to modern allopathic medicines.[3]

Aqueous leaf of *V. amygdalina* is gargled in the mouth for the treatment of toothache in Cameroon,[4] In an *in vivo* study of aqueous leaf extract of *P. americana* against CCl_4-induced toxicity in rat, the extract showed hepatoprotective activity (Brai *et al* 2014) and *in vitro* study revealed the antidiabetic effect on type 2 diabetes[5] with also antifungal activities.[6]

Hexane leaf extracts of *P. americana* have antimicrobial activity [7]An *in vitro* study in Cameroon, of dichloromethane extract of *V. amygdalina* indicated its antiplasmodial activity[8] with no toxicity.

Results of the phytochemicals screening

Phytochemicals study of *P. americana* and *V. amygdalina* leaf extracts indicated that both plants contain alkaloids, flavonoids, saponins, tannins, phlobatannins, terpenoids, anthraquinone, and glycosides.[9] These classes of phytochemicals are in concordance with conclusions made by Asaolu et al. (2010), of the class of phytochemicals found in the aqueous and methanolic extracts of dried leaves of *Persea americana* and *Vernonia amygdalina*.[10] The same author concluded that the presence of phytochemicals and antioxidants provides evidence of the hypotensive activities of *P. americana* and *V. amygdalina* leaf extracts. These plants have been used by local people for the treatment of diseases such as hypertension, diabetes, inflammation, and cancer. This perhaps could probably support the information about the traditional uses of these plants in the treatment and management of these diseases by the people of the African continent.[11]

GC-MS analysis of COA® herbal medicine extract in DCM, Hex, EtOH, and EtOAc

Leaf extracts of *P. americana* and *V. amygdalina* have beneficial pharmacological and biological activities reported in the literature. Much work needs to be done on the identification and isolation of compounds responsible for their reported pharmacological activities. In this study, the intention of using GC-MS analysis is to identify the phytochemicals present in the COA® herbal medicine (standard), COA® extract and leaf extracts of *P. americana* and *V. amygdalina* (from Ghana and South Africa) in four solvents (DCM, Hex, EtOH, and EtOAc).

Results of the GC-MS data showed four major compounds that are common to all four solvent of extraction. These major chemical compounds are octadecanoic acid, phytol acetate, pentadecanoic acid, and heneicosane.

Phytol acetate is an acetylated derivative of phytol. Phytol has anticonvulsant[12] properties and as such, phytol acetate is expected to have similar pharmacological effects. Al-Marzoqi *et al*. (2016) indicated the presence of phytol and phytol acetate in a GC-MS analysis of seed extracts of *Cassia angustifolia* in a study to evaluate the antimicrobial property of the seed.[13] The same author indicated that phytol acetate has anti-inflammatory, antileishmanial and antitrypanosomal properties.

Another author confirmed the presence of phytol in n-hexane extract and phytol acetate in ethyl acetate and methanol extracts of *Ficus mucoso* leaf extracts.[14]

Octadecanoic acid is found in leaf extracts of *S. nigrum* and has been reported to have antibacterial properties in an *in vitro* assay against *Escherichia coli, Proteus mirabilis, Staphylococcus aureus, Pseudomonas aerogenosa,* and *Klebsiella pneumonia*.[15] The presence and pharmacological properties of octadecanoic acid are affirmed by the GC-MS analysis of an ethanolic extract of *Evolvulus alsinoides* amongst others as a chemopreventive agent, anticancer, anti-microbial activity, antioxidant and antidiabetic properties.[16]

Pentadecanoic acid and heneicosane were also major compounds found in the leaf extract of *P. americana* and *V. amygdalina* and affirmed by Jiang *et al* (2015), who in a GC-MS analysis found pentadecanoic acid, heneicosane and octadecanoic acid in leaf extracts of sugarcane and maize.[17]

The presence of these secondary metabolites may contribute to the different therapeutically benefits of COA® herbal medicine.

Variations in the phytochemicals due to the difference in the geographical locations

Substantial differences were observed in the contents of the active ingredients of different extracts collected from the two locations (Ghana and South Africa). Banerjee and Bonde (2011) concluded in their study that, there were major differences in composition and contents of phenolics extracts of the bark of *Bridelia. retusa* due to the dielectric constant of the solvents and geographical locations

of plant and the effect of climate.[18] Reports show a huge effect of geographical location on antioxidant activity and vitamin C contents of guava fruit.[19]

Most phytochemicals were found in *V. amygdalina* (Ghana) i.e. 94 out of 180 compounds. In the case of *P. Americana,* results are more or less similar although that of Ghana has slightly higher compounds. In the case of *P. americana* (Ghana), there were 61 chemical compounds present, where as in *P. americana* (South Africa) 52 compounds out of 167 total. The presence of variations in the chemical constituents in samples collected from the two different locations signifies that the environmental conditions and geographical locations have an impact in the chemical constituents of the plants.

General conclusions

The overall aim of this study was to evaluate and compare the different phytochemicals present in COA® herbal medicine and crude extracts of *V. amygdalina* and *P. americana* collected from Ghana and South Africa by phytochemical screening methods and GC-MS analysis. The purpose of this study was to know the exact phytochemicals present in COA® herbal medicine. Integrating the findings for all of the active ingredients and the geographical factors of each plants extracts, it is concluded that *V. amygdalina* and *P. americana* collected from Ghana have more phytochemicals than the same species in South Africa due to the differences in environmental conditions in the two countries. There is a lack of phytol acetate in *V. amygdalina* (SA) leaf extract. This variation in chemical constituent suggests the diversity in biotic and abiotic factors in both countries such as geographical location, environmental conditions, and seasonal variation. Phytochemicals present in leaf extracts of *V. amygdalina* and *P. americana* gives a scientific basis to validate the different pharmacological and therapeutics benefits of leaf extract of *V. amygdalina* and *P. americana*. Based on results, it is concluded that COA® herbal medicine contains *P. americana* and *V. amygdalina* leaf extracts.

Recommendations for further research

Based on the present study, recommendations are made for these ethnobotanically useful plant species. These are as follows:

- Although the phytochemical screening of both the plants from different locations has been done, there is still a need to validate the reason for the difference in the phytoconstituents.
- Further studies on the isolation of active constituents are required.

- There is a need to advance research for the herbal formulation development and evaluation of these plants.
- Further *in-vitro* and *in-vivo* work is needed to evaluate the pharmacological effects of these plants.
- Further in-*vitro* and *in-vivo* studies using different solvents of extraction and their effects in biological systems are needed.

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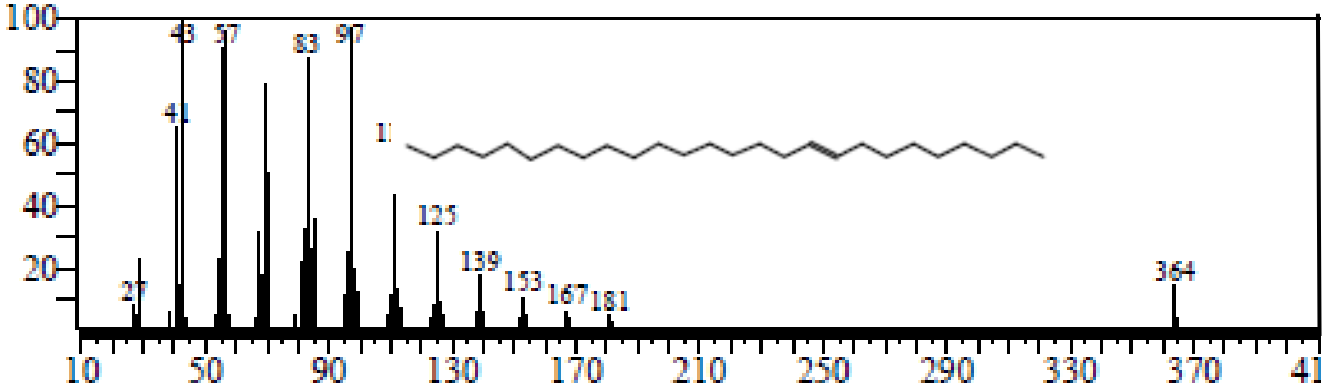

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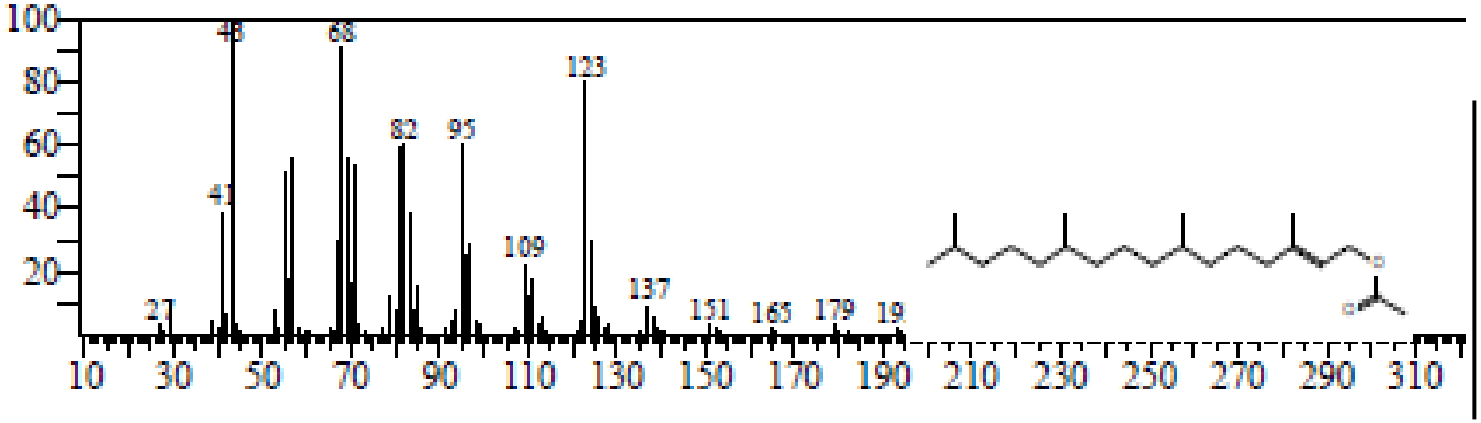

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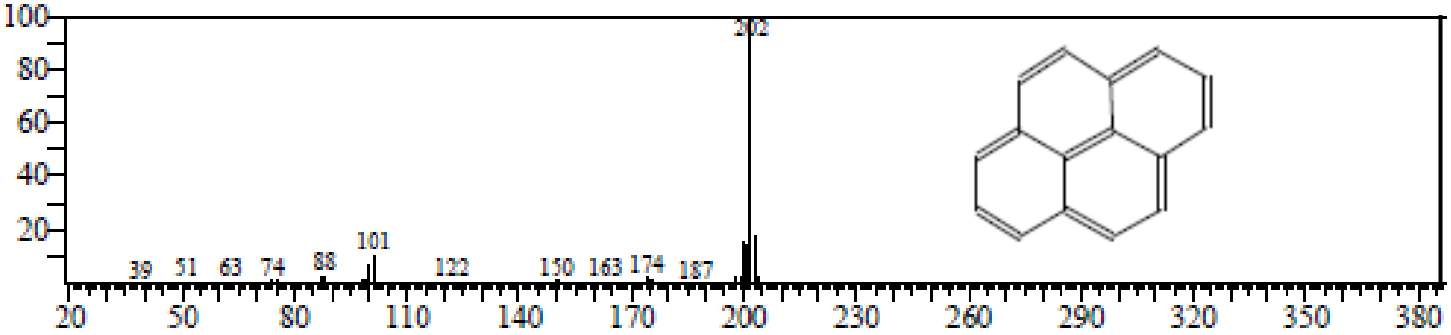

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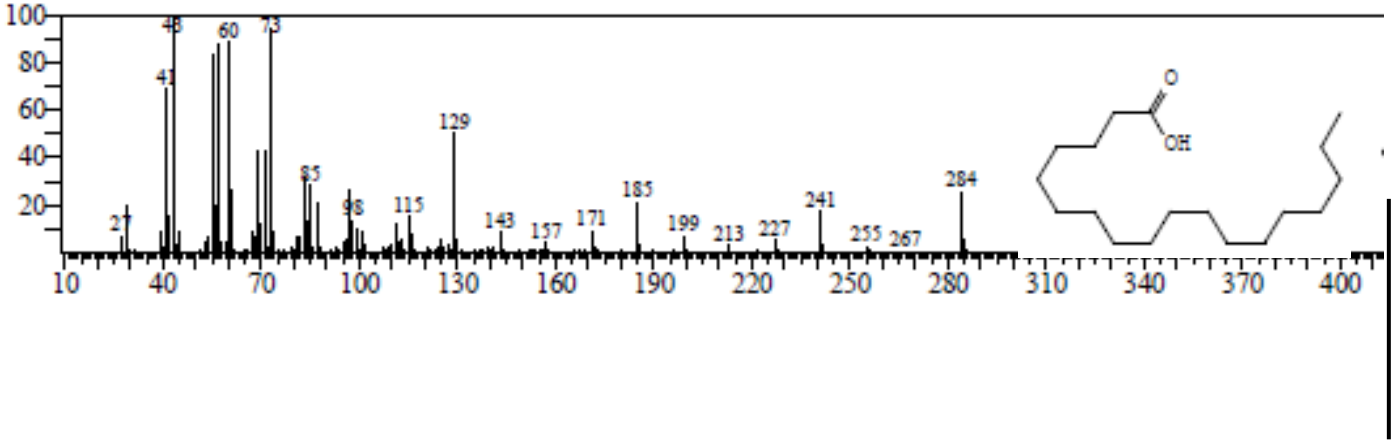

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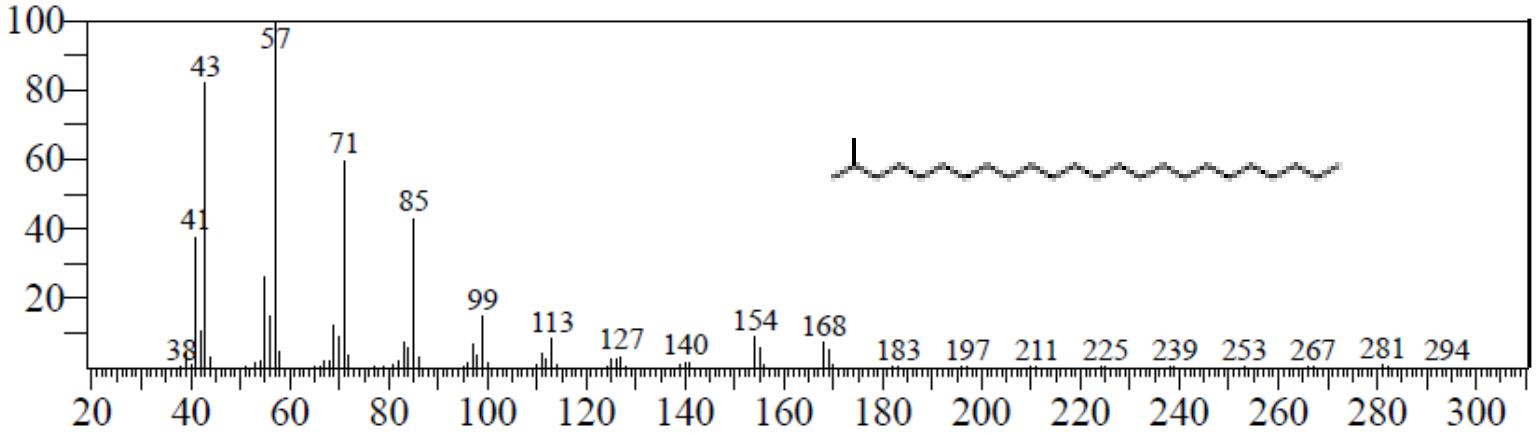

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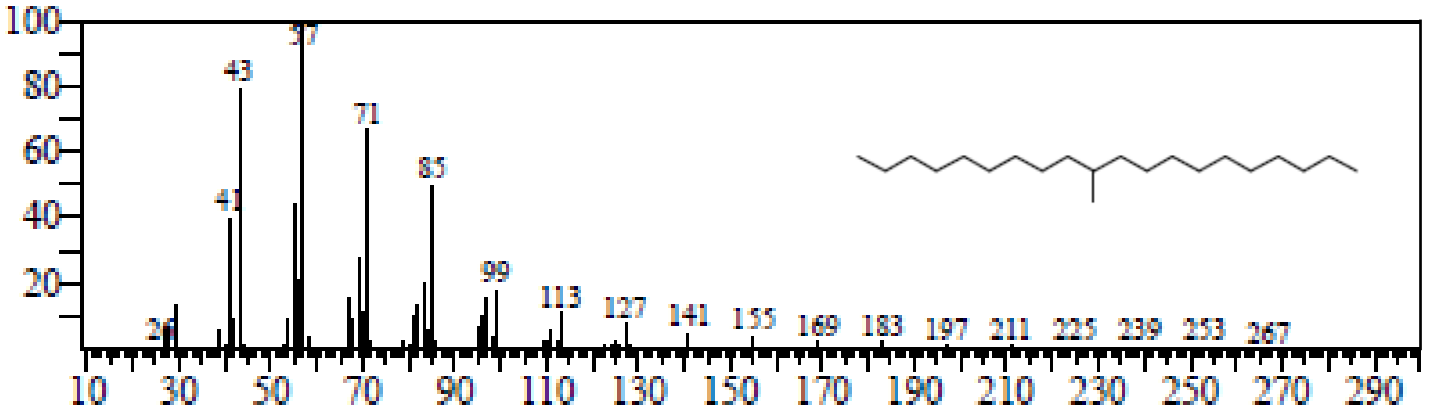

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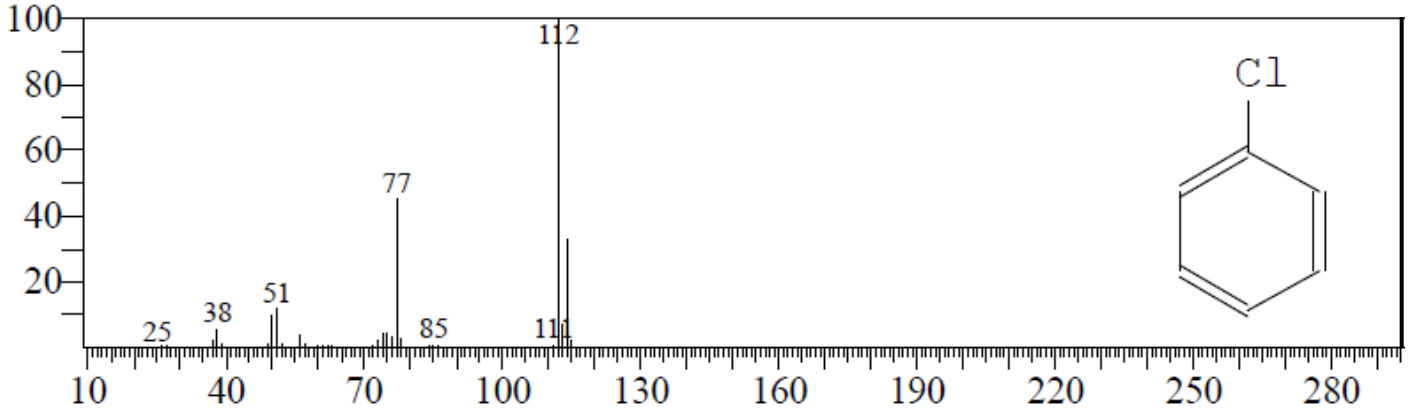

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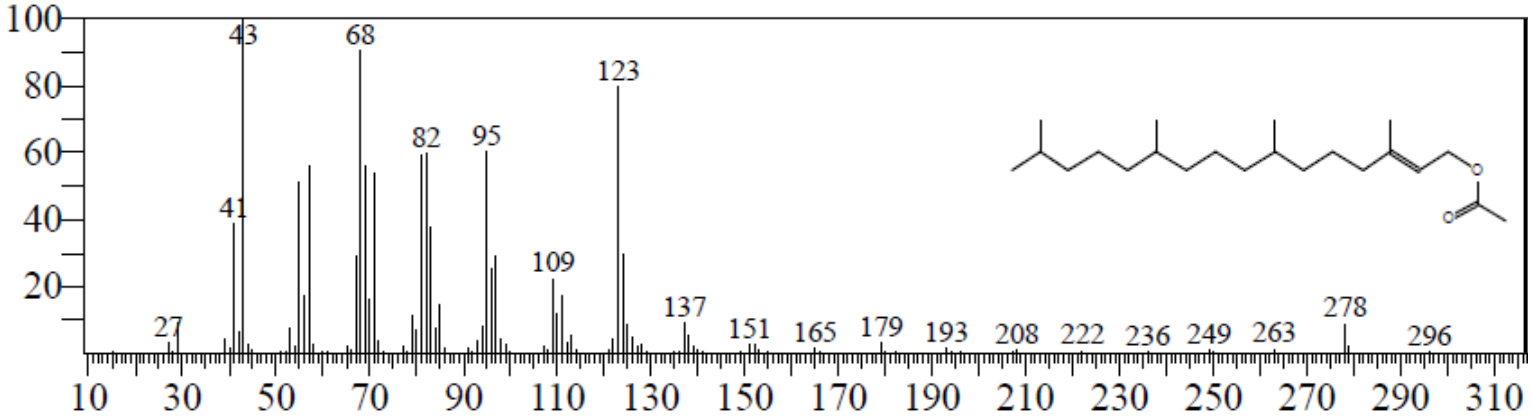

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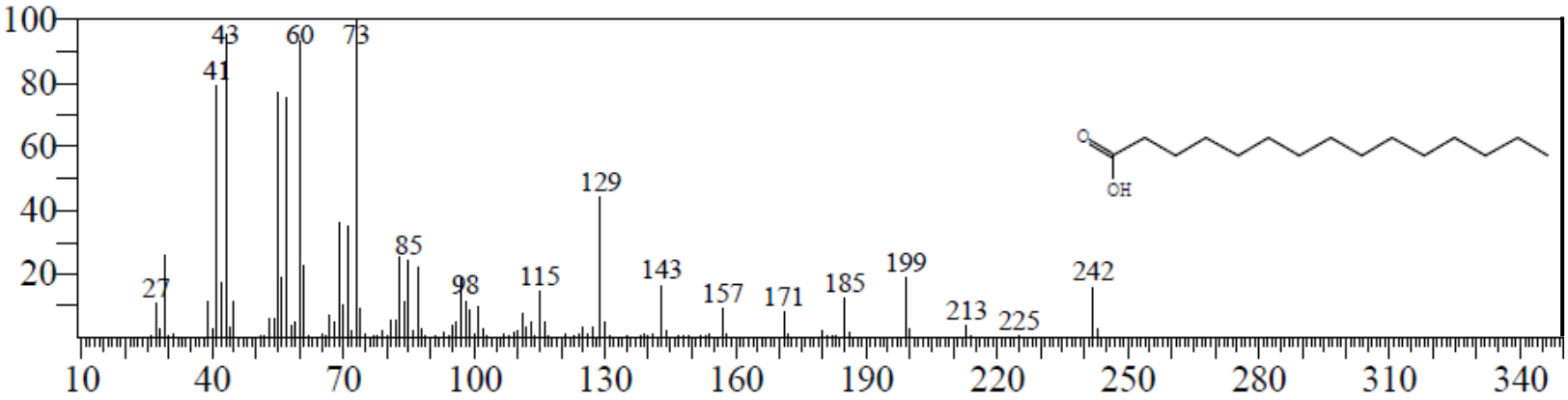

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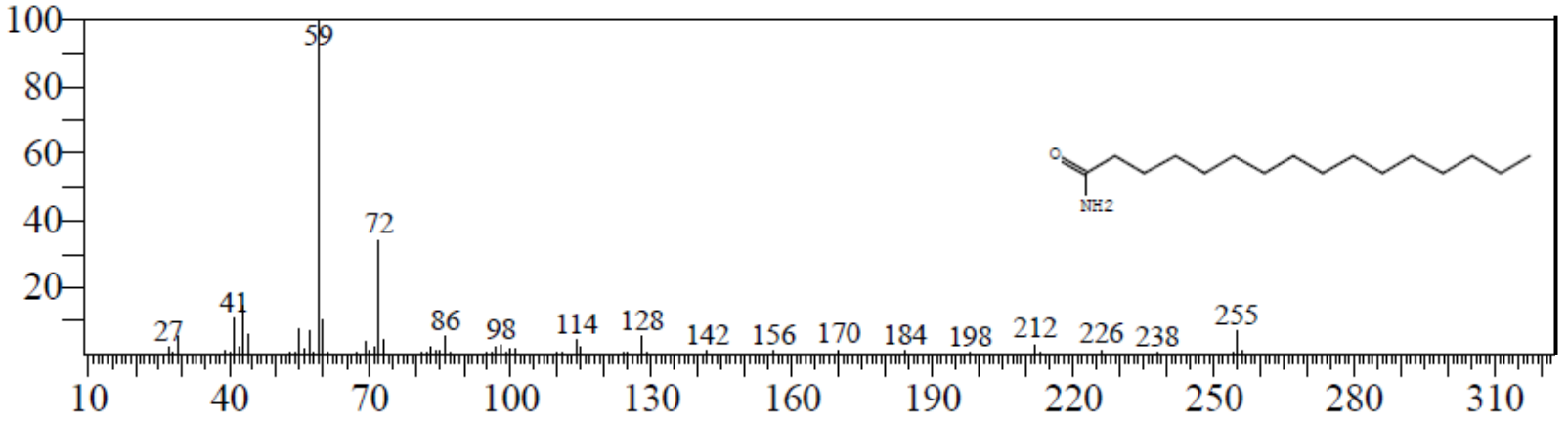

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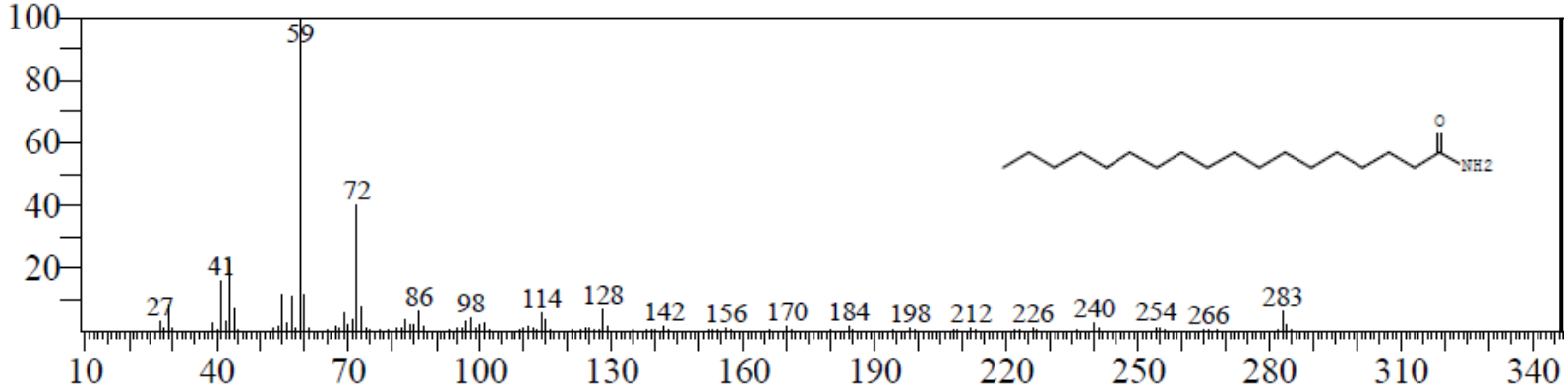

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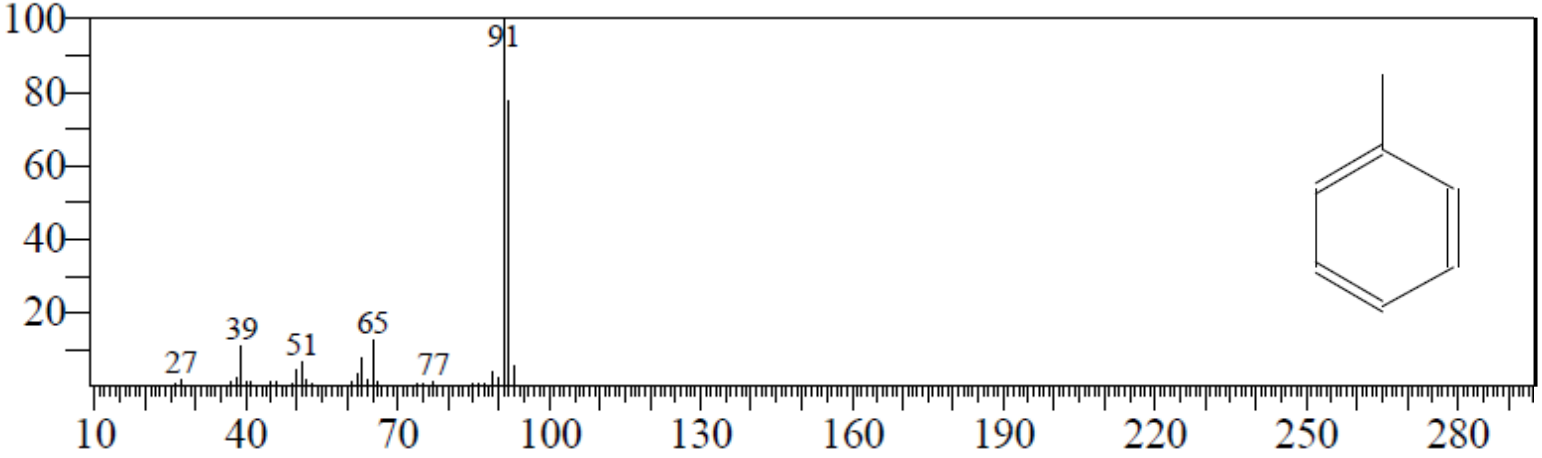

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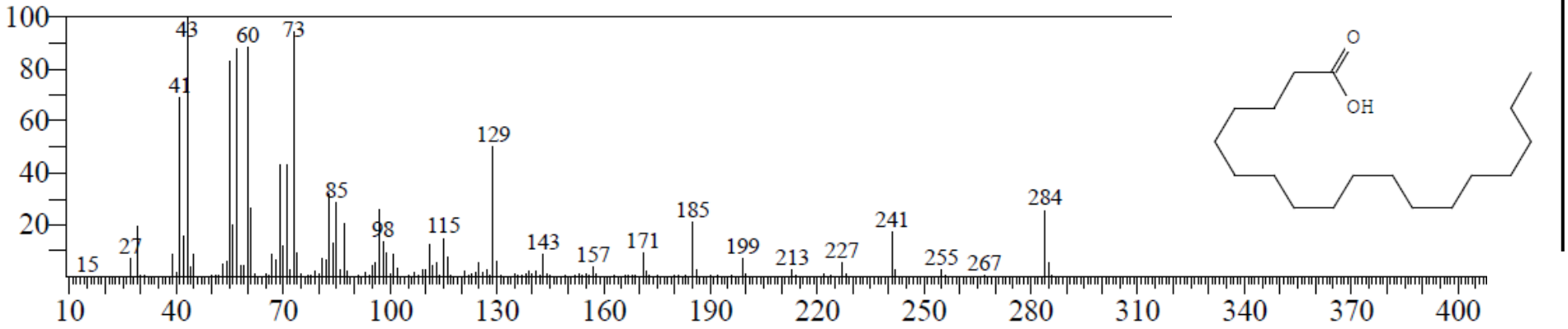

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12 October 2018

Mr B Akwasi (218069763)
Mr J Nwabuife (218084368)
School of Health Sciences
College of Health Sciences
Concordford14@gmail.com

Dear Mr B Akwasi

Protocol: A comparative chemistry of coal herbal medicine and herbal extracts of vermonia amygdaline (bitter leaf), Persea Americana (Avocado) Azardirachta indica (Neem) and Carica papaya (Pawpaw).
Degree: MSc
BREC REF: EXM612/18

I refer to your application to BREC received on 05 October 2018 and wish to advise you that exemption of ethics review has been granted for this study.

This exemption will be **noted** at the next Biomedical Research Ethics Committee meeting to be held on **13 November 2018.**

Yours sincerely

Prof V Rambiritch
Chair: Biomedical Research Ethics Committee

Supervisor: Nlooto@ukzn.ac.za
Co Supervisor: Dr R Kaarpomath
Postgrad admin: Nenep1@ukzn.ac.za

GC-MS ANALYSIS OF COA®
(STANDARD)

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/11/08 3:44:39 PM
Sample Type	: Unknown
Sample Name	: Standard COA
Sample ID	: Akwasi
Vial #	: 9
Injection Volume	: 8.00
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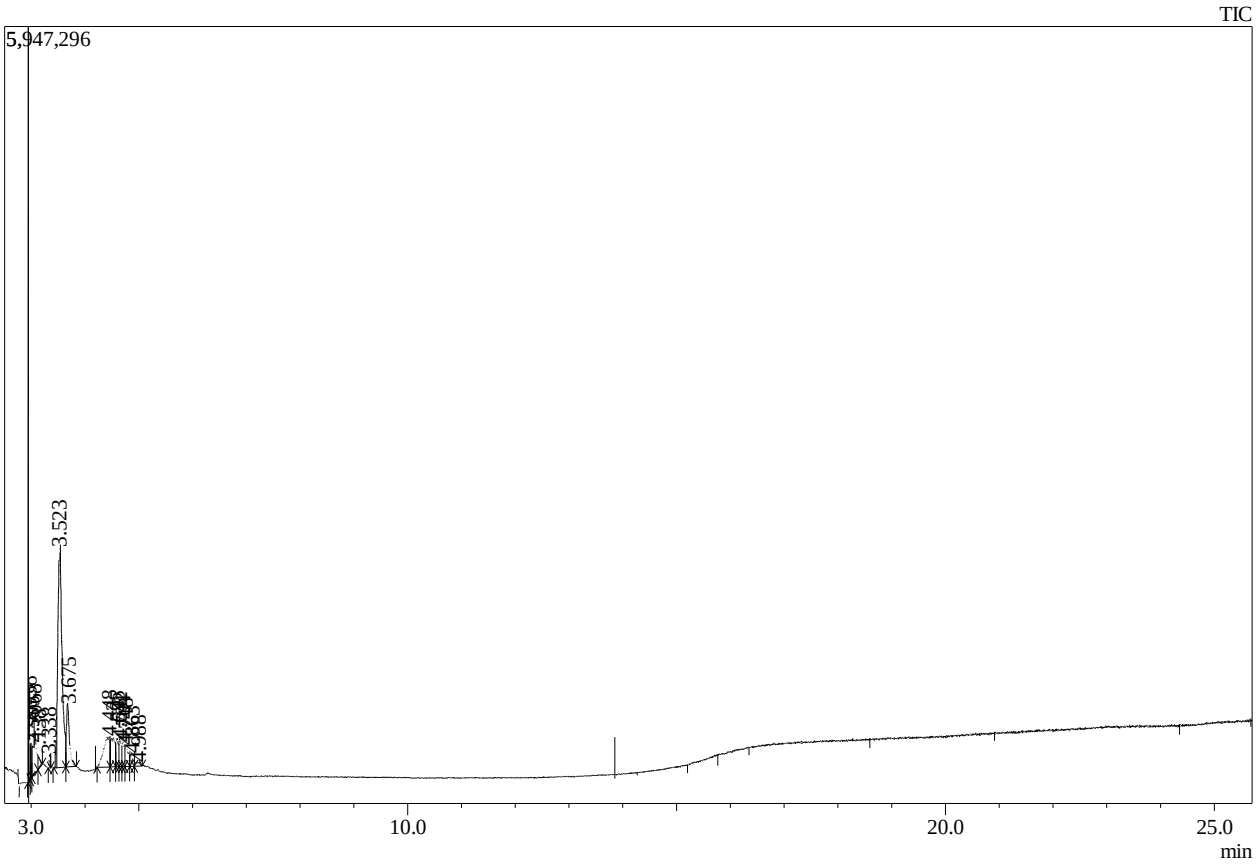
Chromatogram COA-STANDARD



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
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2	2.995	488789	2.28	280026	5.71	1.75	2H-Pyran-2,6(3H)-dione
3	3.068	1449319	6.75	334857	6.83	4.33	3-Piperidinol
4	3.138	230485	1.07	98572	2.01	2.34	Toluene
5	3.338	124156	0.58	103133	2.10	1.20	Acetic acid, butyl ester
6	3.523	9561575	44.53	1733352	35.37	5.52	3-Hexen-1-ol, (E)-
7	3.675	1931871	9.00	497748	10.16	3.88	Formic acid, hexyl ester
8	4.448	1961460	9.13	237515	4.85	8.26	Heptanoic acid
9	4.522	1257864	5.86	226571	4.62	5.55	Butanedioic acid, methyl-
10	4.595	712830	3.32	224076	4.57	3.18	3,4-Dihydroxy-5-methyl-dihydrofuran-2-one
11	4.642	612965	2.85	215262	4.39	2.85	3-Hexenoic acid, (E)-
12	4.704	546450	2.54	188029	3.84	2.91	2-Hexenoic acid
13	4.748	702491	3.27	162534	3.32	4.32	2-Hexenoic acid
14	4.863	345582	1.61	103551	2.11	3.34	2-Hexenoic acid
15	4.988	178753	0.83	29355	0.60	6.09	2-Hexenoic acid
		21473272	100.00	4900941	100.00		

GC-MS ANALYSIS OF *COA*

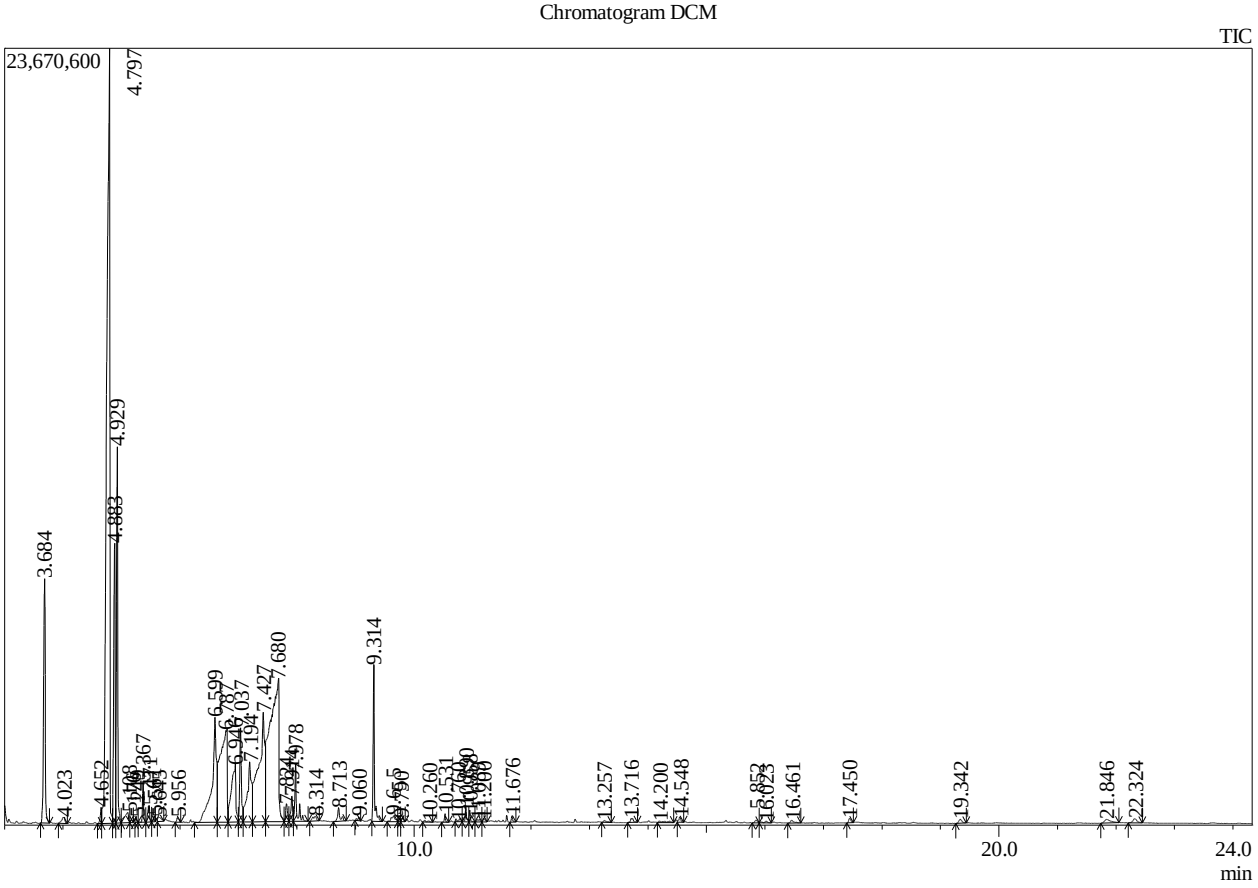
(DCM)- EXTRACT

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/08/01 2:26:03 PM
Sample Type	: Unknown
Sample Name	: DCM
Sample ID	: COA
Vial #	: 76
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\general - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 07 03.qgt

Chromatogram DCM



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	3.684	14384801	4.72	7444540	8.77	1.93	2-Penten-1-ol, (Z)-
2	4.023	690647	0.23	190871	0.22	3.62	Butanoic acid
3	4.652	754226	0.25	494853	0.58	1.52	Butanoic acid, 3-methyl-
4	4.797	88972926	29.19	23608475	27.80	3.77	4-Hexen-1-ol, (Z)-
5	4.883	13923925	4.57	8529009	10.04	1.63	2-Hexen-1-ol, (E)-
6	4.929	17873079	5.86	11465188	13.50	1.56	Formic acid, hexyl ester
7	5.108	1558690	0.51	314312	0.37	4.96	Pentanoic acid
8	5.176	226842	0.07	80949	0.10	2.80	3-Pentanol, 2-chloro-4-methyl-, (R*,S*)-(.+/-.)
9	5.249	221682	0.07	148695	0.18	1.49	o-Xylene
10	5.367	1646337	0.54	1245788	1.47	1.32	Ethanol, 2-butoxy-
11	5.471	1054272	0.35	512462	0.60	2.06	Butanoic acid, 4-hydroxy-
12	5.561	312032	0.10	177993	0.21	1.75	Cyclopentane, 1,2,3,4,5-pentamethyl-
13	5.645	534096	0.18	125586	0.15	4.25	2-Heptanol, acetate
14	5.956	367915	0.12	151555	0.18	2.43	1-Heptene, 4-methyl-
15	6.599	17108345	5.61	3201142	3.77	5.34	2H-Pyran-2,6(3H)-dione
16	6.787	23677541	7.77	2800551	3.30	8.45	Hexanoic acid
17	6.946	9619264	3.16	1762946	2.08	5.46	3-Hexenoic acid, (E)-
18	7.037	5417538	1.78	2668785	3.14	2.03	Methoxymethylaminoacrylonitryl
19	7.194	9404622	3.08	1753858	2.07	5.36	Benzyl alcohol
20	7.427	24628625	8.08	3213569	3.78	7.66	2(3H)-Furanone, 5-ethyldihydro-
21	7.680	46599809	15.29	4269655	5.03	10.91	2-Hexenoic acid, (E)-
22	7.824	1216660	0.40	500418	0.59	2.43	Furyl hydroxymethyl ketone
23	7.914	1624788	0.53	704951	0.83	2.30	.alpha.-Methyl-.alpha.-[4-methyl-3-pentenyl]o
24	7.978	3953682	1.30	1636847	1.93	2.42	1-Cyclohexyl-2-nitropropane-1,3-diol
25	8.314	622326	0.20	133472	0.16	4.66	Maltol
26	8.713	1128405	0.37	382202	0.45	2.95	Cyclohexene,4-butyl-

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	9.060	229019	0.08	118881	0.14	1.93	2H-Pyran-3-ol, 6-ethenyltetrahydro-2,2,6-trim
28	9.314	8078107	2.65	4347571	5.12	1.86	Methyl salicylate
29	9.655	805456	0.26	166059	0.20	4.85	Cyclopentan-1-al, 4-isopropylidene-2-methyl-
30	9.750	258287	0.08	113345	0.13	2.28	1-Methyl-3-butenyl 3-methyl-3-hydroxybutyl e
31	9.790	332168	0.11	97572	0.11	3.40	2-Carene
32	10.260	218990	0.07	35372	0.04	6.19	7-Octen-3-ol, 2,3,6-trimethyl-
33	10.531	349538	0.11	240810	0.28	1.45	Pentadecane
34	10.760	370673	0.12	46898	0.06	7.90	p-Ethoxybenzyl alcohol
35	10.890	995271	0.33	489181	0.58	2.03	2,2'-Bioxepane
36	10.958	437424	0.14	321377	0.38	1.36	2,2'-Bioxepane
37	11.089	232933	0.08	64326	0.08	3.62	Furan, 2,5-diethyltetrahydro-
38	11.200	234369	0.08	91854	0.11	2.55	1,5-Heptadien-4-ol, 3,3,6-trimethyl-
39	11.676	274349	0.09	157378	0.19	1.74	Tetradecane
40	13.257	204387	0.07	58191	0.07	3.51	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4
41	13.716	388184	0.13	113087	0.13	3.43	Z-5-Nonadecene
42	14.200	419644	0.14	50303	0.06	8.34	Epiglobulol
43	14.548	395121	0.13	156244	0.18	2.53	1-Naphthalenol, decahydro-1,4a-dimethyl-7-(1
44	15.852	257378	0.08	82100	0.10	3.13	Oleyl alcohol, trifluoroacetate
45	16.023	241258	0.08	52421	0.06	4.60	1,1,6-trimethyl-3-methylene-2-(3,6,10,13,14-p
46	16.461	341955	0.11	83054	0.10	4.12	2-Methyl-5-(2,6,6-trimethyl-cyclohex-1-enyl)-
47	17.450	342734	0.11	134980	0.16	2.54	Heneicosane
48	19.342	473994	0.16	130876	0.15	3.62	n-Tetracosanol-1
49	21.846	836397	0.27	116123	0.14	7.20	n-Nonadecanol-1
50	22.324	614584	0.20	123912	0.15	4.96	Heneicosane
		304855295	100.00	84910587	100.00		

GC-MS ANALYSIS OF COA®
(HEXANE)

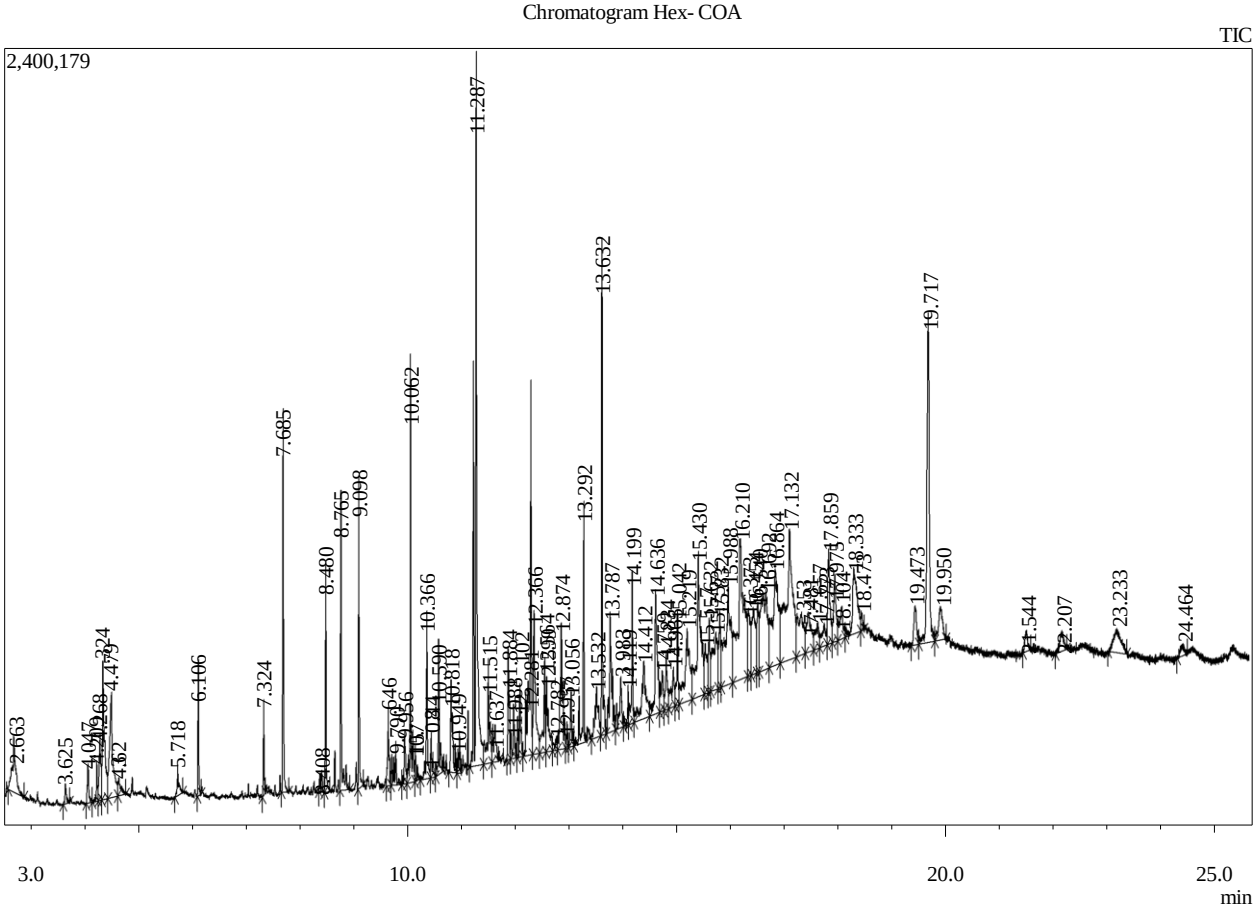
Qualitative Analysis Report

GC-MS ANALYSIS COA HEXANE EXTRACT

Sample Information

Analyzed	: 2018/08/02 12:14:05 AM
Sample Type	: Unknown
Sample Name	: Hexane Extract -COA
Sample ID	: Akwasi
Vial #	: 78
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 07 03.qgt

Chromatogram Hex- COA



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	2.663	778958	1.07	161051	0.67	4.84	Formamide, N,N-dimethyl-
2	3.625	120940	0.17	58621	0.24	2.06	Butanoic acid, 4-hydroxy-
3	4.047	223064	0.31	114252	0.48	1.95	Benzonitrile, 3-hydroxy-
4	4.209	185807	0.26	112389	0.47	1.65	Aniline
5	4.268	471337	0.65	184052	0.77	2.56	3-Hexenoic acid, (E)-
6	4.324	1205361	1.66	413057	1.72	2.92	2H-Pyran-2,6(3H)-dione
7	4.479	1347776	1.86	321557	1.34	4.19	2-Hexenoic acid, (E)-
8	4.623	189033	0.26	54530	0.23	3.47	Dehydromevalonic lactone
9	5.718	194870	0.27	77748	0.32	2.51	1-Octadecyne
10	6.106	420081	0.58	383580	1.60	1.10	Dodecane
11	7.324	268507	0.37	268792	1.12	1.00	Tridecane, 5-methyl-
12	7.685	1230117	1.70	971056	4.05	1.27	Tetradecane
13	8.408	148227	0.20	66700	0.28	2.22	Tetradecane, 4-methyl-
14	8.480	783934	1.08	642633	2.68	1.22	Butylated Hydroxytoluene
15	8.765	1129563	1.56	851685	3.55	1.33	Tetradecane, 5-methyl-
16	9.098	1003128	1.38	895865	3.74	1.12	Hexadecane
17	9.646	393664	0.54	203783	0.85	1.93	Pentadecane, 8-hexyl-
18	9.790	367323	0.51	118908	0.50	3.09	Pentadecane, 8-hexyl-
19	9.956	293210	0.40	149113	0.62	1.97	Heptadecane, 3-methyl-
20	10.062	1656359	2.28	1066922	4.45	1.55	Tetradecane, 5-methyl-
21	10.157	116886	0.16	93947	0.39	1.24	2-Methylhexadec-1-ene
22	10.366	747710	1.03	488455	2.04	1.53	Heptadecane
23	10.448	240092	0.33	127358	0.53	1.89	2-methyltetracosane
24	10.590	610159	0.84	332145	1.39	1.84	Phytol, acetate
25	10.818	753696	1.04	264795	1.11	2.85	Eicosane
26	10.949	204827	0.28	64548	0.27	3.17	Tetradecane, 5-methyl-

Qualitative Analysis Report

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	11.287	4979187	6.86	1886799	7.87	2.64	l-(+)-Ascorbic acid 2,6-dihexadecanoate
28	11.515	805612	1.11	306001	1.28	2.63	Eicosane
29	11.637	445991	0.61	110282	0.46	4.04	Methyl 2-bromo-5-methoxybenzoate
30	11.884	582737	0.80	320143	1.34	1.82	Pentadecane, 8-hexyl-
31	11.988	357830	0.49	152438	0.64	2.35	Trifluoroacetoxy hexadecane
32	12.102	491221	0.68	195788	0.82	2.51	Pyrene
33	12.281	2075165	2.86	166555	0.70	12.46	9-Octadecenal, (Z)-
34	12.366	1219202	1.68	432441	1.80	2.82	Octadecanoic acid
35	12.564	604559	0.83	276795	1.16	2.18	Eicosane
36	12.599	602835	0.83	214205	0.89	2.81	1-Dodecanol, 2-octyl-
37	12.782	116332	0.16	38994	0.16	2.98	Hexadecanoic acid, 2-hydroxyethyl ester
38	12.874	1179933	1.63	381355	1.59	3.09	Pentadecane, 8-hexyl-
39	12.957	108682	0.15	35656	0.15	3.05	2-methyltetracosane
40	13.056	301585	0.42	162837	0.68	1.85	Tetracosane, 11-decyl-
41	13.292	1052914	1.45	667706	2.79	1.58	Docosane, 2,4-dimethyl-
42	13.532	616670	0.85	152108	0.63	4.05	Tetratetracontane
43	13.632	1835402	2.53	1453195	6.06	1.26	Phenol, 2,2'-methylenebis[6-(1,1-dimethylethy
44	13.787	1128730	1.56	336187	1.40	3.36	Hexatriacontane
45	13.983	479001	0.66	145955	0.61	3.28	2-methylhexacosane
46	14.119	169364	0.23	96418	0.40	1.76	2-methyloctacosane
47	14.199	656372	0.90	442347	1.85	1.48	Docosane, 7-hexyl-
48	14.412	995660	1.37	173246	0.72	5.75	Hexatriacontane
49	14.636	1173440	1.62	346092	1.44	3.39	Tetratetracontane
50	14.759	399803	0.55	119226	0.50	3.35	2-methyloctacosane
51	14.834	379649	0.52	159570	0.67	2.38	Tetratetracontane
52	14.965	459810	0.63	117974	0.49	3.90	3,3,13,13-Tetraethylpentadecane
53	15.042	491996	0.68	259778	1.08	1.89	Tetracosane, 11-decyl-
54	15.219	1388505	1.91	212483	0.89	6.53	Tetratetracontane
55	15.430	2164601	2.98	419866	1.75	5.16	Tetratetracontane
56	15.557	532749	0.73	144058	0.60	3.70	2-methylhexacosane
57	15.632	455687	0.63	218721	0.91	2.08	2-methylhexacosane
58	15.757	1119538	1.54	173867	0.73	6.44	Hexadecane, 1-iodo-
59	15.832	507747	0.70	232439	0.97	2.18	Docosane, 2,4-dimethyl-
60	15.988	2178389	3.00	300630	1.25	7.25	Tetratetracontane
61	16.210	3616869	4.99	429166	1.79	8.43	Hexatriacontane
62	16.373	708467	0.98	195273	0.81	3.63	Sulfurous acid, octadecyl 2-propyl ester
63	16.454	1008344	1.39	199745	0.83	5.05	Hexatriacontane
64	16.540	555638	0.77	210273	0.88	2.64	Cholesta-4,6-dien-3-ol, (3.beta.)-
65	16.692	2148935	2.96	236841	0.99	9.07	Octadecane, 5-methyl-
66	16.864	2324649	3.20	295415	1.23	7.87	Hexatriacontane
67	17.132	3524432	4.86	399319	1.67	8.83	Hexatriacontane
68	17.353	1086268	1.50	124464	0.52	8.73	Decane, 1-bromo-2-methyl-
69	17.481	736123	1.01	119307	0.50	6.17	Stigmasterol
70	17.657	389680	0.54	74444	0.31	5.23	6,6-Diethylhoctadecane
71	17.773	339423	0.47	60727	0.25	5.59	Sulfurous acid, octadecyl 2-propyl ester
72	17.859	837788	1.15	278411	1.16	3.01	.gamma.-Sitosterol
73	17.975	676353	0.93	201522	0.84	3.36	2-methylhexacosane
74	18.104	465918	0.64	97390	0.41	4.78	Silane, dimethyl(docosyloxy)butoxy-
75	18.333	1234800	1.70	193522	0.81	6.38	Triacontane, 1-bromo-
76	18.473	129385	0.18	54852	0.23	2.36	Testosterone Valerate
77	19.473	432381	0.60	120761	0.50	3.58	Tetratetracontane
78	19.717	3204314	4.42	995181	4.15	3.22	Acetic acid, (2,4-dichlorophenoxy)-, isooctyl e
79	19.950	540229	0.74	96557	0.40	5.59	Hexatriacontane
80	21.544	232439	0.32	58842	0.25	3.95	Hexatriacontane
81	22.207	322847	0.45	65441	0.27	4.93	Nonahexacontanoic acid
82	23.233	705182	0.97	75586	0.32	9.33	Hexadecanoic acid, octadecyl ester
83	24.464	179221	0.25	39018	0.16	4.59	Nonahexacontanoic acid
		72541212	100.00	23961754	100.00		

GC-MS ANALYSIS OF COA®
(ETHANOL)

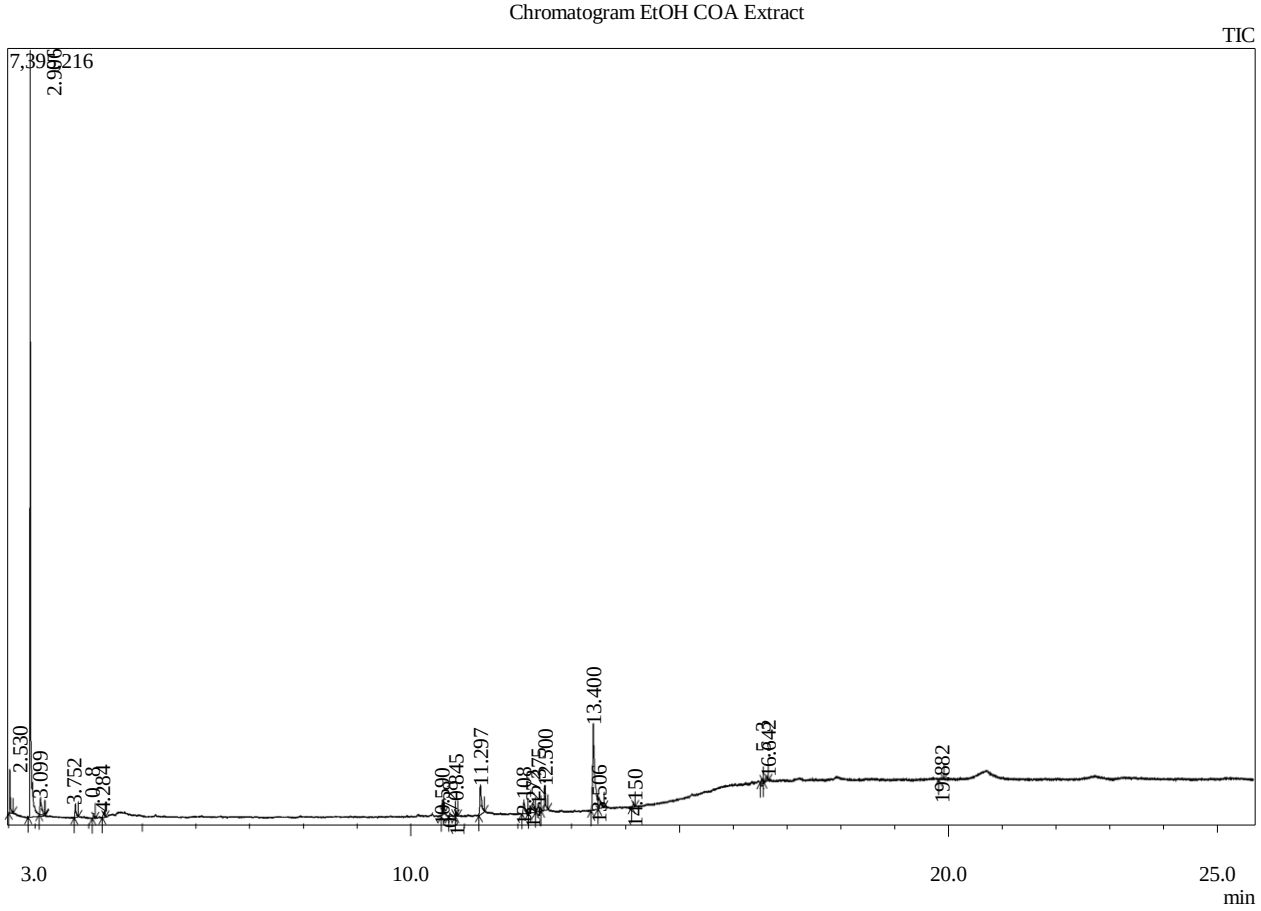
Qualitative Analysis Report

GC-MS ANALYSIS COA ETHANOL EXTRACT

Sample Information

Analyzed	: 2018/08/02 12:50:08 AM
Sample Type	: Unknown
Sample Name	: EtOH COA Extract
Sample ID	: Akwasi
Vial #	: 79
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 07 03.qgt

Chromatogram EtOH COA Extract



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	2.530	285984	2.22	371563	3.76	0.77	Triethylamine
2	2.906	7089544	55.04	7015466	71.01	1.01	Formamide, N,N-dimethyl-
3	3.099	354614	2.75	161407	1.63	2.20	Benzene, chloro-
4	3.752	159969	1.24	128487	1.30	1.25	Butanoic acid, 4-hydroxy-
5	4.088	52912	0.41	48461	0.49	1.09	Benzene, isocyanato-
6	4.284	100589	0.78	56113	0.57	1.79	Aniline
7	10.590	150477	1.17	138940	1.41	1.08	Phytol, acetate
8	10.728	58125	0.45	29354	0.30	1.98	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
9	10.845	51431	0.40	47734	0.48	1.08	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
10	11.297	616739	4.79	280537	2.84	2.20	Pentadecanoic acid
11	12.108	230632	1.79	135127	1.37	1.71	Pyrene
12	12.272	309975	2.41	97108	0.98	3.19	7-Tetradecenal, (Z)-
13	12.375	194855	1.51	62566	0.63	3.11	Octadecanoic acid
14	12.500	374603	2.91	235420	2.38	1.59	Hexadecanamide
15	13.400	2302424	17.88	824212	8.34	2.79	9-Octadecenamide, (Z)-
16	13.506	322573	2.50	107679	1.09	3.00	Octadecanamide
17	14.150	64201	0.50	50028	0.51	1.28	Diisooctyl phthalate
18	16.543	52066	0.40	32696	0.33	1.59	Cholesta-4,6-dien-3-ol, (3.beta.)-
19	16.642	54894	0.43	37590	0.38	1.46	Stigmast-5-en-3-ol, oleate
20	19.882	53484	0.42	18548	0.19	2.88	1,4-Methanoazulen-9-ol, decahydro-1,5,5,8a-t
		12880091	100.00	9879036	100.00		

GC-MS ANALYSIS OF COA®
(ETHYL ACETATE)

Qualitative Analysis Report

COA ETHYL ACETATE EXTRACT

GC-MS ANALYSIS

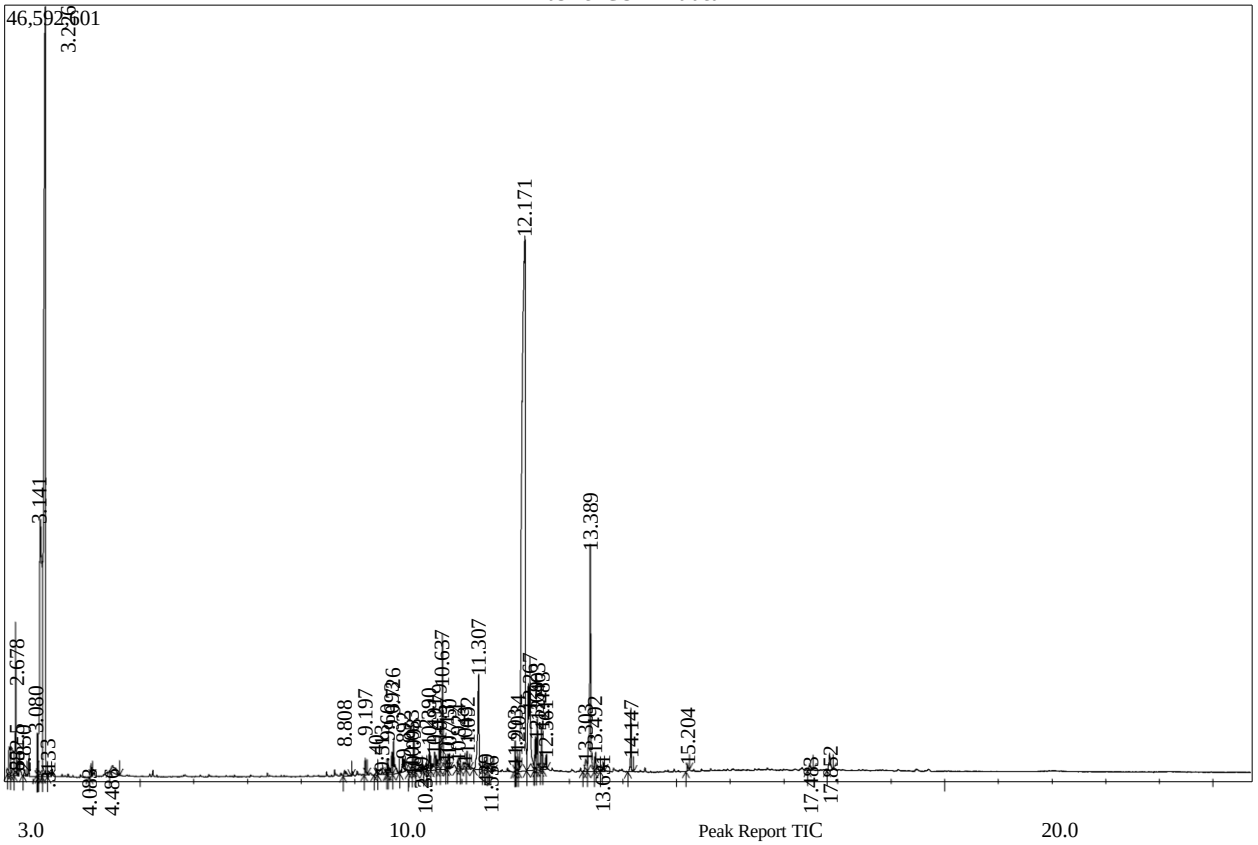
Sample Information

Analyzed : 2018/08/02 1:26:30 AM
Sample Type : Unknown
Sample Name : EtOAc- COA Extract
Sample ID : Akwasi

Vial # : 80
Injection Volume : 1.00

Method File : C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File : C:\GCMSsolution\System\Tune1\Normal Conc - 2018 07 03.qgt

Chromatogram
EtOAc- COA Extract TIC



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	2.555	3431691	0.80	1649895	0.92	2.08	Toluene
2	2.588	5905792	1.38	1683071	0.94	3.51	Spiro[2,4]hepta-4,6-diene
3	2.678	8296405	1.93	8862016	4.93	0.94	Toluene
4	2.850	531202	0.12	321887	0.18	1.65	1,3-Octanediol
5	3.080	996793	0.23	1180274	0.66	0.84	Furfural
6	3.141	51891650	12.08	15230582	8.47	3.41	Benzene, chloro-
7	3.226	97578624	22.72	46143666	25.65	2.11	Benzene, chloro-
8	3.313	673716	0.16	280520	0.16	2.40	Hexyl chloroformate
9	4.083	708808	0.17	751885	0.42	0.94	2-Furancarboxaldehyde, 5-methyl-
10	4.486	2064553	0.48	402532	0.22	5.13	2-Hexenoic acid, (E)-
11	8.808	983653	0.23	334391	0.19	2.94	.alpha.-Calacorene
12	9.197	1073231	0.25	945089	0.53	1.14	4,6-Heptadien-2-one, 3,6-dimethyl-3-(1-methy
13	9.403	467305	0.11	371504	0.21	1.26	Di-epi-.alpha.-cedrene-(I)
14	9.513	1127272	0.26	245917	0.14	4.58	1-Naphthalenol, 1,2,3,4,4a,7,8,8a-octahydro-1
15	9.609	717031	0.17	621897	0.35	1.15	2-Naphthalenemethanol, decahydro-.alpha.,.al
16	9.693	2230671	0.52	1295239	0.72	1.72	Naphthalene, 1,6-dimethyl-4-(1-methylethyl)-
17	9.726	3523123	0.82	2063957	1.15	1.71	Benzene, 1,2,4,5-tetraethyl-
18	9.892	1926500	0.45	890231	0.49	2.16	9-Undecenal, 2,10-dimethyl-
19	10.053	725650	0.17	362707	0.20	2.00	Neoclovene oxide
20	10.098	3479310	0.81	2869991	1.60	1.21	1-Cyclohexanone, 2-methyl-2-(3-methyl-2-oxo
21	10.239	1210327	0.28	383065	0.21	3.16	4-(2-Acetyl-5,5-dimethylcyclopent-2-enyliden
22	10.323	631094	0.15	442009	0.25	1.43	Pentafluoropropionic acid, tridecyl ester
23	10.390	2053703	0.48	1547257	0.86	1.33	2-Naphthalenemethanol, 2,3,4,4a,5,6,7,8-octa
24	10.491	2478934	0.58	1033291	0.57	2.40	7-Oxocholesteryl isocaproate
25	10.579	3409167	0.79	1858172	1.03	1.83	(1S,2E,4S,5R,7E,11E)-Cembra-2,7,11-trien-4,
26	10.637	11557210	2.69	7933844	4.41	1.46	2-Naphthalenemethanol, 2,3,4,4a,5,6,7,8-octa

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	10.713	879058	0.20	496928	0.28	1.77	3,3,5,5-Tetramethyl-4,5-dihydro-3H-benzo[c]a
28	10.750	992751	0.23	848798	0.47	1.17	Phthalic acid, hexyl 4-trifluoromethoxybenzyl
29	10.924	1053068	0.25	576891	0.32	1.83	4,6,10,10-Tetramethyl-5-oxatricyclo[4.4.0.0(1
30	11.043	1189389	0.28	428359	0.24	2.78	3-Heptyn-2-one, 5-cyclopentyl-6-hydroxy-6-m
31	11.092	1720716	0.40	1054833	0.59	1.63	Pentadecanoic acid, 14-methyl-, methyl ester
32	11.307	11269016	2.62	5699367	3.17	1.98	Pentadecanoic acid
33	11.429	1714721	0.40	541332	0.30	3.17	Metolachlor
34	11.536	1721046	0.40	488170	0.27	3.53	3-Buten-2-one, 4-(2-hydroxy-2,6,6-trimethylc
35	11.993	532876	0.12	407903	0.23	1.31	n-Nonadecanol-1
36	12.034	1496316	0.35	1284267	0.71	1.17	Tricyclo[3.3.1.1(3,7)]decane, 1-(phenylmethyl
37	12.171	134992357	31.43	32159456	17.88	4.20	Benzene, 1,1'-(1,3-butadiyne-1,4-diyl)bis-
38	12.267	19968705	4.65	6511323	3.62	3.07	cis-9-Hexadecenal
39	12.369	2527124	0.59	1996069	1.11	1.27	Octadecanoic acid
40	12.403	4204492	0.98	3121640	1.74	1.35	1,4-Ethenoanthracene, 1,4-dihydro-
41	12.483	2786109	0.65	2594846	1.44	1.07	Hexadecanamide
42	12.561	1277369	0.30	811146	0.45	1.57	Heneicosane
43	13.303	1898633	0.44	726686	0.40	2.61	3-Furan-2-yl-acrylic acid 4-[(2-chloro-benzoyl
44	13.389	19513229	4.54	13280547	7.38	1.47	9-Octadecenamide, (Z)-
45	13.492	1891488	0.44	1129293	0.63	1.67	Octadecanamide
46	13.631	640000	0.15	560934	0.31	1.14	Phenol, 2,2'-methylenebis[6-(1,1-dimethylethy
47	14.147	3930903	0.92	3667898	2.04	1.07	Diisooctyl phthalate
48	15.204	556045	0.13	438181	0.24	1.27	13-Docosenamide, (Z)-
49	17.483	774186	0.18	381948	0.21	2.03	Stigmasterol
50	17.852	2290068	0.53	984875	0.55	2.33	.gamma.-Sitosterol
		429493080	100.00	179896579	100.00		

GC-MS ANALYSIS OF *VERNONIA AMYGDALINA*

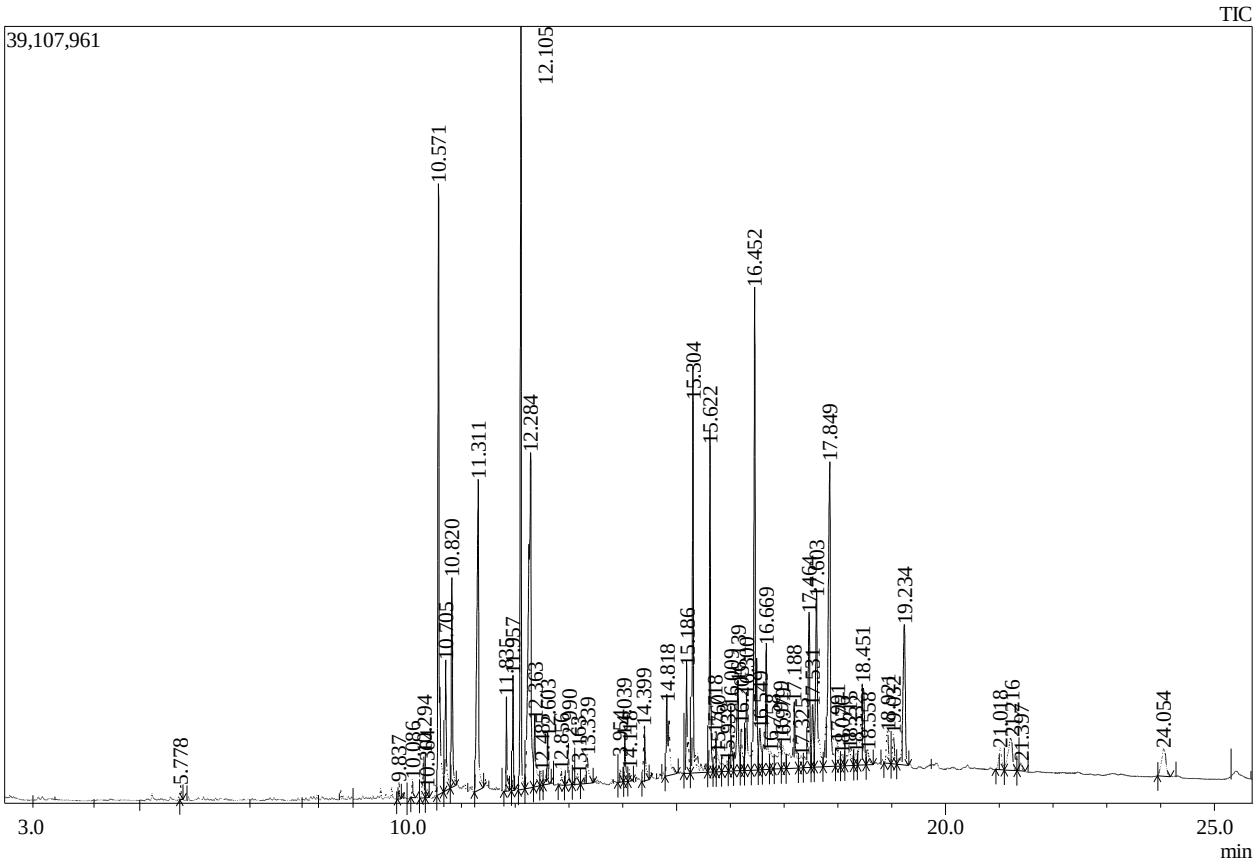
(DCM)- Ghana

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 9:02:40 PM
Sample Type	: Unknown
Sample Name	: VA-DCM-Ghana
Sample ID	: Akwasi
Vial #	: 41
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram VA-DCM-Ghana



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	5.778	881097	0.13	726286	0.24	1.21	3,4-Dimethyl-2-pentanone
2	9.837	897101	0.13	794506	0.26	1.13	Heptadecanal
3	10.086	1165830	0.17	894254	0.29	1.30	Tetradecanoic acid
4	10.294	3875585	0.56	2230507	0.73	1.74	Acetic acid, 2-(2,2,6-trimethyl-7-oxa-bicyclo[4
5	10.364	999375	0.15	458528	0.15	2.18	2,6,8-Trimethylbicyclo[4.2.0]oct-2-ene-1,8-di
6	10.571	48610023	7.08	30733448	9.99	1.58	Phytol, acetate
7	10.705	10968697	1.60	6544932	2.13	1.68	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
8	10.820	16581367	2.41	10400815	3.38	1.59	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
9	11.311	37364304	5.44	15581853	5.07	2.40	Pentadecanoic acid
10	11.835	5868939	0.85	4727780	1.54	1.24	9-Octadecen-1-ol, (Z)-
11	11.957	5954359	0.87	5682186	1.85	1.05	1-Heneicosanol
12	12.105	52972208	7.71	37722450	12.27	1.40	Phytol
13	12.284	59060153	8.60	16871347	5.49	3.50	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-
14	12.363	8290349	1.21	3401625	1.11	2.44	Octadecanoic acid
15	12.485	1814993	0.26	694961	0.23	2.61	Cyclopropanemethanol, 2-methyl-2-(4-methyl-
16	12.603	3731836	0.54	2406030	0.78	1.55	Phytol, acetate
17	12.856	1385375	0.20	754551	0.25	1.84	E-10,13,13-Trimethyl-11-tetradecen-1-ol aceta
18	12.990	2779561	0.40	1925104	0.63	1.44	1-Heneicosanol
19	13.163	1630238	0.24	515512	0.17	3.16	Methyl 12-oxo-9-dodecenoate
20	13.339	4484665	0.65	1439669	0.47	3.12	Eicosanoic acid
21	13.954	1582875	0.23	664048	0.22	2.38	1-Octadecanesulphonyl chloride
22	14.039	3965335	0.58	2326952	0.76	1.70	Hexadecanoic acid, 2-hydroxy-1-(hydroxymet
23	14.118	928680	0.14	583221	0.19	1.59	Diisooctyl phthalate
24	14.399	7441805	1.08	2717062	0.88	2.74	Preg-4-en-3-one, 17.alpha.-hydroxy-17.beta.-c
25	14.818	12942917	1.88	3940618	1.28	3.28	12-Methyl-E,E-2,13-octadecadien-1-ol
26	15.186	9337238	1.36	5201412	1.69	1.80	13-Docosenamide, (Z)-

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	15.304	27888842	4.06	19634726	6.38	1.42	Squalene
28	15.622	22804189	3.32	15240456	4.96	1.50	Tetratetracontane
29	15.718	2922623	0.43	1924203	0.63	1.52	Oxirane, 2,2-dimethyl-3-(3,7,12,16,20-pentam
30	15.760	3497493	0.51	967982	0.31	3.61	Hexadeca-2,6,10,14-tetraen-1-ol, 3,7,11,16-tet
31	15.938	2652839	0.39	514994	0.17	5.15	17-(1,5-Dimethyl-hex-2-enyl)-10,13-dimethyl-
32	16.009	7051965	1.03	3102219	1.01	2.27	Tetratetracontane
33	16.139	12631996	1.84	4451875	1.45	2.84	2-(8-Chloro-3,7-dimethyl-octa-2,6-dienyloxy)t
34	16.205	5089864	0.74	2274109	0.74	2.24	Hexadecanal
35	16.300	9998034	1.46	3698920	1.20	2.70	.gamma.-Tocopherol
36	16.452	48271235	7.03	23512079	7.65	2.05	Tetratetracontane
37	16.549	6106688	0.89	2026150	0.66	3.01	9-Octadecanone
38	16.669	15866580	2.31	6029833	1.96	2.63	Vitamin E
39	16.758	3020953	0.44	875257	0.28	3.45	9-Octadecenoic acid, 1,2,3-propanetriyl ester,
40	16.919	7214188	1.05	1502641	0.49	4.80	Tetratetracontane
41	16.979	4870583	0.71	1151497	0.37	4.23	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
42	17.188	13200336	1.92	3228711	1.05	4.09	Hexadecanal
43	17.325	2806246	0.41	590271	0.19	4.75	Ergost-5-en-3-ol, (3.beta.)-
44	17.464	18831707	2.74	7752465	2.52	2.43	Stigmasterol
45	17.531	6970876	1.01	3123133	1.02	2.23	Docosyl trifluoroacetate
46	17.603	23912086	3.48	8613996	2.80	2.78	Chondrillasterol
47	17.849	48239190	7.02	14813625	4.82	3.26	Chondrillasterol
48	17.991	4002729	0.58	1204679	0.39	3.32	Methanesulfonic acid, 2-(3-hydroxy-4,4,10,13
49	18.076	2020826	0.29	568880	0.18	3.55	Stigmasterol
50	18.215	4841222	0.70	829639	0.27	5.84	4,4,6a,6b,8a,11,11,14b-Octamethyl-1,4,4a,5,6,
51	18.332	2000489	0.29	554655	0.18	3.61	2,2,4-Trimethyl-3-(3,8,12,16-tetramethyl-hept
52	18.451	19229023	2.80	4039320	1.31	4.76	Hexadecanal
53	18.558	2196393	0.32	762203	0.25	2.88	.alpha.-Amyrin
54	18.921	4570887	0.67	1590052	0.52	2.87	Tetracosyl pentafluoropropionate
55	19.032	3831667	0.56	1491578	0.49	2.57	2-Nonadecanone
56	19.234	21801442	3.17	7011728	2.28	3.11	Phytol, acetate
57	21.018	4302276	0.63	1096575	0.36	3.92	E,E,Z-1,3,12-Nonadecatriene-5,14-diol
58	21.216	9288796	1.35	1610124	0.52	5.77	Phytol, acetate
59	21.397	1342660	0.20	425629	0.14	3.15	Oxirane, 2,2-dimethyl-3-(3,7,12,16,20-pentam
60	24.054	8007943	1.17	1359557	0.44	5.89	Phytol, acetate
		686799771	100.00	307513418	100.00		

GC-MS ANALYSIS OF *VERNONIA AMYGDALINA*

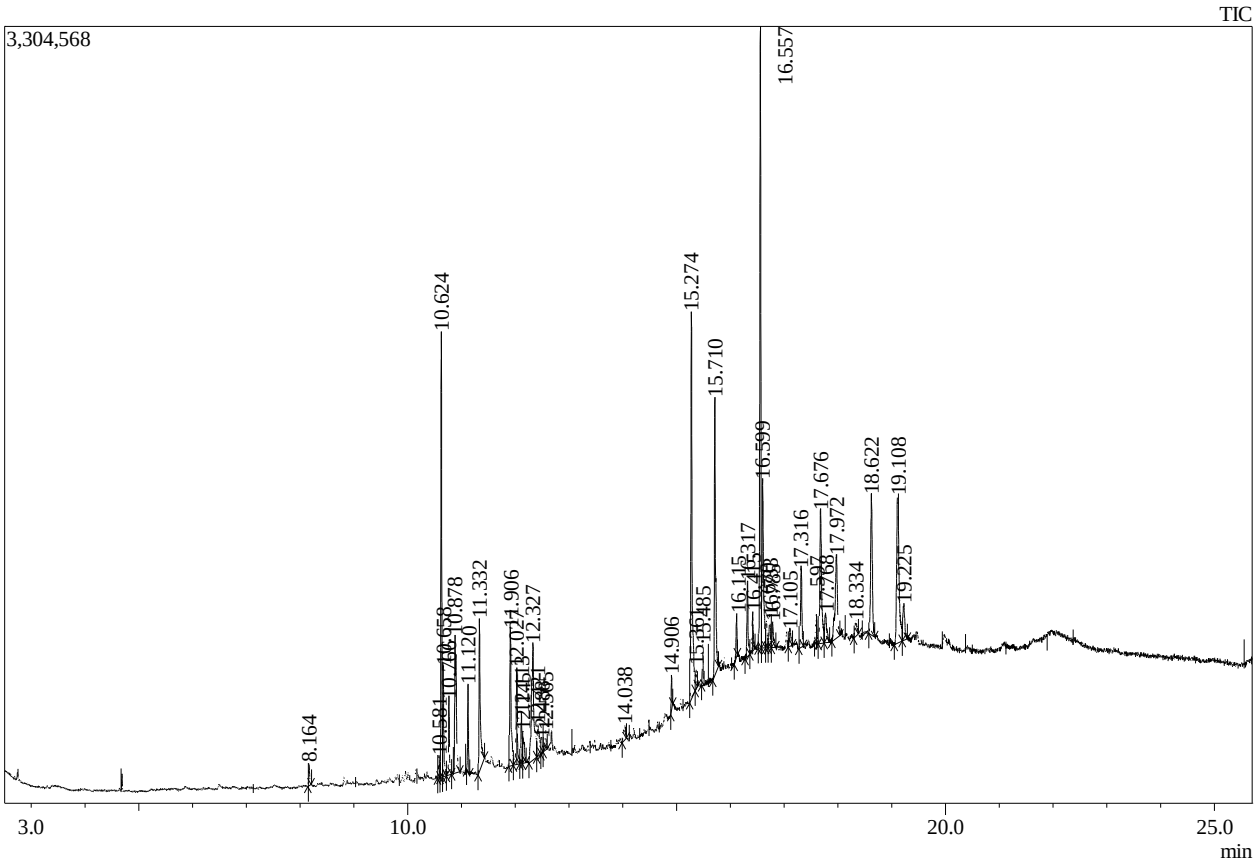
(DCM)- South Africa

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 4:13:22 PM
Sample Type	: Unknown
Sample Name	: VA-DCM-SA
Sample ID	: Akwasi
Vial #	: 33
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram VA-DCM-SA



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	8.164	124297	0.43	94622	0.55	1.31	Undecane, 3,8-dimethyl-
2	10.581	109371	0.37	94789	0.55	1.15	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
3	10.624	2046288	7.01	1877804	10.93	1.09	Phytol, acetate
4	10.658	602137	2.06	472150	2.75	1.28	2-Pentadecanone, 6,10,14-trimethyl-
5	10.766	398755	1.37	323468	1.88	1.23	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
6	10.878	1206715	4.13	578325	3.37	2.09	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
7	11.120	376794	1.29	359916	2.10	1.05	1-Hexacosene
8	11.332	1115619	3.82	639501	3.72	1.74	Pentadecanoic acid
9	11.906	968756	3.32	584006	3.40	1.66	9-Octadecen-1-ol, (Z)-
10	12.027	682153	2.34	403899	2.35	1.69	1-Octadecanol
11	12.113	260382	0.89	217880	1.27	1.20	2-Ethylnon-1-en-3-ol
12	12.145	247533	0.85	135659	0.79	1.82	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
13	12.327	1556556	5.33	489561	2.85	3.18	13-Octadecenal, (Z)-
14	12.421	398323	1.36	138913	0.81	2.87	Octadecanoic acid
15	12.488	124315	0.43	64778	0.38	1.92	Pentadecanal-
16	12.565	142053	0.49	82009	0.48	1.73	11,13-Dimethyl-12-tetradecen-1-ol acetate
17	14.038	149894	0.51	62454	0.36	2.40	2-methyltetracosane
18	14.906	152034	0.52	141422	0.82	1.08	Tetratetracontane
19	15.274	2605200	8.92	1618064	9.42	1.61	13-Docosenamide, (Z)-
20	15.361	117448	0.40	96741	0.56	1.21	Tetradecanamide
21	15.485	186776	0.64	175395	1.02	1.06	Hexadecanal
22	15.710	1716603	5.88	1146698	6.68	1.50	2-methyloctacosane
23	16.115	226684	0.78	182737	1.06	1.24	Tetratetracontane
24	16.317	468033	1.60	317285	1.85	1.48	Hexadecanal
25	16.415	256931	0.88	167816	0.98	1.53	.gamma.-Tocopherol
26	16.557	3502682	12.00	2598139	15.13	1.35	Tetratetracontane

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	16.599	1372315	4.70	714454	4.16	1.92	n-Tetracosanol-1
28	16.670	214356	0.73	112527	0.66	1.90	2-Nonadecanone
29	16.733	234440	0.80	134684	0.78	1.74	Stigmast-5-en-3-ol, oleate
30	16.785	192236	0.66	108953	0.63	1.76	1,19-Eicosadiene
31	17.105	153373	0.53	75126	0.44	2.04	Tetracosyl acetate
32	17.316	581332	1.99	337887	1.97	1.72	Oxirane, heptadecyl-
33	17.597	202365	0.69	120079	0.70	1.69	2-methylhexacosane
34	17.676	1300732	4.46	569094	3.31	2.29	Tetracosyl trifluoroacetate
35	17.768	328712	1.13	125667	0.73	2.62	2-Nonadecanone
36	17.972	1062396	3.64	353809	2.06	3.00	.gamma.-Sitosterol
37	18.334	179944	0.62	67616	0.39	2.66	Triacontyl acetate
38	18.622	1269150	4.35	601834	3.50	2.11	Hexadecanal
39	19.108	1964694	6.73	632535	3.68	3.11	Tetracosyl trifluoroacetate
40	19.225	395354	1.35	155050	0.90	2.55	2-Nonadecanone
		29193731	100.00	17173346	100.00		

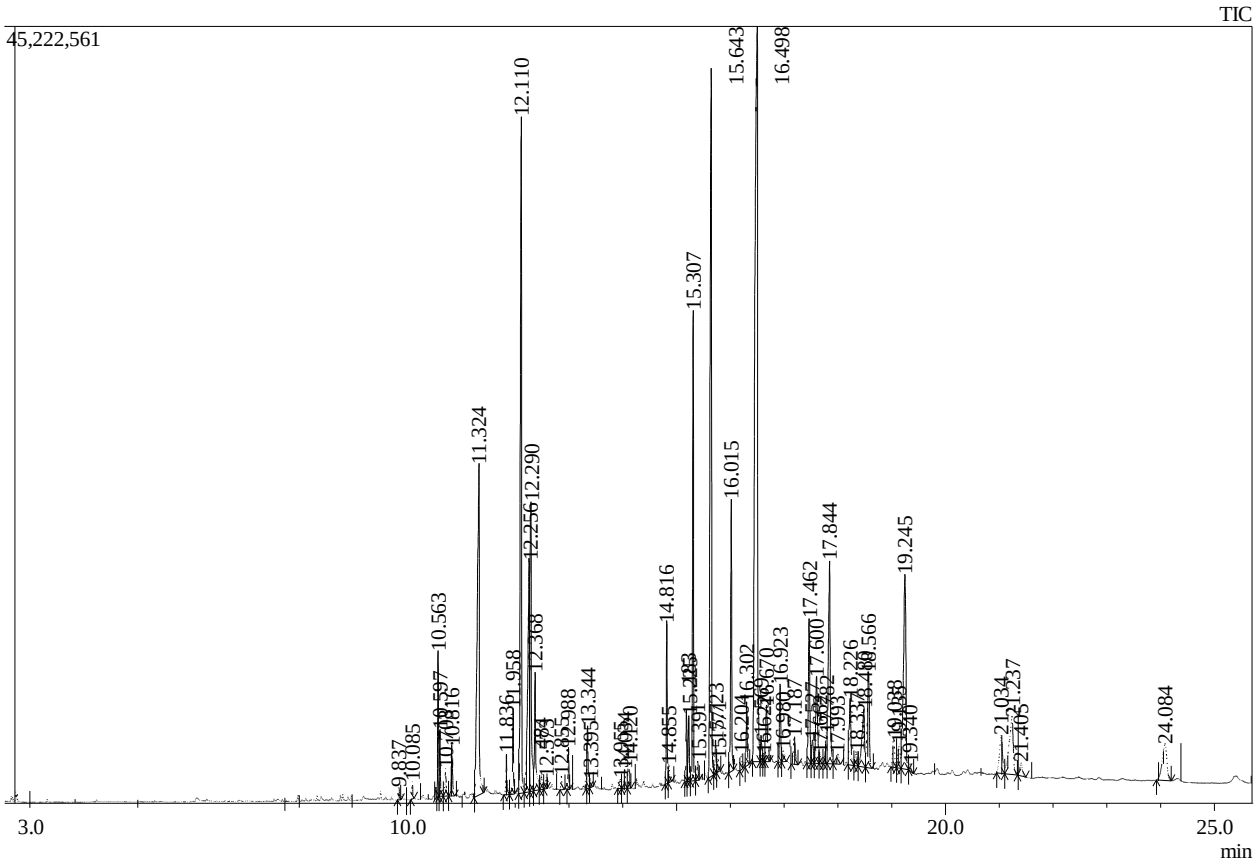
GC-MS ANALYSIS OF *VERNONIA AMYGDALINA* (HEXANE)- Ghana

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 8:26:56 PM
Sample Type	: Unknown
Sample Name	: V A-Hex-Ghana
Sample ID	: Akwasi
Vial #	: 40
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram VA-HEX-Ghana



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	9.837	769743	0.10	701234	0.19	1.10	Heptadecanal
2	10.085	1138544	0.15	1009474	0.28	1.13	Tetradecanoic acid
3	10.563	9229048	1.20	8215009	2.26	1.12	Phytol, acetate
4	10.597	4414447	0.57	3837959	1.05	1.15	2-Pentadecanone, 6,10,14-trimethyl-
5	10.703	2705229	0.35	1562678	0.43	1.73	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
6	10.816	4722113	0.61	2912503	0.80	1.62	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
7	11.324	52517995	6.82	19261863	5.29	2.73	Pentadecanoic acid
8	11.836	3277261	0.43	2341653	0.64	1.40	9-Octadecen-1-ol, (Z)-
9	11.958	5259160	0.68	4923016	1.35	1.07	1-Heneicosanol
10	12.110	63535434	8.25	39066723	10.73	1.63	Phytol
11	12.256	30965505	4.02	13500096	3.71	2.29	9,12-Octadecadienoic acid (Z,Z)-
12	12.290	32379915	4.21	16581062	4.55	1.95	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-
13	12.368	11910288	1.55	6892852	1.89	1.73	Octadecanoic acid
14	12.484	2215785	0.29	797248	0.22	2.78	Cyclopropanemethanol, 2-methyl-2-(4-methyl-
15	12.573	1275869	0.17	631441	0.17	2.02	1-Hexadecanol, 2-methyl-
16	12.855	1128528	0.15	812201	0.22	1.39	E-10,13,13-Trimethyl-11-tetradecen-1-ol aceta
17	12.988	2546477	0.33	2456650	0.67	1.04	n-Nonadecanol-1
18	13.344	5422732	0.70	3565878	0.98	1.52	Eicosanoic acid
19	13.395	860058	0.11	501841	0.14	1.71	2,6,10,14,18-Pentamethyl-2,6,10,14,18-eicosa
20	13.955	1060431	0.14	550809	0.15	1.93	2-methyloctacosane
21	14.034	1616303	0.21	1097276	0.30	1.47	Hexadecanoic acid, 2-hydroxy-1-(hydroxymet
22	14.120	1568266	0.20	1395346	0.38	1.12	Diisooctyl phthalate
23	14.816	10744963	1.40	9409559	2.58	1.14	2-methyloctacosane
24	14.855	1864484	0.24	1054572	0.29	1.77	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-
25	15.183	5215420	0.68	4136639	1.14	1.26	9-Octadecenamide, (Z)-
26	15.225	5179383	0.67	3808556	1.05	1.36	Heneicosane

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	15.307	32133414	4.17	27001978	7.42	1.19	Squalene
28	15.391	1501687	0.20	1159625	0.32	1.29	Hexadecanal
29	15.643	78426983	10.19	40858065	11.22	1.92	Tetratetracontane
30	15.723	2505569	0.33	1937133	0.53	1.29	1,6,10,14,18,22-Tetracosahexaen-3-ol, 2,6,10,
31	15.771	1386520	0.18	755575	0.21	1.84	Oxirane, 2,2-dimethyl-3-(3,7,12,16,20-pentam
32	16.015	20991464	2.73	15684562	4.31	1.34	Tetratetracontane
33	16.204	1448853	0.19	983679	0.27	1.47	Hexadecanal
34	16.302	4864351	0.63	3669115	1.01	1.33	.gamma.-Tocopherol
35	16.498	150899546	19.60	42626669	11.71	3.54	Tetratetracontane
36	16.569	2700948	0.35	1546960	0.42	1.75	2-Nonadecanone
37	16.622	861977	0.11	479698	0.13	1.80	Stigmast-5-en-3-ol, oleate
38	16.670	7004232	0.91	3286146	0.90	2.13	Vitamin E
39	16.923	7141864	0.93	4495347	1.23	1.59	Tetratetracontane
40	16.980	1562392	0.20	673228	0.18	2.32	11,13-Dimethyl-12-tetradecen-1-ol acetate
41	17.187	4001839	0.52	1605988	0.44	2.49	Ergosterol
42	17.462	15673863	2.04	8323491	2.29	1.88	Cholest-22-ene-21-ol, 3,5-dehydro-6-methoxy
43	17.527	3168869	0.41	1327192	0.36	2.39	Triacontyl trifluoroacetate
44	17.600	11813013	1.53	5054987	1.39	2.34	Chondrillasterol
45	17.662	1287899	0.17	666646	0.18	1.93	.gamma.-Ergostenol
46	17.782	3282397	0.43	1791749	0.49	1.83	Phytol, acetate
47	17.844	27789991	3.61	11606866	3.19	2.39	Chondrillasterol
48	17.993	2055191	0.27	612769	0.17	3.35	9,19-Cyclolanost-23-ene-3,25-diol, 3-acetate,
49	18.226	8719520	1.13	3879128	1.07	2.25	4,4,6a,6b,8a,11,11,14b-Octamethyl-1,4,4a,5,6,
50	18.337	2274805	0.30	837055	0.23	2.72	Lup-20(29)-en-3-ol, acetate, (3.beta.)-
51	18.480	11224234	1.46	3330496	0.91	3.37	dl-.alpha.-Tocopherol
52	18.566	14619549	1.90	5538058	1.52	2.64	.alpha.-Amyrin
53	19.038	4376566	0.57	1674959	0.46	2.61	2-Nonadecanone
54	19.135	3965265	0.52	1415880	0.39	2.80	Lanosterol
55	19.245	35602506	4.62	11324134	3.11	3.14	Phytol, acetate
56	19.340	913279	0.12	404499	0.11	2.26	Hexadecanoic acid, octadecyl ester
57	21.034	8871453	1.15	2206548	0.61	4.02	E,E,Z-1,3,12-Nonadecatriene-5,14-diol
58	21.237	17838439	2.32	3361694	0.92	5.31	Phytol, acetate
59	21.405	2638012	0.34	783765	0.22	3.37	Hexadecanoic acid, octadecyl ester
60	24.084	12828621	1.67	2141716	0.59	5.99	Phytol, acetate
		769898492	100.00	364069538	100.00		

GC-MS ANALYSIS OF *VERNONIA AMYGDALINA*

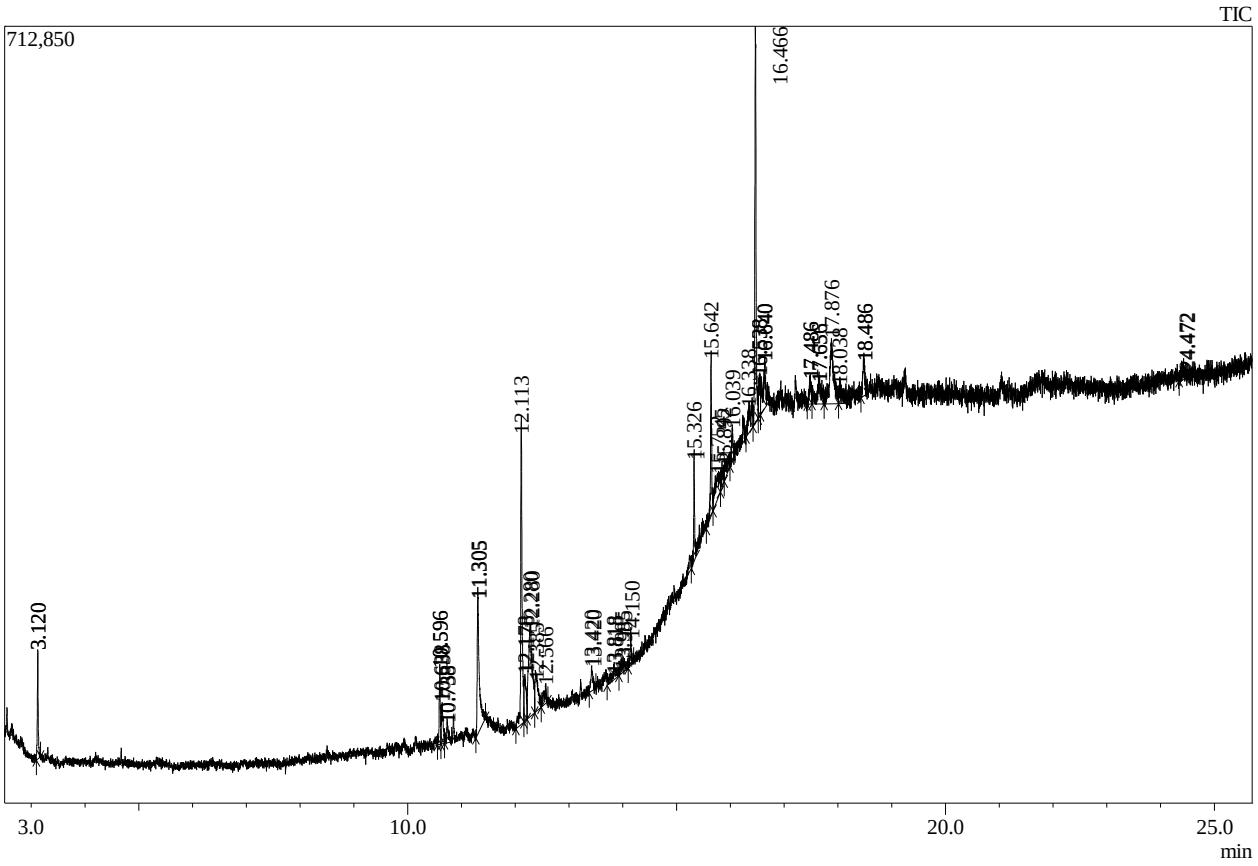
(HEXANE)- South Africa

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/08/02 2:02:44 AM
Sample Type	: Unknown
Sample Name	: VA-HEX(SA)
Sample ID	: Akwasi
Vial #	: 81
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 07 03.qgt

Chromatogram VA-HEX(SA)



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	3.120	117887	2.62	90915	5.14	1.30	Benzene, chloro-
2	10.596	107147	2.38	68602	3.88	1.56	Phytol, acetate
3	10.638	79251	1.76	33177	1.88	2.39	9-(3,3-Dimethyloxiran-2-yl)-2,7-dimethylnona
4	10.738	49084	1.09	16987	0.96	2.89	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
5	11.305	353396	7.86	129368	7.32	2.73	Pentadecanoic acid
6	12.113	523562	11.64	274165	15.51	1.91	Pyrene
7	12.178	111578	2.48	38968	2.20	2.86	Phytol
8	12.280	414474	9.22	81439	4.61	5.09	cis-9-Hexadecenal
9	12.385	142020	3.16	33416	1.89	4.25	Octadecanoic acid
10	12.566	53647	1.19	16201	0.92	3.31	Sulfurous acid, 2-propyl tridecyl ester
11	13.420	60907	1.35	20774	1.18	2.93	13-Docosenamide, (Z)-
12	13.818	69784	1.55	6040	0.34	11.55	Isosteviol
13	13.985	52920	1.18	8214	0.46	6.44	1,2-Epoxynonane
14	14.150	50998	1.13	29238	1.65	1.74	Bis(tridecyl) phthalate
15	15.326	125413	2.79	93587	5.29	1.34	Squalene
16	15.642	218742	4.86	131763	7.45	1.66	Hexatriacontane
17	15.752	137086	3.05	21026	1.19	6.52	Cyclopropa[5,6]stigmast-22-en-3-ol, 3',6-dihy
18	15.845	55928	1.24	15453	0.87	3.62	17-Acetyl-14-hydroxy-16-methoxy-10,13-dim
19	15.892	60125	1.34	14308	0.81	4.20	Trimethylsilyl 3-methyl-4-[(trimethylsilyl)oxy]
20	16.039	52347	1.16	29124	1.65	1.80	2-methyltetracosane
21	16.338	113661	2.53	19890	1.13	5.71	.gamma.-Tocopherol
22	16.466	587218	13.06	354537	20.06	1.66	Hexatriacontane
23	16.538	70353	1.56	35596	2.01	1.98	Cholesta-4,6-dien-3-ol, (3.beta.)-
24	16.640	147643	3.28	41364	2.34	3.57	Stigmast-5-en-3-ol, oleate
25	17.486	64630	1.44	23217	1.31	2.78	Octadecane, 1,1'-[1,3-propanediylbis(oxy)]bis
26	17.656	133959	2.98	22637	1.28	5.92	d-Glucitol, 2,5-anhydro-1-O-octyl-

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	17.876	319758	7.11	55612	3.15	5.75	.gamma.-Sitosterol
28	18.038	65900	1.47	17172	0.97	3.84	2-t-Butyl-5-(dimethoxy-phosphoryl)-3-methyl-
29	18.486	105390	2.34	33909	1.92	3.11	Oxirane, hexadecyl-
30	24.472	52042	1.16	10874	0.62	4.79	2-t-Butyl-5-(dimethoxy-phosphoryl)-3-methyl-
		4496850	100.00	1767573	100.00		

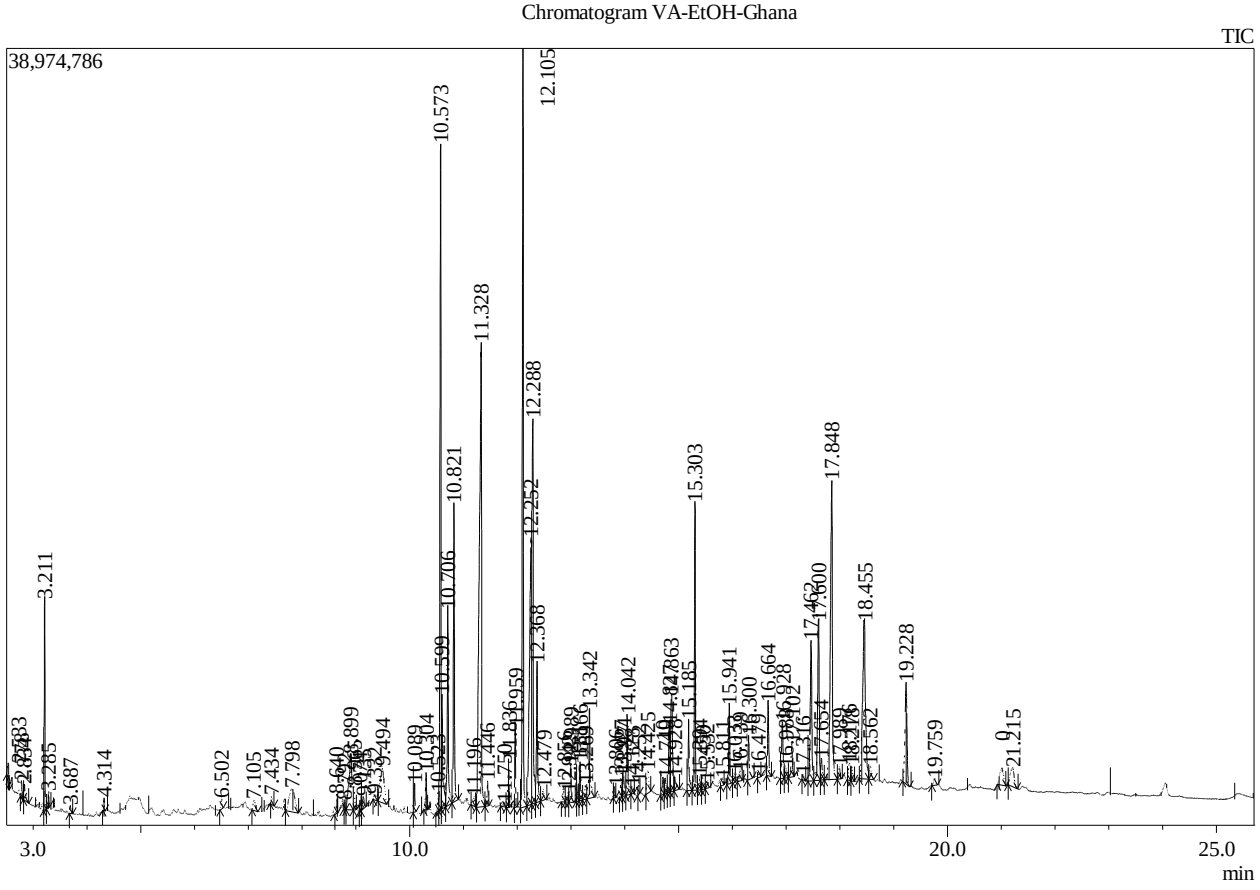
GC-MS ANALYSIS OF *VERNONIA AMYGDALINA* (ETHANOL)- Ghana

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 9:38:40 PM
Sample Type	: Unknown
Sample Name	: VA-EtOH-Ghana
Sample ID	: Akwasi
Vial #	: 42
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram VA-EtOH-Ghana



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	2.533	1497907	0.25	1016738	0.32	1.47	2,3-Hexanedione
2	2.778	817387	0.14	848887	0.27	0.96	2,3-Butanediol, [R-(R*,R*)]-
3	2.834	1344181	0.23	774024	0.24	1.74	2,3-Butanediol
4	3.211	8873596	1.49	10627624	3.33	0.83	Methoxyacetic acid, 2-pentyl ester
5	3.285	697983	0.12	843195	0.26	0.83	3-Hexanol, 2-methyl-
6	3.687	525932	0.09	388523	0.12	1.35	Butyrolactone
7	4.314	535408	0.09	633513	0.20	0.85	Pentane, 1,1-diethoxy-
8	6.502	752380	0.13	552345	0.17	1.36	Benzeneacetic acid
9	7.105	1595578	0.27	526813	0.16	3.03	Valeric acid, 2,3-epoxy-3,4-dimethyl-, ethyl es
10	7.434	574908	0.10	433782	0.14	1.33	(E)-1-(2,3,6-trimethylphenyl)buta-1,3-diene (T
11	7.798	8439601	1.42	1132290	0.35	7.45	Erythritol
12	8.640	846428	0.14	933023	0.29	0.91	Phosphoric acid, diethyl octyl ester
13	8.792	667748	0.11	464809	0.15	1.44	Dodecanoic acid
14	8.899	11524959	1.94	2789786	0.87	4.13	D-Allose
15	8.965	2023419	0.34	1131790	0.35	1.79	.beta.-D-Glucopyranose, 1,6-anhydro-
16	9.074	520825	0.09	559735	0.18	0.93	Hexadecane
17	9.135	1459582	0.25	630144	0.20	2.32	.beta.-d-Ribopyranoside, methyl, 3-acetate
18	9.332	1084259	0.18	222581	0.07	4.87	Megastigmatrienone
19	9.494	9330547	1.57	1842562	0.58	5.06	d-Glycero-d-tallo-heptose
20	10.089	1487951	0.25	1419989	0.44	1.05	Tetradecanoic acid
21	10.304	2572518	0.43	1809967	0.57	1.42	5-Isopropyl-6-methyl-hepta-3,5-dien-2-ol
22	10.523	1215289	0.20	939093	0.29	1.29	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
23	10.573	42382536	7.13	32746889	10.25	1.29	Phytol, acetate
24	10.599	8837778	1.49	5683858	1.78	1.55	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
25	10.706	14338603	2.41	9787188	3.06	1.47	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
26	10.821	19038662	3.20	14995126	4.69	1.27	3,7,11,15-Tetramethyl-2-hexadecen-1-ol

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	11.196	1105195	0.19	486646	0.15	2.27	1-Hexadecen-3-ol, 3,5,11,15-tetramethyl-
28	11.328	68111282	11.46	23252197	7.28	2.93	Pentadecanoic acid
29	11.446	1853069	0.31	1238651	0.39	1.50	Docosanoic acid, ethyl ester
30	11.750	624705	0.11	229958	0.07	2.72	Octanoic acid, octyl ester
31	11.836	3507171	0.59	2371022	0.74	1.48	9-Octadecen-1-ol, (Z)-
32	11.959	4336606	0.73	4164016	1.30	1.04	3-Eicosene, (E)-
33	12.105	50795107	8.55	37742314	11.81	1.35	Phytol
34	12.252	34685449	5.84	13400199	4.19	2.59	9,12-Octadecadienoic acid (Z,Z)-
35	12.288	33880284	5.70	19284488	6.03	1.76	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-
36	12.368	10469480	1.76	7031181	2.20	1.49	Octadecanoic acid
37	12.479	1357921	0.23	595893	0.19	2.28	trans-2-[2'-(2"-Methyl-1"-propenyl)cyclopropy
38	12.856	1165982	0.20	669137	0.21	1.74	Pentalene, octahydro-2-[(2-octyl)decyl]-
39	12.942	1099452	0.18	502547	0.16	2.19	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-
40	12.989	3160070	0.53	1891173	0.59	1.67	1-Heneicosanol
41	13.135	1476581	0.25	863802	0.27	1.71	2-Dodecylcyclobutanone
42	13.166	3057413	0.51	1934472	0.61	1.58	9-Octadecenal, (Z)-
43	13.269	1702097	0.29	774700	0.24	2.20	Eicosane, 2-cyclohexyl-
44	13.342	7945894	1.34	4494695	1.41	1.77	Eicosanoic acid
45	13.806	721201	0.12	531329	0.17	1.36	Hexadecanal, 2-methyl-
46	13.927	1164715	0.20	926803	0.29	1.26	Hexadecanoic acid, 2-hydroxy-1-(hydroxymet
47	13.971	2594294	0.44	1153145	0.36	2.25	Bicyclo[6.1.0]nonane, 9-(1-methylethylidene)-
48	14.042	5686004	0.96	3992765	1.25	1.42	Hexadecanoic acid, 2-hydroxy-1-(hydroxymet
49	14.128	740700	0.12	401946	0.13	1.84	Methanol, [6,8,9-trimethyl-4-(1-propenyl)-3-o
50	14.252	653551	0.11	609356	0.19	1.07	Octadecanoic acid
51	14.425	3375540	0.57	1145575	0.36	2.95	Preg-4-en-3-one, 17.alpha.-hydroxy-17.beta.-c
52	14.710	1662485	0.28	851450	0.27	1.95	9,12-Octadecadienoic acid (Z,Z)-, 2,3-dihydro
53	14.749	1458435	0.25	783204	0.25	1.86	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-
54	14.827	5700871	0.96	3387304	1.06	1.68	9,12-Octadecadienoic acid (Z,Z)-, 2-hydroxy-
55	14.863	11352654	1.91	4861279	1.52	2.34	1,6,10,14-Hexadecatetraen-3-ol, 3,7,11,15-tetr
56	14.928	1586442	0.27	654854	0.20	2.42	Octadecanoic acid, 2,3-dihydroxypropyl ester
57	15.185	4559859	0.77	3582225	1.12	1.27	9-Octadecenamide, (Z)-
58	15.303	16365666	2.75	14266379	4.46	1.15	Squalene
59	15.394	915964	0.15	636693	0.20	1.44	(E,E,E)-3,7,11,15-Tetramethylhexadeca-1,3,6,
60	15.450	696041	0.12	345366	0.11	2.02	Hexadecane, 1-bromo-
61	15.532	694530	0.12	355532	0.11	1.95	Cyclohexane, 1,2,3,5-tetraisopropyl-
62	15.811	982484	0.17	376284	0.12	2.61	3H-Naphtho[2,3-b]furan-2-one, 4-hydroxy-4a,
63	15.941	4977897	0.84	4002493	1.25	1.24	17-(1,5-Dimethyl-hex-2-enyl)-10,13-dimethyl-
64	16.033	626045	0.11	480377	0.15	1.30	(E,E,E)-3,7,11,15-Tetramethylhexadeca-1,3,6,
65	16.139	1058472	0.18	560383	0.18	1.89	2-(8-Chloro-3,7-dimethyl-octa-2,6-dienyloxy)t
66	16.300	3652917	0.61	2220671	0.69	1.64	.gamma.-Tocopherol
67	16.479	618559	0.10	282536	0.09	2.19	n-Tetracosanol-1
68	16.664	5833967	0.98	3736334	1.17	1.56	Vitamin E
69	16.928	5591153	0.94	2830574	0.89	1.98	Lycoxanthin
70	16.988	1462144	0.25	581338	0.18	2.52	1,6,10,14,18,22-Tetracosahexaen-3-ol, 2,6,10,
71	17.102	2934923	0.49	1663703	0.52	1.76	15,17,19,21-Hexatriacontatetrayne
72	17.316	565776	0.10	302488	0.09	1.87	Ergost-5-en-3-ol, (3.beta.)-
73	17.462	14031956	2.36	7006273	2.19	2.00	Stigmasterol
74	17.600	17230416	2.90	8056241	2.52	2.14	Chondrillasterol
75	17.654	2055597	0.35	1088500	0.34	1.89	Stigmasta-4,7,22-trien-3.alpha.-ol
76	17.848	39690421	6.68	14992554	4.69	2.65	Chondrillasterol
77	17.989	1255007	0.21	640919	0.20	1.96	Cholest-7-en-3-ol, 2,2-dimethyl-, (3.beta.,5.alp
78	18.176	2271686	0.38	900990	0.28	2.52	5,8,11-Eicosatriynoic acid, methyl ester
79	18.218	1620602	0.27	755810	0.24	2.14	4,4,6a,6b,8a,11,11,14b-Octamethyl-1,4,4a,5,6,
80	18.455	29983075	5.05	7956485	2.49	3.77	10,12,14-Nonacosatriynoic acid
81	18.562	1800031	0.30	677690	0.21	2.66	.alpha.-Amyrin
82	19.228	12950173	2.18	5077346	1.59	2.55	Phytol, acetate
83	19.759	994329	0.17	349477	0.11	2.85	cis-13,16-Docasadienoic acid, tert-butyldimeth
84	21.015	4075163	0.69	885126	0.28	4.60	E,E,Z-1,3,12-Nonadecatriene-5,14-diol
85	21.215	4792844	0.81	975675	0.31	4.91	Phytol, acetate
		594304312	100.00	319546837	100.00		

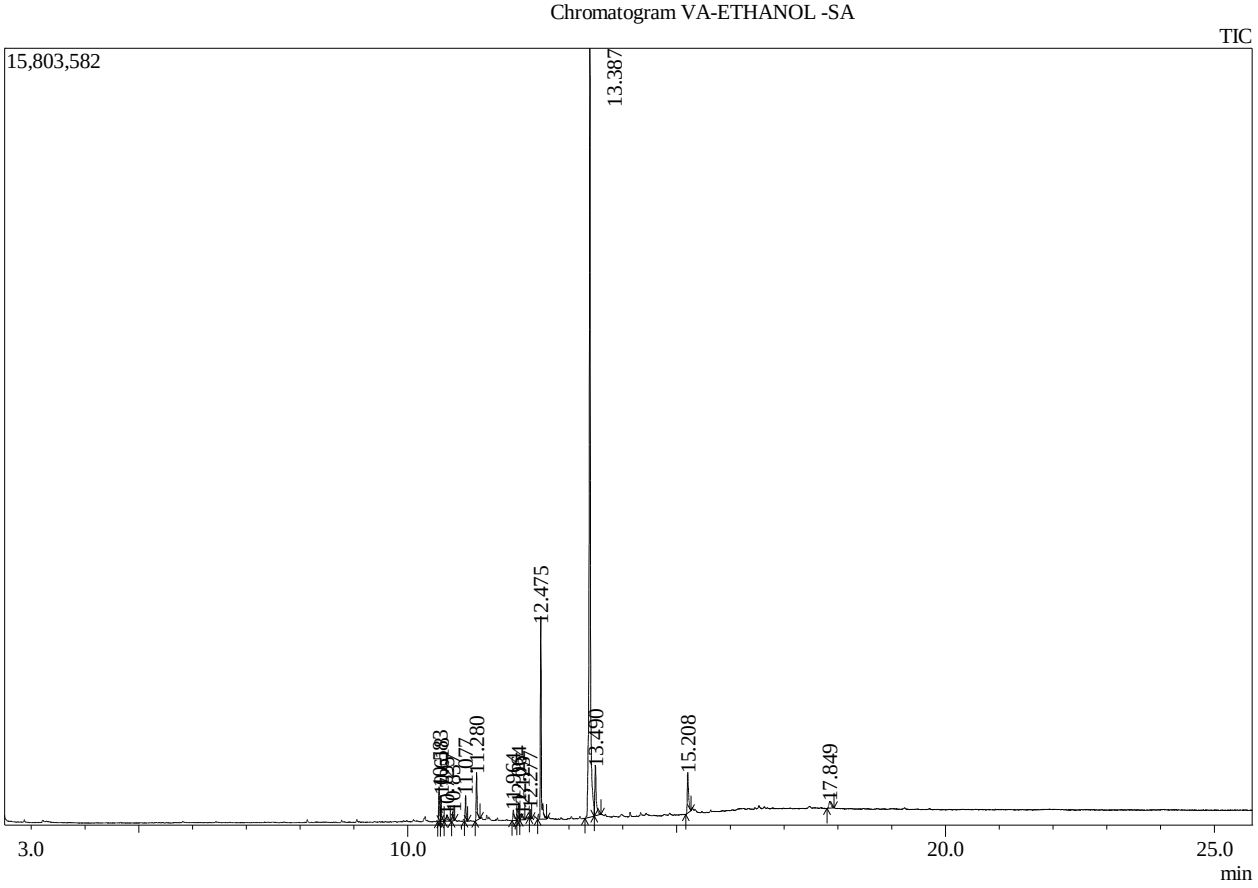
GC-MS ANALYSIS OF *VERNONIA AMYGDALINA* (ETHANOL)- SOUTH AFRICA

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/08/02 6:16:20 AM
Sample Type	: Unknown
Sample Name	: VA-ETHANOL(SA)
Sample ID	: Akwasi
Vial #	: 88
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 07 03.qgt

Chromatogram VA-ETHANOL -SA



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	10.583	780767	2.02	700188	2.79	1.12	Phytol, acetate
2	10.618	656846	1.70	528857	2.11	1.24	2-Pentadecanone, 6,10,14-trimethyl-
3	10.725	200459	0.52	115712	0.46	1.73	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
4	10.837	208119	0.54	172787	0.69	1.20	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
5	11.077	540213	1.40	500707	1.99	1.08	1-Hexacosene
6	11.280	1198329	3.10	914798	3.64	1.31	Pentadecanoic acid
7	11.964	348535	0.90	206901	0.82	1.68	Pentanoic acid, 4-hexadecyl ester
8	12.064	401512	1.04	338788	1.35	1.19	.gamma.-Dodecalactone
9	12.123	309138	0.80	107949	0.43	2.86	Phytol
10	12.277	166382	0.43	177281	0.71	0.94	Cyclopentanone, 2-(5-oxohexyl)-
11	12.475	4914997	12.70	3952207	15.74	1.24	Hexadecanamide
12	13.387	25327386	65.45	15428897	61.45	1.64	9-Octadecenamide, (Z)-
13	13.490	1804619	4.66	1008121	4.02	1.79	Octadecanamide
14	15.208	1416745	3.66	816913	3.25	1.73	13-Docosenamide, (Z)-
15	17.849	420698	1.09	138471	0.55	3.04	Ergosta-7,22-dien-3-ol, (3.beta.,5.alpha.,22E)-
		38694745	100.00	25108577	100.00		

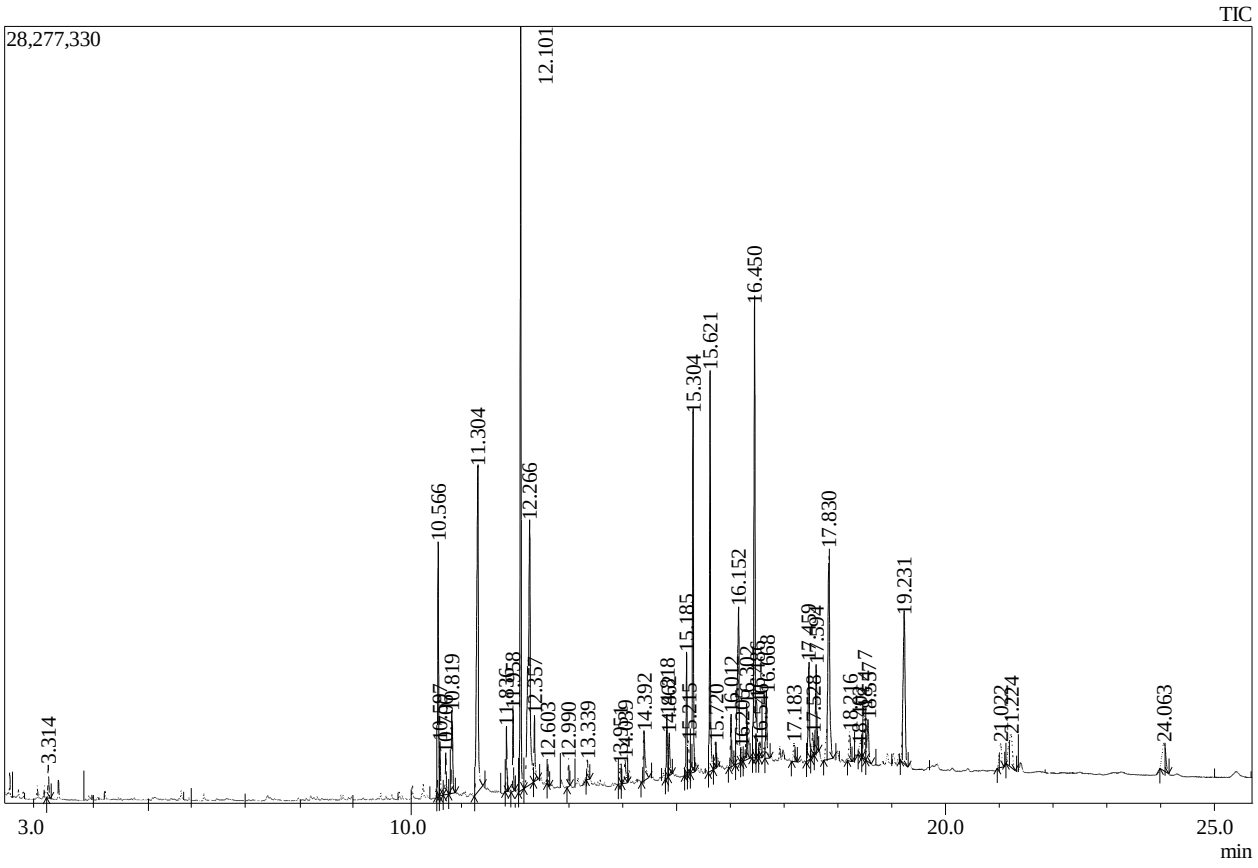
GC-MS ANALYSIS OF *VERNONIA AMYGDALINA* (ETHYL ACETATE)- Ghana

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 6:03:18 PM
Sample Type	: Unknown
Sample Name	: VA-EtoAc-Ghana
Sample ID	: Akwasi
Vial #	: 36
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram VA-EtoAc-Ghana



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	3.314	1395966	0.46	1168969	0.66	1.19	o-Xylene
2	10.566	10202701	3.35	9107503	5.10	1.12	Phytol, acetate
3	10.597	2805994	0.92	1939413	1.09	1.45	2-Pentadecanone, 6,10,14-trimethyl-
4	10.706	2324582	0.76	1513050	0.85	1.54	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
5	10.819	5004996	1.64	3004419	1.68	1.67	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
6	11.304	26581750	8.74	11877628	6.66	2.24	Pentadecanoic acid
7	11.836	2436680	0.80	2333896	1.31	1.04	9-Octadecen-1-ol, (Z)-
8	11.958	3125913	1.03	2939995	1.65	1.06	1-Heneicosanol
9	12.101	32510057	10.68	27471990	15.39	1.18	Phytol
10	12.266	27054865	8.89	9555225	5.35	2.83	cis,cis,cis-7,10,13-Hexadecatrienal
11	12.357	3429463	1.13	2312170	1.30	1.48	Octadecanoic acid
12	12.603	855478	0.28	908350	0.51	0.94	Phytol, acetate
13	12.990	1064516	0.35	987074	0.55	1.08	n-Nonadecanol-1
14	13.339	1280035	0.42	746406	0.42	1.71	Eicosanoic acid
15	13.951	1481370	0.49	731792	0.41	2.02	8-Methylenecyclooctene-3,4-diol
16	14.039	1775818	0.58	911975	0.51	1.95	Hexadecanoic acid, 2-hydroxy-1-(hydroxymet
17	14.392	4656246	1.53	1821377	1.02	2.56	Preg-4-en-3-one, 17.alpha.-hydroxy-17.beta.-c
18	14.818	3485012	1.15	2295433	1.29	1.52	2-methylhexacosane
19	14.862	2615073	0.86	1562671	0.88	1.67	(.+/-.)-Lavandulol, chlorodifluoroacetate
20	15.185	5257807	1.73	4411565	2.47	1.19	9-Octadecenamide, (Z)-
21	15.215	2037814	0.67	1226985	0.69	1.66	2-methyltetracosane
22	15.304	16415047	5.39	13171407	7.38	1.25	Squalene
23	15.621	17076404	5.61	14504064	8.13	1.18	Tetratetracontane
24	15.720	1102762	0.36	952222	0.53	1.16	Oxirane, 2,2-dimethyl-3-(3,7,12,16,20-pentam
25	16.012	3334148	1.10	1922382	1.08	1.73	Tetratetracontane
26	16.152	13778123	4.53	5656884	3.17	2.44	2-(8-Chloro-3,7-dimethyl-octa-2,6-dienyloxy)t

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	16.205	1039026	0.34	531160	0.30	1.96	Oxirane, heptadecyl-
28	16.302	3805492	1.25	2029463	1.14	1.88	.gamma.-Tocopherol
29	16.450	24217103	7.96	16560056	9.28	1.46	Tetratetracontane
30	16.486	3086421	1.01	2113146	1.18	1.46	n-Tetracosanol-1
31	16.546	1244223	0.41	571065	0.32	2.18	9-Octadecanone
32	16.668	3890718	1.28	2413873	1.35	1.61	Vitamin E
33	17.183	1582954	0.52	674648	0.38	2.35	Ergosterol
34	17.459	6238814	2.05	3482536	1.95	1.79	Stigmasterol
35	17.528	1601840	0.53	862270	0.48	1.86	Eicosyl heptafluorobutyrate
36	17.594	5947134	1.95	3177992	1.78	1.87	Chondrillasterol
37	17.830	19243189	6.32	7600899	4.26	2.53	Chondrillasterol
38	18.216	1995717	0.66	1064344	0.60	1.88	.alpha.-Amyrin
39	18.402	1922817	0.63	424545	0.24	4.53	1-Phenanthrenemethanol, 1,2,3,4,4a,9,10,10a-
40	18.477	4160282	1.37	1807473	1.01	2.30	dl-.alpha.-Tocopherol
41	18.557	3463919	1.14	1497140	0.84	2.31	.alpha.-Amyrin
42	19.231	14399026	4.73	5490131	3.08	2.62	Phytol, acetate
43	21.022	2732679	0.90	893480	0.50	3.06	1,1':3',1"-Tercyclopentane, 2'-dodecyl-
44	21.224	5883634	1.93	1216640	0.68	4.84	Phytol, acetate
45	24.063	4729179	1.55	1016412	0.57	4.65	Phytol, acetate
		304272787	100.00	178462118	100.00		

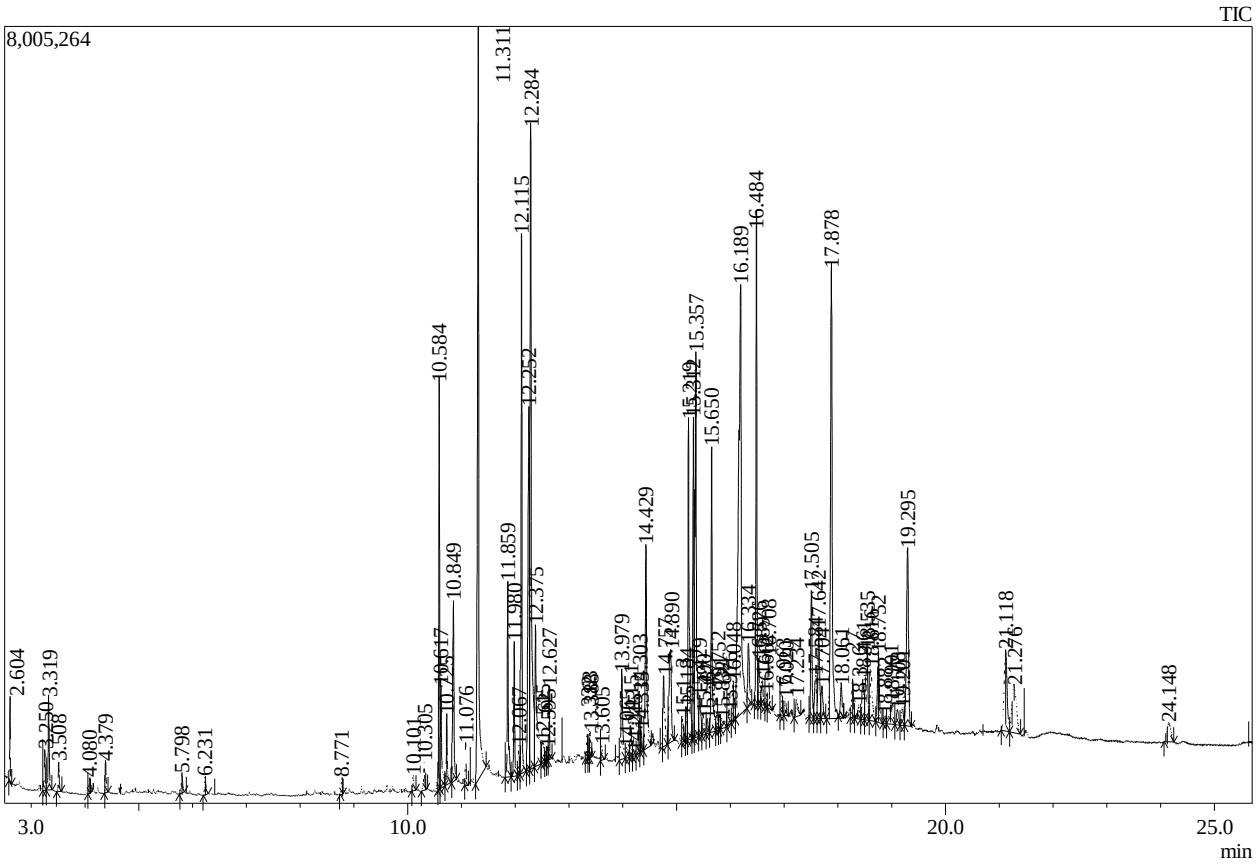
GC-MS ANALYSIS OF *VERNONIA AMYGDALINA* (ETHYL ACETATE)- South Africa

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 1:47:54 PM
Sample Type	: Unknown
Sample Name	: VA-EtoAc-SA
Sample ID	: Akwasi
Vial #	: 29
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram VA-EtoAc-SA



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	2.604	663071	0.38	836994	0.91	0.79	Toluene
2	3.250	525007	0.30	402784	0.44	1.30	Ethylbenzene
3	3.319	1411084	0.80	961925	1.04	1.47	Benzene, 1,3-dimethyl-
4	3.508	387362	0.22	303272	0.33	1.28	Benzene, 1,3-dimethyl-
5	4.080	199178	0.11	160828	0.17	1.24	Benzene, 1-ethyl-3-methyl-
6	4.379	451332	0.26	327879	0.36	1.38	Benzene, 1-ethyl-3-methyl-
7	5.798	401823	0.23	207608	0.22	1.94	3,4-Dimethyl-2-pentanone
8	6.231	369656	0.21	189367	0.21	1.95	1-Heptanol, 2-propyl-
9	8.771	219862	0.13	172398	0.19	1.28	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4
10	10.101	254783	0.15	164484	0.18	1.55	Tetradecanoic acid
11	10.305	436586	0.25	288019	0.31	1.52	Acetic acid, 2-(2,2,6-trimethyl-7-oxa-bicyclo[4
12	10.584	4552682	2.59	4218883	4.57	1.08	Phytol, acetate
13	10.617	1707808	0.97	1032209	1.12	1.65	2-Pentadecanone, 6,10,14-trimethyl-
14	10.725	1076617	0.61	730041	0.79	1.47	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
15	10.849	3117507	1.77	1845836	2.00	1.69	3-Eicosene, (E)-
16	11.076	462562	0.26	423681	0.46	1.09	1-Hexacosene
17	11.311	13442458	7.65	7759172	8.41	1.73	Pentadecanoic acid
18	11.859	2663246	1.52	1984259	2.15	1.34	9-Octadecen-1-ol, (Z)-
19	11.980	1743170	0.99	1344412	1.46	1.30	1-Heneicosanol
20	12.067	314996	0.18	306026	0.33	1.03	.gamma.-Dodecalactone
21	12.115	6036154	3.44	5560245	6.02	1.09	Phytol
22	12.252	7292956	4.15	3712259	4.02	1.96	9,12-Octadecadienoic acid (Z,Z)-
23	12.284	12816635	7.30	6636583	7.19	1.93	cis,cis,cis-7,10,13-Hexadecatrienal
24	12.375	3836348	2.18	1458934	1.58	2.63	Octadecanoic acid
25	12.515	729265	0.42	216406	0.23	3.37	2-Dodecen-1-yl(-)succinic anhydride
26	12.562	178469	0.10	105169	0.11	1.70	Tetracosane, 1-bromo-

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	12.595	180837	0.10	137568	0.15	1.31	3,7,11-Trimethyldodecylacetate
28	12.627	879866	0.50	741292	0.80	1.19	Phytol, acetate
29	13.332	453279	0.26	242484	0.26	1.87	4,8,12,16-Tetramethylheptadecan-4-olide
30	13.363	404773	0.23	298126	0.32	1.36	Eicosanoic acid
31	13.388	422459	0.24	269393	0.29	1.57	9-Octadecenamide, (Z)-
32	13.605	234091	0.13	157406	0.17	1.49	3,4-Xylyl 3,5-di-tert-butylbenzoate
33	13.979	2068751	1.18	924146	1.00	2.24	Preg-4-en-3-one, 17.alpha.-hydroxy-17.beta.-c
34	14.065	351913	0.20	109140	0.12	3.22	Glycerol 1-palmitate
35	14.151	538401	0.31	354256	0.38	1.52	Diisooctyl phthalate
36	14.215	173033	0.10	49885	0.05	3.47	12,15-Octadecadiynoic acid, methyl ester
37	14.303	1051530	0.60	631070	0.68	1.67	Cyclohexane, 1,2-diethenyl-4-(1-methylethylid
38	14.335	343886	0.20	230471	0.25	1.49	1,1,3,3-Tetramethyl-1,3-disilacyclopentane
39	14.429	4386064	2.50	2104072	2.28	2.08	Preg-4-en-3-one, 17.alpha.-hydroxy-17.beta.-c
40	14.757	1478114	0.84	726633	0.79	2.03	6,10-Dodecadien-1-yn-3-ol, 3,7,11-trimethyl-
41	14.890	2919645	1.66	945038	1.02	3.09	2,6-Octadiene, 4,5-dimethyl-
42	15.113	401904	0.23	259398	0.28	1.55	Cholesta-8,24-dien-3-ol, 4-methyl-, (3.beta.,4.
43	15.184	462545	0.26	352621	0.38	1.31	4-(2,4,4-Trimethyl-cyclohexa-1,5-dienyl)-but-
44	15.219	5440903	3.10	3256640	3.53	1.67	13-Docosenamide, (Z)-
45	15.312	4609909	2.62	3249648	3.52	1.42	Isolongifolene, 9,10-dehydro-
46	15.357	7860416	4.47	3856724	4.18	2.04	(3,7-Dimethyl-octa-2,6-dienyl)-benzene
47	15.429	955802	0.54	418446	0.45	2.28	Cholesta-8,24-dien-3-ol, 4-methyl-, (3.beta.,4.
48	15.490	584384	0.33	239473	0.26	2.44	erythro-7,8-Bromochlorodisparlure
49	15.567	331105	0.19	154038	0.17	2.15	Cyclohexane, 1,2,3,5-tetraisopropyl-
50	15.650	4119801	2.35	2892186	3.13	1.42	2-methyloctacosane
51	15.752	593536	0.34	423879	0.46	1.40	2,6,10,14-Hexadecatetraen-1-ol, 3,7,11,15-tetr
52	15.791	273871	0.16	163772	0.18	1.67	Squalene
53	15.832	374853	0.21	107832	0.12	3.48	.beta.-D-Mannofuranoside, farnesyl-
54	15.976	286614	0.16	149465	0.16	1.92	Card-20(22)-enolide, 3,14-dihydroxy-, (3.beta
55	16.048	971643	0.55	412961	0.45	2.35	2-methyloctacosane
56	16.189	16385833	9.33	4392905	4.76	3.73	2-(8-Chloro-3,7-dimethyl-octa-2,6-dienyloxy)t
57	16.334	1428738	0.81	651179	0.71	2.19	Androst-5,7-dien-3-ol-17-one, acetate
58	16.484	6243268	3.55	4988831	5.41	1.25	Tetratetracontane
59	16.526	831698	0.47	468379	0.51	1.78	n-Tetracosanol-1
60	16.589	561063	0.32	305153	0.33	1.84	Cholesta-4,6-dien-3-ol, (3.beta.)-
61	16.662	245289	0.14	165515	0.18	1.48	22,23-Dibromostigmasterol acetate
62	16.708	804300	0.46	537766	0.58	1.50	Vitamin E
63	16.963	336309	0.19	193955	0.21	1.73	2-methylhexacosane
64	17.020	361352	0.21	164810	0.18	2.19	Hexacosyl acetate
65	17.234	542435	0.31	210847	0.23	2.57	Ethyl iso-allocholate
66	17.505	2342562	1.33	1310550	1.42	1.79	Stigmasterol
67	17.584	1054988	0.60	433573	0.47	2.43	1-Heptacosanol
68	17.642	2020591	1.15	925235	1.00	2.18	Chondrillasterol
69	17.704	840406	0.48	333399	0.36	2.52	Ergosta-8(14),15,22-trien-3-ol, (3.beta.,5.alph
70	17.878	10643116	6.06	4670414	5.06	2.28	Chondrillasterol
71	18.061	712537	0.41	342569	0.37	2.08	7,22-Ergostadienone
72	18.271	884594	0.50	401694	0.44	2.20	4,4,6a,6b,8a,11,11,14b-Octamethyl-1,4,4a,5,6,
73	18.386	339028	0.19	147064	0.16	2.31	4,4,6a,6b,8a,11,11,14b-Octamethyl-1,4,4a,5,6,
74	18.461	1052740	0.60	408762	0.44	2.58	10,12,14-Nonacosatriynoic acid
75	18.535	2095420	1.19	724282	0.78	2.89	dl-.alpha.-Tocopherol
76	18.616	1519009	0.86	550744	0.60	2.76	.alpha.-Amyrin
77	18.752	1842835	1.05	724835	0.79	2.54	2-Azapentane-1,5-dione, 4-methyl-1,5-diphen
78	18.882	206360	0.12	90362	0.10	2.28	Tricyclo[20.8.0.0(7,16)]triacontane, 1(22),7(1
79	18.991	938001	0.53	228313	0.25	4.11	Docosyl trifluoroacetate
80	19.108	488737	0.28	155431	0.17	3.14	2-Nonadecanone
81	19.201	550018	0.31	189978	0.21	2.90	Lanosterol
82	19.295	4804113	2.73	1834463	1.99	2.62	Phytol, acetate
83	21.118	3247083	1.85	842912	0.91	3.85	E,E,Z-1,3,12-Nonadecatriene-5,14-diol
84	21.276	2900606	1.65	491954	0.53	5.90	Tricyclo[20.8.0.0(7,16)]triacontane, 1(22),7(1
85	24.148	990772	0.56	201664	0.22	4.91	Phytol, acetate
		175658346	100.00	92295274	100.00		

GC-MS ANALYSIS OF *PERSEA AMERICANA*

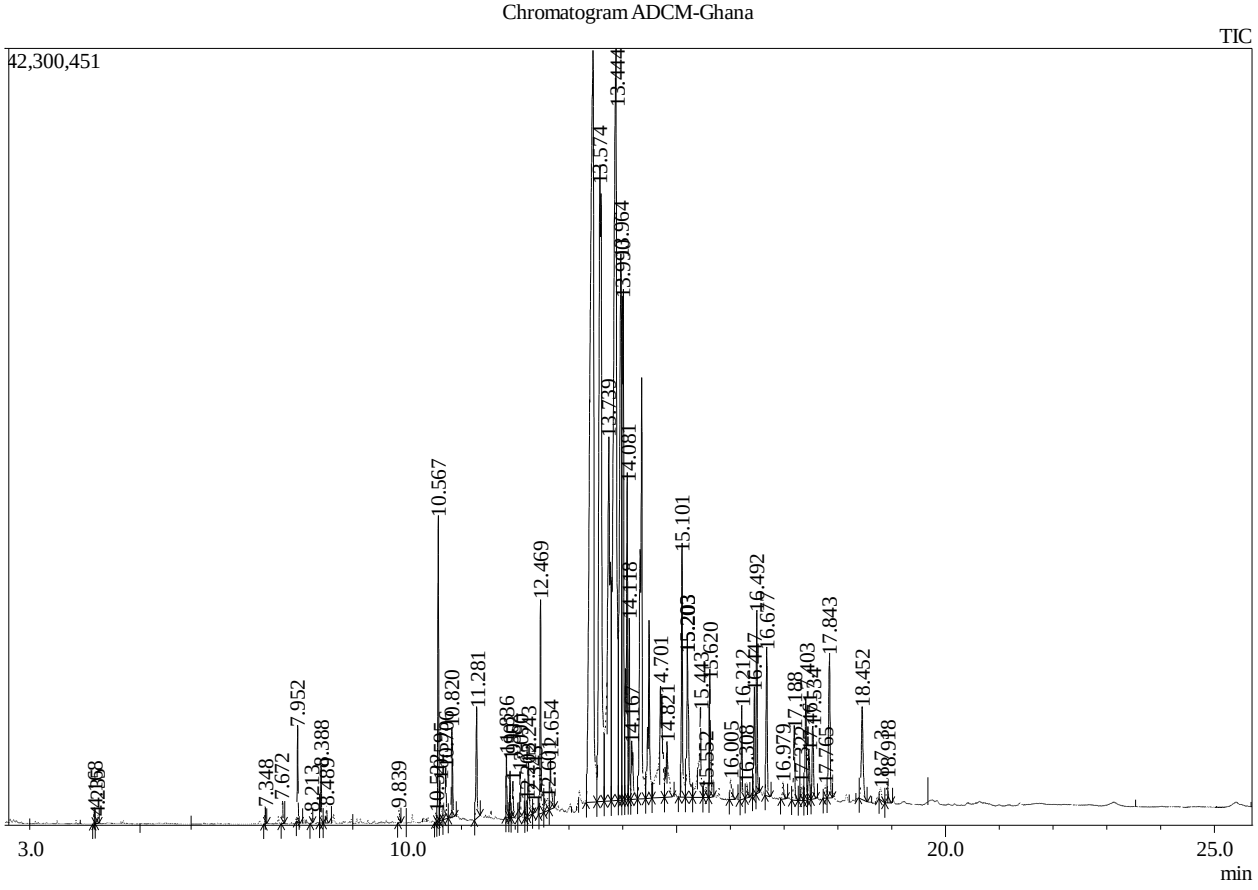
(DCM)- Ghana

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 6:39:10 PM
Sample Type	: Unknown
Sample Name	: PA-DCM-Ghana
Sample ID	: Akwasi
Vial #	: 37
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram ADCM-Ghana



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	4.168	770666	0.07	872660	0.20	0.88	Bicyclo[3.1.0]hexane, 4-methylene-1-(1-meth
2	4.235	417025	0.04	412259	0.10	1.01	.beta.-Pinene
3	7.348	945353	0.08	912788	0.21	1.04	.alpha.-Cubebene
4	7.672	1200101	0.10	1237181	0.29	0.97	.beta.-copaene
5	7.952	5267034	0.46	5189459	1.21	1.01	Caryophyllene
6	8.213	555096	0.05	568538	0.13	0.98	Humulene
7	8.388	3298699	0.29	3075676	0.71	1.07	1,6-Cyclodecadiene, 1-methyl-5-methylene-8-
8	8.489	1211256	0.11	909308	0.21	1.33	Cyclohexane, 1-ethenyl-1-methyl-2-(1-methyle
9	9.839	842291	0.07	774241	0.18	1.09	Heptadecanal
10	10.522	682766	0.06	513806	0.12	1.33	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
11	10.567	18429724	1.60	16611007	3.86	1.11	Phytol, acetate
12	10.595	3336649	0.29	2188942	0.51	1.52	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
13	10.706	3920393	0.34	2766663	0.64	1.42	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
14	10.820	8015438	0.70	4892914	1.14	1.64	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
15	11.281	8623919	0.75	6026836	1.40	1.43	Pentadecanoic acid
16	11.836	4117748	0.36	3417784	0.79	1.20	9-Octadecen-1-ol, (Z)-
17	11.902	2367970	0.21	2531373	0.59	0.94	9,17-Octadecadienal, (Z)-
18	11.955	2180753	0.19	1978266	0.46	1.10	n-Nonadecanol-1
19	12.090	2574786	0.22	2401052	0.56	1.07	Phytol
20	12.205	1378778	0.12	880467	0.20	1.57	11,14-Eicosadienoic acid, methyl ester
21	12.243	5489383	0.48	2837640	0.66	1.93	7-Tetradecenal, (Z)-
22	12.345	596132	0.05	514850	0.12	1.16	Octadecanoic acid
23	12.469	13978145	1.22	11502031	2.67	1.22	Di(1-decynyl)mercury
24	12.601	3062869	0.27	671103	0.16	4.56	6,11-Undecadiene, 1-acetoxy-3,7-dimethyl-
25	12.654	4508072	0.39	2763558	0.64	1.63	cis-9-Hexadecenoic acid, trimethylsilyl ester
26	13.444	196275969	17.06	40389895	9.39	4.86	2-Hydroxy-(Z)9-pentadecenyl propanoate

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	13.574	126916833	11.03	33598559	7.81	3.78	E-11-Methyl-12-tetradecen-1-ol acetate
28	13.739	79170965	6.88	19583207	4.55	4.04	11-Dodecyn-1-ol acetate
29	13.869	170552886	14.83	40209830	9.35	4.24	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
30	13.964	80852358	7.03	29570142	6.87	2.73	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
31	13.990	60325980	5.24	27194025	6.32	2.22	2,3,4-Trimethyl-5-hexen-3-ol
32	14.081	31328133	2.72	15939774	3.71	1.97	9-Methyl-Z,Z-10,12-hexadecadien-1-ol acetat
33	14.118	11040188	0.96	8559790	1.99	1.29	3-Isopropyl-4-methyl-dec-1-en-4-ol
34	14.167	7251656	0.63	3053299	0.71	2.38	Estran-3-one, 17-(acetyloxy)-2-methyl-, (2.alp
35	14.351	53888786	4.69	22300797	5.18	2.42	Cyclopropanecarboxylic acid,pentadecyl ester
36	14.488	20571224	1.79	9029662	2.10	2.28	(R)-(-)-14-Methyl-8-hexadecyn-1-ol
37	14.701	28921441	2.51	5614918	1.31	5.15	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
38	14.821	9739537	0.85	2896122	0.67	3.36	Tetratetracontane
39	15.101	20386112	1.77	12881330	2.99	1.58	Ethanol, 2-(9,12-octadecadienyloxy)-, (Z,Z)-
40	15.203	18723150	1.63	8067740	1.88	2.32	13-Docosenamide, (Z)-
41	15.443	15445732	1.34	4695002	1.09	3.29	9-Methyl-10,12-hexadecadien-1-ol acetate
42	15.552	2306386	0.20	519297	0.12	4.44	11-Dodecyn-1-ol acetate
43	15.620	9253737	0.80	6559165	1.52	1.41	Tetratetracontane
44	16.005	2109558	0.18	1100922	0.26	1.92	2-methylhexacosane
45	16.212	6870102	0.60	4996778	1.16	1.37	Hexadecanal
46	16.308	1265109	0.11	753121	0.18	1.68	.gamma.-Tocopherol
47	16.447	8651426	0.75	5537120	1.29	1.56	Heneicosane
48	16.492	12950372	1.13	9805195	2.28	1.32	1-Heptacosanol
49	16.677	12202017	1.06	7897203	1.84	1.55	Vitamin E
50	16.979	1677639	0.15	924208	0.21	1.82	Heptacosyl acetate
51	17.188	6363941	0.55	3743499	0.87	1.70	Hexadecanal
52	17.322	1671984	0.15	885923	0.21	1.89	Ergost-5-en-3-ol, (3.beta.)-
53	17.403	9344753	0.81	5462196	1.27	1.71	16-Hentriacontanone
54	17.461	4896966	0.43	2643746	0.61	1.85	Stigmasterol
55	17.534	7465869	0.65	3915883	0.91	1.91	Docosyl trifluoroacetate
56	17.765	1540314	0.13	713960	0.17	2.16	Hexadecanal
57	17.843	15846102	1.38	7684765	1.79	2.06	.gamma.-Sitosterol
58	18.452	11867788	1.03	4995939	1.16	2.38	Hexadecanal
59	18.793	1779440	0.15	679027	0.16	2.62	17-Pentatriacontene
60	18.918	2980554	0.26	1356861	0.32	2.20	Heptacosyl trifluoroacetate
		1150206053	100.00	430209302	100.00		

GC-MS ANALYSIS OF *PERSEA AMERICANA*

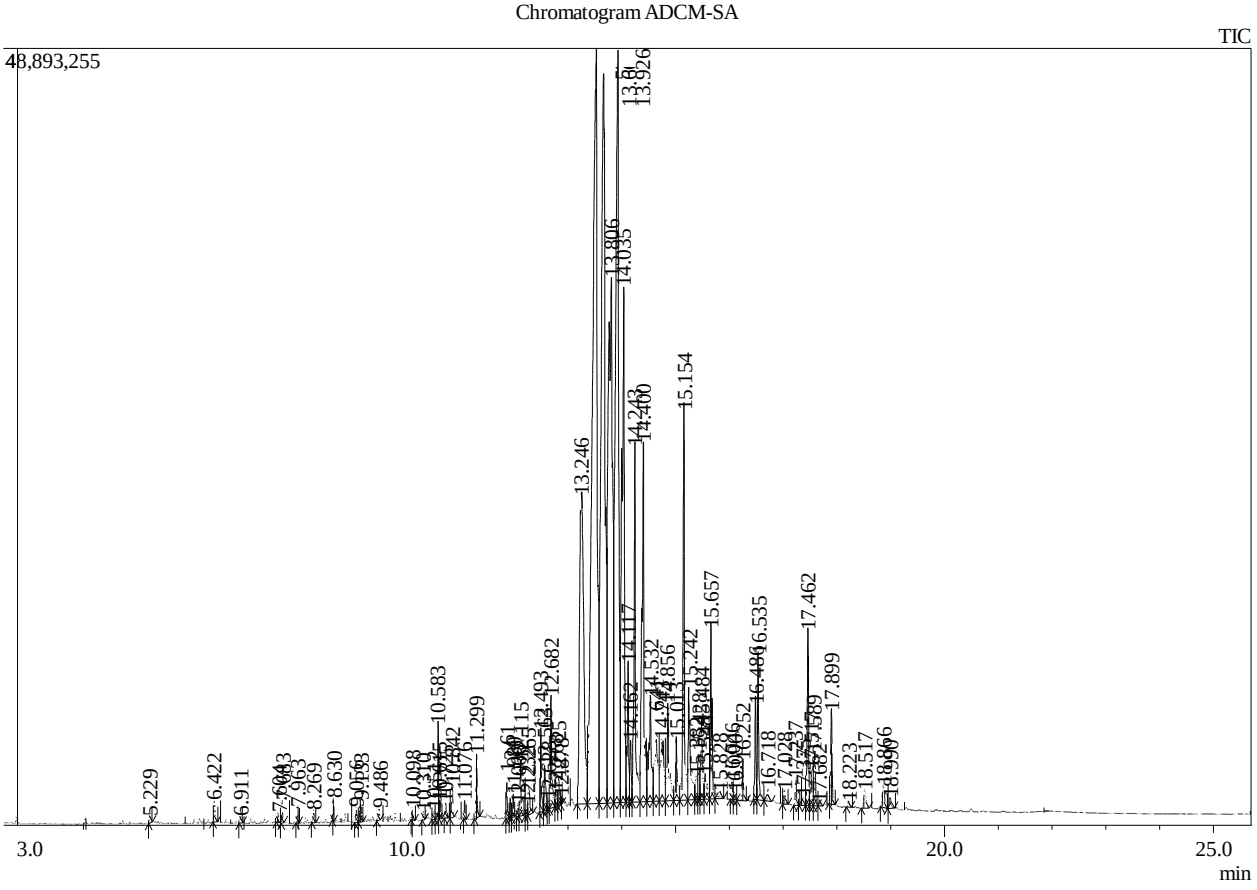
(DCM)- South Africa

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 3:01:00 PM
Sample Type	: Unknown
Sample Name	:PA-DCM-SA
Sample ID	: Akwasi
Vial #	: 31
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram ADCM-SA



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	5.229	773873	0.05	398152	0.09	1.94	1,2,3-Propanetriol, 1-acetate
2	6.422	2182739	0.13	1353919	0.29	1.61	1,2,3-Propanetriol, 1-acetate
3	6.911	726718	0.04	421781	0.09	1.72	4-Penten-2-one, 3-cyclohexyl-
4	7.604	582455	0.04	611048	0.13	0.95	.alfa.-Copaene
5	7.683	1363988	0.08	1379156	0.30	0.99	1H-Cyclopenta[1,3]cyclopropa[1,2]benzene, o
6	7.963	1030436	0.06	1030890	0.22	1.00	Caryophyllene
7	8.269	915439	0.06	761223	0.16	1.20	2-Butanone, 4-(2,6,6-trimethyl-2-cyclohexen-1
8	8.630	1217402	0.07	1385281	0.30	0.88	cubedol
9	9.056	1520070	0.09	935345	0.20	1.63	Bicyclo[7.2.0]undec-4-ene, 4,11,11-trimethyl-
10	9.133	2727655	0.17	1353225	0.29	2.02	Caryophyllene oxide
11	9.486	1059007	0.06	741525	0.16	1.43	Tetracyclo[6.3.2.0(2,5).0(1,8)]tridecan-9-ol, 4
12	10.098	1047332	0.06	767206	0.17	1.37	2-Piperidinone, N-[4-bromo-n-butyl]-
13	10.310	717871	0.04	549844	0.12	1.31	7-Oxabicyclo[4.1.0]heptane, 1-methyl-4-(2-m
14	10.512	1289102	0.08	642953	0.14	2.00	3-Buten-2-one, 4-(3-hydroxy-6,6-dimethyl-2-
15	10.583	6890088	0.42	6157687	1.33	1.12	Phytol, acetate
16	10.615	1703675	0.10	1178753	0.25	1.45	2-Pentadecanone, 6,10,14-trimethyl-
17	10.725	1187059	0.07	981208	0.21	1.21	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
18	10.842	3785484	0.23	2058485	0.44	1.84	Oxirane, hexadecyl-
19	11.076	1142564	0.07	1156664	0.25	0.99	1-Hexacosene
20	11.299	5650406	0.34	3956048	0.85	1.43	Pentadecanoic acid
21	11.861	2880196	0.18	2206950	0.48	1.31	9-Octadecen-1-ol, (Z)-
22	11.924	1321133	0.08	1222998	0.26	1.08	9,17-Octadecadienal, (Z)-
23	11.980	1991909	0.12	1395022	0.30	1.43	1-Heneicosanol
24	12.066	1045151	0.06	796477	0.17	1.31	2-Ethylnon-1-en-3-ol
25	12.115	3895762	0.24	3459876	0.75	1.13	Phytol
26	12.232	1002424	0.06	695683	0.15	1.44	11,14-Eicosadienoic acid, methyl ester

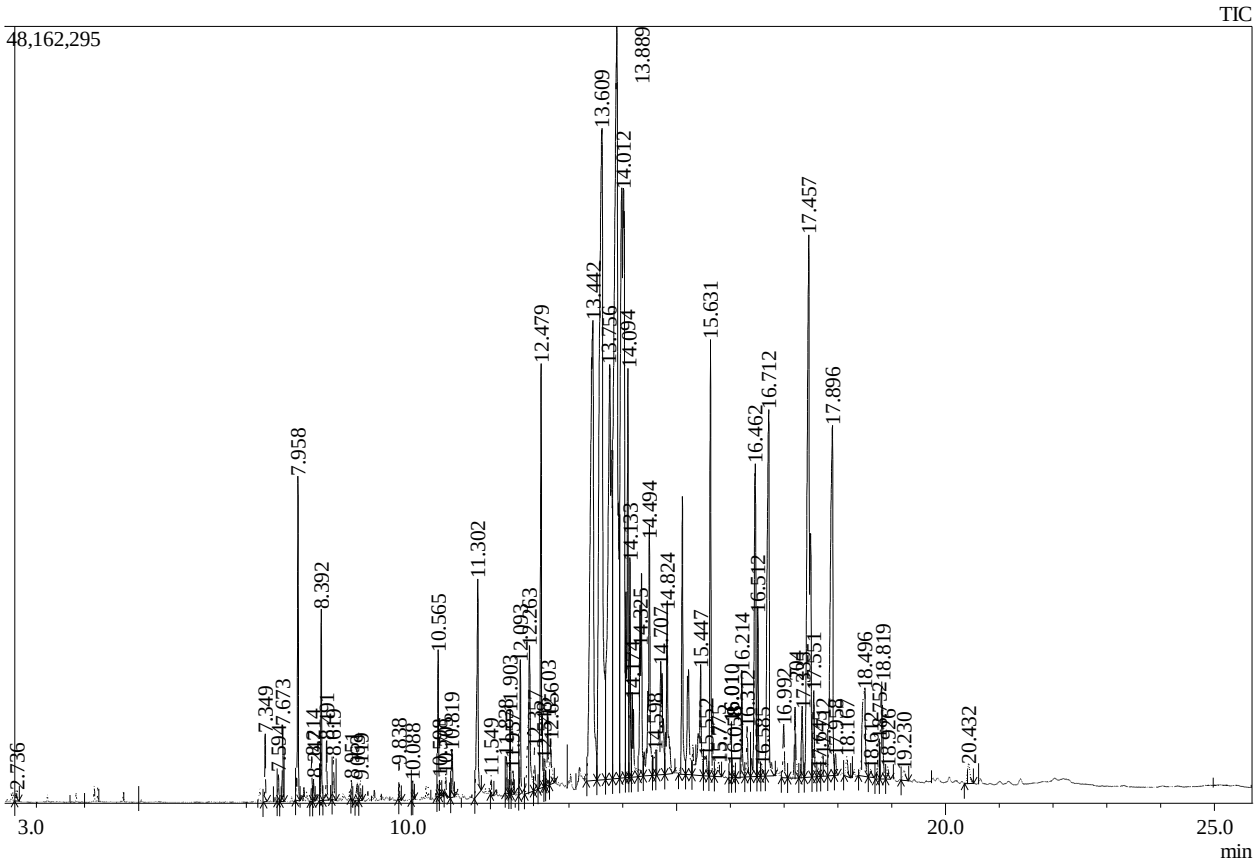
Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	12.265	4314736	0.26	1790340	0.39	2.41	7-Tetradecenal, (Z)-
28	12.493	6585436	0.40	5170299	1.12	1.27	Di(1-decynyl)mercury
29	12.563	5693485	0.35	2777729	0.60	2.05	Silane, dimethyl(3-methylbut-3-enyloxy)isobu
30	12.628	988887	0.06	641857	0.14	1.54	Phytol, acetate
31	12.682	11559995	0.70	7098513	1.53	1.63	cis-9-Hexadecenoic acid, trimethylsilyl ester
32	12.768	1785436	0.11	929127	0.20	1.92	cis, 6-Octadecenoic acid, trimethylsilyl ester
33	12.825	2830063	0.17	1701487	0.37	1.66	Decalin-8a-ol-7-one, 4a,8-dimethyl-2-[2-(t-but
34	12.878	670472	0.04	513432	0.11	1.31	5-Hexadecanol
35	13.246	105628941	6.43	19550271	4.22	5.40	3-Methyl-2-(2-methylene-cyclohexyl)-butan-2
36	13.525	295671007	18.01	47343637	10.21	6.25	2-Hydroxy-(Z)9-pentadecenyl propanoate
37	13.661	243787851	14.85	45868262	9.89	5.31	E-11-Methyl-12-tetradecen-1-ol acetate
38	13.806	191108228	11.64	32752968	7.07	5.83	11-Dodecyn-1-ol acetate
39	13.926	200924034	12.24	46889365	10.11	4.29	7-Octen-3-ol, 2,3,6-trimethyl-
40	14.035	108113382	6.58	31649424	6.83	3.42	7-Octen-3-ol, 2,3,6-trimethyl-
41	14.117	17434174	1.06	8377397	1.81	2.08	9-Methyl-Z,Z-10,12-hexadecadien-1-ol acetat
42	14.162	6767299	0.41	3861772	0.83	1.75	3-Isopropyl-4-methyl-dec-1-en-4-ol
43	14.243	44161367	2.69	21190189	4.57	2.08	2-Nonadecanone, O-methyloxime
44	14.400	60395230	3.68	22270244	4.80	2.71	Cyclopropanecarboxylic acid,pentadecyl ester
45	14.532	22755111	1.39	6313600	1.36	3.60	9-Eicosyne
46	14.641	17730158	1.08	4151768	0.90	4.27	Acetic acid, 3-ethylpent-3-yl ester
47	14.743	17109596	1.04	3838085	0.83	4.46	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
48	14.856	15457829	0.94	5981344	1.29	2.58	Tetratetracontane
49	15.013	9099973	0.55	3836630	0.83	2.37	3-Methyl-2-(2-methylene-cyclohexyl)-butan-2
50	15.154	46882820	2.86	24347572	5.25	1.93	Ethanol, 2-(9,12-octadecadienyloxy)-, (Z,Z)-
51	15.242	19085367	1.16	6401905	1.38	2.98	13-Docosenamide, (Z)-
52	15.382	3891986	0.24	1496732	0.32	2.60	3-Methyl-2-(2-methylene-cyclohexyl)-butan-2
53	15.428	5029232	0.31	2789197	0.60	1.80	Hexadecanal
54	15.484	11086084	0.68	4581011	0.99	2.42	9-Methyl-10,12-hexadecadien-1-ol acetate
55	15.548	4718670	0.29	1562546	0.34	3.02	Z-5,17-Octadecadien-1-ol acetate
56	15.657	20068351	1.22	10344994	2.23	1.94	Tetratetracontane
57	15.828	1753176	0.11	467053	0.10	3.75	Hexadecanal
58	16.046	1870984	0.11	1146290	0.25	1.63	Tetratetracontane
59	16.090	883631	0.05	490289	0.11	1.80	Heptacosyl acetate
60	16.252	3537671	0.22	2377165	0.51	1.49	Hexadecanal
61	16.486	9125132	0.56	6170634	1.33	1.48	Tetratetracontane
62	16.535	14711609	0.90	9187339	1.98	1.60	1-Heptacosanol
63	16.718	1583589	0.10	759197	0.16	2.09	Vitamin E
64	17.028	1601759	0.10	910107	0.20	1.76	Octacosyl acetate
65	17.237	2723439	0.17	1658891	0.36	1.64	Hexadecanal
66	17.375	1075345	0.07	564914	0.12	1.90	Ergost-5-en-3-ol, (3.beta.)-
67	17.462	19436074	1.18	11052496	2.38	1.76	16-Hentriacontanone
68	17.517	4464947	0.27	2266594	0.49	1.97	Stigmasterol
69	17.589	6456464	0.39	3359491	0.72	1.92	1-Heptacosanol
70	17.682	625436	0.04	248810	0.05	2.51	2-Nonadecanone
71	17.899	11180284	0.68	5718453	1.23	1.96	.gamma.-Sitosterol
72	18.223	888156	0.05	383586	0.08	2.32	Octacosyl acetate
73	18.517	3478300	0.21	1163947	0.25	2.99	Hexadecanal
74	18.866	3814940	0.23	1378725	0.30	2.77	Oleic acid, 3-(octadecyloxy)propyl ester
75	18.990	1635467	0.10	664488	0.14	2.46	Tetracosyl trifluoroacetate
		1641831541	100.00	463587565	100.00		

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 5:27:29 PM
Sample Type	: Unknown
Sample Name	: PA-HEX-Ghana
Sample ID	: Akwasi
Vial #	: 35
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram PA HEX-Ghana



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	2.736	1170614	0.06	654321	0.10	1.79	2-Pentanol, 4-methyl-
2	7.349	4082955	0.23	4177731	0.67	0.98	.alpha.-Cubebene
3	7.594	1674778	0.09	1703690	0.27	0.98	.alfa.-Copaene
4	7.673	4495281	0.25	4640672	0.74	0.97	.beta.-copaene
5	7.958	22556018	1.24	19876073	3.17	1.13	Caryophyllene
6	8.214	2573716	0.14	2724015	0.43	0.94	Humulene
7	8.247	1294144	0.07	1338502	0.21	0.97	Aromandendrene
8	8.392	12577100	0.69	11833209	1.89	1.06	1,6-Cyclodecadiene, 1-methyl-5-methylene-8-
9	8.491	4718328	0.26	3689702	0.59	1.28	.gamma.-Elemene
10	8.619	3319346	0.18	2646019	0.42	1.25	cubedol
11	8.951	1192059	0.07	1216913	0.19	0.98	1,5-Cyclodecadiene, 1,5-dimethyl-8-(1-methyl
12	9.063	1895864	0.10	1098980	0.18	1.73	(-)-Spathulenol
13	9.119	1467715	0.08	1201503	0.19	1.22	Caryophyllene oxide
14	9.838	2189642	0.12	2173340	0.35	1.01	Heptadecanal
15	10.088	1376785	0.08	1177351	0.19	1.17	Tetradecanoic acid
16	10.565	9511043	0.52	9016094	1.44	1.05	Phytol, acetate
17	10.598	1316206	0.07	1212889	0.19	1.09	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
18	10.705	1348511	0.07	1260576	0.20	1.07	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
19	10.819	4825455	0.27	2988594	0.48	1.61	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
20	11.302	24832928	1.37	13277794	2.12	1.87	Pentadecanoic acid
21	11.549	1066821	0.06	1190084	0.19	0.90	Cyclohexane, 2-chloro-4-methyl-1-(1-methyle
22	11.838	2632233	0.15	2309688	0.37	1.14	9-Octadecen-1-ol, (Z)-
23	11.903	4818834	0.27	5114707	0.82	0.94	9,17-Octadecadienal, (Z)-
24	11.957	1771318	0.10	1470596	0.23	1.20	Tetradecyl trifluoroacetate
25	12.093	8815864	0.49	8148044	1.30	1.08	Phytol
26	12.263	25364944	1.40	9042697	1.44	2.81	Cyclopropaneoctanoic acid, 2-[[2-[(2-ethylcyc

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	12.357	4200595	0.23	2611233	0.42	1.61	Octadecanoic acid
28	12.479	36976676	2.04	25930036	4.14	1.43	Di(1-decynyl)mercury
29	12.548	2642238	0.15	1734193	0.28	1.52	3-Methyl-1-dodecyn-3-ol
30	12.603	7763947	0.43	4161035	0.66	1.87	Phytol, acetate
31	12.656	5395519	0.30	2689241	0.43	2.01	cis-9-Hexadecenoic acid, trimethylsilyl ester
32	13.442	126342403	6.97	28490962	4.55	4.43	2-Hydroxy-(Z)9-pentadecenyl propanoate
33	13.609	190677707	10.52	40136957	6.41	4.75	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
34	13.756	124694638	6.88	25468771	4.07	4.90	11-Dodecyn-1-ol acetate
35	13.889	232036684	12.80	46031300	7.35	5.04	7-Octen-3-ol, 2,3,6-trimethyl-
36	14.012	183926768	10.14	36386136	5.81	5.05	7-Octen-3-ol, 2,3,6-trimethyl-
37	14.094	46613411	2.57	23022646	3.67	2.02	11-Dodecyn-1-ol acetate
38	14.133	17775395	0.98	12618320	2.01	1.41	3-Isopropyl-4-methyl-dec-1-en-4-ol
39	14.174	11333302	0.63	4637064	0.74	2.44	Estran-3-one, 17-(acetyloxy)-2-methyl-, (2.alp
40	14.325	30061310	1.66	9366168	1.49	3.21	1,2-15,16-Diepoxyhexadecane
41	14.494	34810901	1.92	14489580	2.31	2.40	(R)-(-)-14-Methyl-8-hexadecyn-1-ol
42	14.598	6767225	0.37	1533070	0.24	4.41	1,3-Dioxolane, 2-(1,1-dimethylethyl)-2-methyl
43	14.707	29113704	1.61	6783469	1.08	4.29	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
44	14.824	22561030	1.24	10460121	1.67	2.16	2-methyloctacosane
45	15.108	27592054	1.52	15595613	2.49	1.77	Ethanol, 2-(9,12-octadecadienyloxy)-, (Z,Z)-
46	15.206	18443623	1.02	5923441	0.95	3.11	13-Docosenamide, (Z)-
47	15.447	22665586	1.25	6323463	1.01	3.58	9-Methyl-10,12-hexadecadien-1-ol acetate
48	15.552	5470767	0.30	1224507	0.20	4.47	Z-5,17-Octadecadien-1-ol acetate
49	15.631	39148085	2.16	26113757	4.17	1.50	Tetratetracontane
50	15.775	2979921	0.16	826082	0.13	3.61	Oxirane, 2,2-dimethyl-3-(3,7,12,16,20-pentam
51	16.010	5659740	0.31	3454649	0.55	1.64	Tetratetracontane
52	16.058	1462400	0.08	880369	0.14	1.66	Tetracosyl acetate
53	16.214	8812469	0.49	6166114	0.98	1.43	Hexadecanal
54	16.312	5142702	0.28	3015099	0.48	1.71	.gamma.-Tocopherol
55	16.462	43046791	2.37	18739096	2.99	2.30	Tetracosane
56	16.512	15402462	0.85	10424429	1.66	1.48	1-Heptacosanol
57	16.585	2090731	0.12	756719	0.12	2.76	6,10,14-Trimethyl-pentadecan-2-ol
58	16.712	60719459	3.35	22451831	3.58	2.70	Vitamin E
59	16.992	5884643	0.32	3267728	0.52	1.80	Triacontyl acetate
60	17.204	8928751	0.49	4316639	0.69	2.07	Hexadecanal
61	17.335	8250714	0.46	4150671	0.66	1.99	Ergost-5-en-3-ol, (3.beta.)-
62	17.457	116541806	6.43	33257695	5.31	3.50	16-Hentriacontanone
63	17.551	10499880	0.58	5249735	0.84	2.00	Heneicosyl trifluoroacetate
64	17.645	1203003	0.07	466038	0.07	2.58	Oxirane, [(hexadecyloxy)methyl]-
65	17.712	5628981	0.31	1263291	0.20	4.46	dl-.alpha.-Tocopherol
66	17.896	71154937	3.92	21581642	3.44	3.30	.gamma.-Sitosterol
67	17.958	2390918	0.13	1263007	0.20	1.89	Stigmastanol
68	18.167	2899764	0.16	1227432	0.20	2.36	Triacontyl acetate
69	18.496	21888105	1.21	5399402	0.86	4.05	dl-.alpha.-Tocopherol
70	18.612	1161716	0.06	486869	0.08	2.39	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
71	18.752	6053659	0.33	2355009	0.38	2.57	Cholest-4-en-3-one
72	18.819	17134778	0.95	5970378	0.95	2.87	17-Pentatriacontene
73	18.926	1691932	0.09	771764	0.12	2.19	Docosyl trifluoroacetate
74	19.230	2353637	0.13	636897	0.10	3.70	Phytol, acetate
75	20.432	4226100	0.23	1229137	0.20	3.44	9,19-Cyclolanostan-3-ol, 24-methylene-, (3.be
		1813112069	100.00	626522589	100.00		

GC-MS ANALYSIS OF *PERSEA AMERICANA*

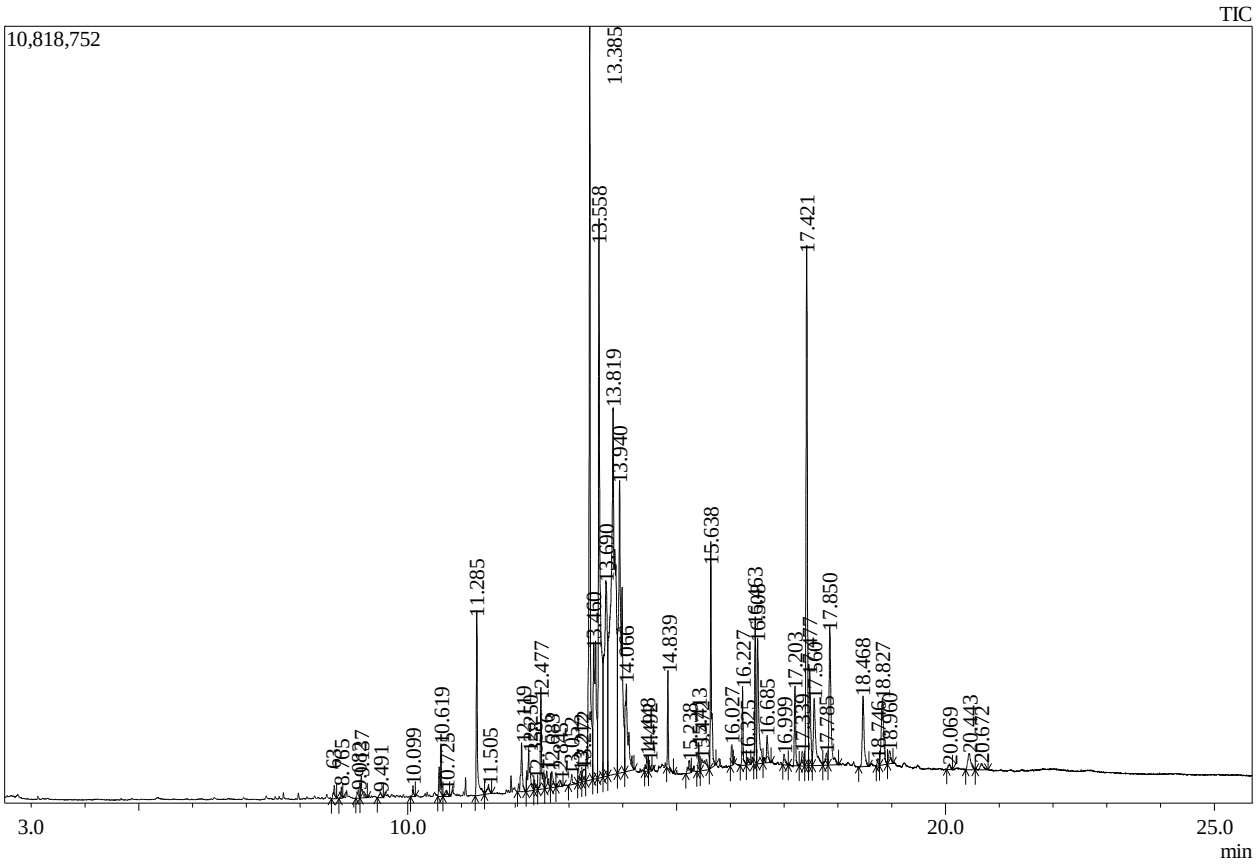
(HEXANE)- South Africa

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/08/02 4:27:36 AM
Sample Type	: Unknown
Sample Name	: PA-HEX-SA
Sample ID	: Akwasi
Vial #	: 85
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 07 03.qgt

Chromatogram PA-HEX-SA



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	8.635	324278	0.19	165307	0.25	1.96	cubedol
2	8.765	192186	0.11	153172	0.23	1.25	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4
3	9.082	267783	0.16	107478	0.16	2.49	1H-Cycloprop[e]azulen-7-ol, decahydro-1,1,7
4	9.137	548342	0.32	286087	0.43	1.92	Caryophyllene oxide
5	9.491	190081	0.11	68407	0.10	2.78	1H-Cycloprop[e]azulene, decahydro-1,1,7-tri
6	10.099	199870	0.12	147912	0.22	1.35	Eicosanoic acid
7	10.619	1251009	0.73	705738	1.06	1.77	2-Pentadecanone, 6,10,14-trimethyl-
8	10.725	217666	0.13	78836	0.12	2.76	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
9	11.285	3807647	2.23	2516234	3.79	1.51	Pentadecanoic acid
10	11.505	392853	0.23	121771	0.18	3.23	6,11-Dimethyl-2,6,10-dodecatrien-1-ol
11	12.119	1438438	0.84	664086	1.00	2.17	Phytol
12	12.250	1731546	1.01	549314	0.83	3.15	Z,Z-8,10-Hexadecadien-1-ol
13	12.358	255914	0.15	140938	0.21	1.82	Octadecanoic acid
14	12.477	2103879	1.23	1356112	2.04	1.55	Hexadecanamide
15	12.586	715879	0.42	241456	0.36	2.96	8-Hexadecanol
16	12.683	544926	0.32	220697	0.33	2.47	30-Norlupan-28-oic acid, 3-hydroxy-21-metho
17	12.845	203299	0.12	77705	0.12	2.62	Kauran-18-al, 17-(acetyloxy)-, (4.beta.)-
18	13.052	261722	0.15	136433	0.21	1.92	Eicosane
19	13.212	451003	0.26	139381	0.21	3.24	cis-9-Hexadecenoic acid, trimethylsilyl ester
20	13.272	620492	0.36	168162	0.25	3.69	1-Heptanol, 7-(octylthio)-
21	13.385	17740101	10.37	10105459	15.21	1.76	9-Octadecenamide, (Z)-
22	13.460	7010793	4.10	1887810	2.84	3.71	11-Dodecyn-1-ol acetate
23	13.558	18045162	10.55	7578841	11.41	2.38	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
24	13.690	10878089	6.36	2681581	4.04	4.06	3-Methyl-2-(2-methylene-cyclohexyl)-butan-2
25	13.819	30064895	17.58	5044126	7.59	5.96	7-Octen-3-ol, 2,3,6-trimethyl-
26	13.940	15937393	9.32	3898461	5.87	4.09	10-Methyl-E-11-tridece-1-ol acetate

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	14.066	4309785	2.52	1172018	1.76	3.68	10-Methyl-E-11-tridece-1-ol acetate
28	14.448	413719	0.24	204770	0.31	2.02	cis-4,7,10,13,16,19-Docosahexaenoic acid, ter
29	14.492	244590	0.14	149979	0.23	1.63	2-Octylcyclopropene-1-heptanol
30	14.839	1393993	0.81	1276889	1.92	1.09	Tetratetracontane
31	15.238	440139	0.26	166644	0.25	2.64	Eicosane
32	15.413	458368	0.27	373607	0.56	1.23	Hexadecanal
33	15.472	984328	0.58	165130	0.25	5.96	Octadecanoic acid, 9,10-epoxy-, isopropyl este
34	15.638	4428688	2.59	3035183	4.57	1.46	Tetratetracontane
35	16.027	461430	0.27	279293	0.42	1.65	Tetratetracontane
36	16.227	1404054	0.82	995668	1.50	1.41	Oxirane, heptadecyl-
37	16.325	209382	0.12	96531	0.15	2.17	.gamma.-Tocopherol
38	16.463	2635646	1.54	1957781	2.95	1.35	Tetratetracontane
39	16.508	3935032	2.30	1694852	2.55	2.32	1-Heptacosanol
40	16.685	852472	0.50	371015	0.56	2.30	.alpha.-Tocopheryl acetate
41	16.999	325173	0.19	142765	0.21	2.28	Octacosyl acetate
42	17.203	1733538	1.01	1059928	1.60	1.64	Oxirane, heptadecyl-
43	17.339	491310	0.29	202193	0.30	2.43	Ergost-5-en-3-ol, (3.beta.)-
44	17.421	12589073	7.36	7073076	10.65	1.78	16-Hentriacontanone
45	17.477	2737164	1.60	1284298	1.93	2.13	Stigmasterol
46	17.560	2915564	1.70	922549	1.39	3.16	1-Heptacosanol
47	17.785	456245	0.27	154993	0.23	2.94	Oxirane, heptadecyl-
48	17.850	4729636	2.77	1886252	2.84	2.51	.gamma.-Sitosterol
49	18.468	2441277	1.43	959556	1.44	2.54	Hexadecanal
50	18.746	217854	0.13	101290	0.15	2.15	Stigmast-4-en-3-one
51	18.827	2824333	1.65	908603	1.37	3.11	Oxirane, hexadecyl-
52	18.960	512477	0.30	178438	0.27	2.87	1-Heptacosanol
53	20.069	204068	0.12	64709	0.10	3.15	Stigmastane-3,6-dione, (5.alpha.)-
54	20.443	905894	0.53	222684	0.34	4.07	9,19-Cyclolanostan-3-ol, 24-methylene-, (3.be
55	20.672	396052	0.23	87082	0.13	4.55	9,19-Cyclolanost-23-ene-3,25-diol, 3-acetate,
		171046530	100.00	66429280	100.00		

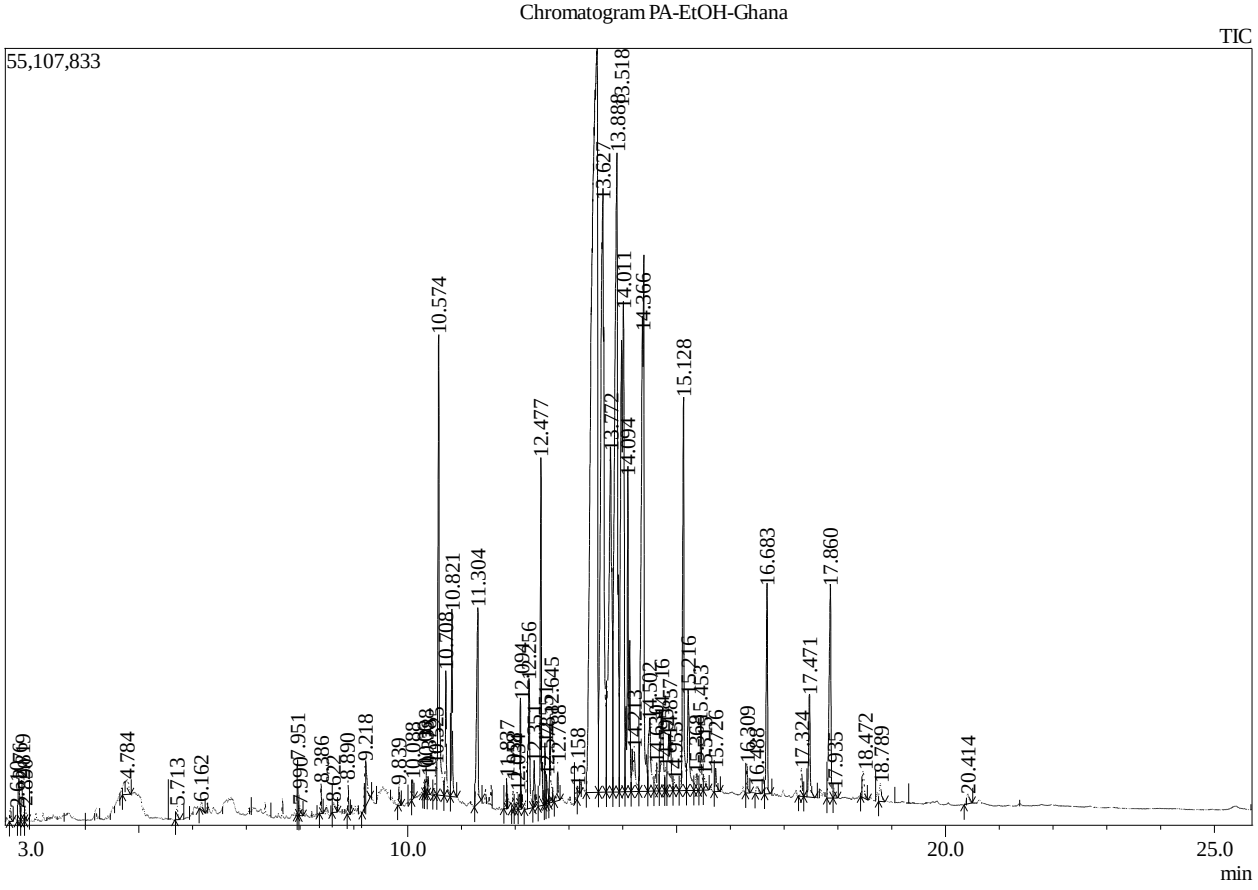
GC-MS ANALYSIS OF *PERSEA AMERICANA* (ETHANOL)- Ghana

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 7:15:08 PM
Sample Type	: Unknown
Sample Name	: PA-EtOH-Ghana
Sample ID	: Akwasi
Vial #	: 38
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram PA-EtOH-Ghana



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	2.610	1491989	0.09	936134	0.18	1.59	2-Propenoic acid, oxiranylmethyl ester
2	2.769	5544574	0.32	2968253	0.56	1.87	2,3-Butanediol, [R-(R*,R*)]-
3	2.819	5817217	0.33	2898591	0.55	2.01	2,3-Butanediol, [R-(R*,R*)]-
4	2.890	1422862	0.08	1052859	0.20	1.35	Ethyl orthoformate
5	4.784	6552196	0.37	1061861	0.20	6.17	(3-Methyl-oxiran-2-yl)-methanol
6	5.713	1723466	0.10	962008	0.18	1.79	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6
7	6.162	1486424	0.08	509265	0.10	2.92	Isosorbide
8	7.951	4045057	0.23	3919848	0.74	1.03	Caryophyllene
9	7.990	1571670	0.09	734603	0.14	2.14	6-Methyl-6-(5-methylfuran-2-yl)heptan-2-one
10	8.386	2374679	0.14	2047078	0.39	1.16	1,6-Cyclodecadiene, 1-methyl-5-methylene-8-
11	8.622	2514781	0.14	712129	0.13	3.53	4-epi-cubedol
12	8.890	2594982	0.15	2360809	0.45	1.10	2-Hexanone, 3-methyl-4-methylene-
13	9.218	10808149	0.62	3215947	0.61	3.36	1,3-Benzenediol, 4-propyl-
14	9.839	1632225	0.09	1446447	0.27	1.13	2-Cyclohexen-1-one, 4-(3-hydroxybutyl)-3,5,5
15	10.088	1417337	0.08	1399871	0.26	1.01	Tetradecanoic acid
16	10.306	1578918	0.09	1045877	0.20	1.51	5-Isopropyl-6-methyl-hepta-3,5-dien-2-ol
17	10.338	2191614	0.13	1940716	0.37	1.13	3,7,11,Trimethyl-8,10- dodecedienylacetate
18	10.378	1693795	0.10	1269361	0.24	1.33	6,11-Undecadiene, 1-acetoxy-3,7-dimethyl-
19	10.525	3811936	0.22	2148555	0.41	1.77	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
20	10.574	54674146	3.13	32639326	6.18	1.68	Phytol, acetate
21	10.708	13003171	0.74	8889329	1.68	1.46	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
22	10.821	17240757	0.99	13232472	2.50	1.30	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
23	11.304	28757321	1.64	13728394	2.60	2.09	Pentadecanoic acid
24	11.837	3268251	0.19	2131832	0.40	1.53	9-Octadecen-1-ol, (Z)-
25	11.958	1619093	0.09	1277463	0.24	1.27	1-Heneicosanol
26	12.034	2345244	0.13	1219305	0.23	1.92	2-Nonadecanone, O-methyloxime

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	12.094	9680710	0.55	7796836	1.48	1.24	Phytol
28	12.256	23984124	1.37	9128622	1.73	2.63	Cyclopropaneoctanoic acid, 2-[[2-[(2-ethylcyc
29	12.351	3631783	0.21	3264046	0.62	1.11	Octadecanoic acid
30	12.477	32384567	1.85	23994088	4.54	1.35	Di(1-decynyl)mercury
31	12.551	5501805	0.31	4362303	0.83	1.26	3-Methyl-1-dodecyn-3-ol
32	12.578	3908334	0.22	2059923	0.39	1.90	3-Methyl-1-dodecyn-3-ol
33	12.645	9194133	0.53	6229440	1.18	1.48	Cyclopropanecarboxylic acid, undec-2-enyl es
34	12.788	3736418	0.21	2711476	0.51	1.38	Cyclopropanecarboxyllic acid, tridec-2-ynyl es
35	13.158	1654313	0.09	822499	0.16	2.01	(2-Methyl-[1,3]dioxolan-2-yl)-acetic acid, phe
36	13.518	415816028	23.77	52404801	9.92	7.93	2-Hydroxy-(Z)9-pentadecenyl propanoate
37	13.627	186980622	10.69	41971455	7.94	4.45	Z,Z-4,6-Nonadecadien-1-ol acetate
38	13.772	104356962	5.97	23806142	4.51	4.38	11-Dodecyn-1-ol acetate
39	13.888	179968235	10.29	44226102	8.37	4.07	7-Octen-3-ol, 2,3,6-trimethyl-
40	14.011	142656539	8.16	33429410	6.33	4.27	7-Octen-3-ol, 2,3,6-trimethyl-
41	14.094	52374975	2.99	22766360	4.31	2.30	10-Methyl-E-11-tridece-1-ol acetate
42	14.213	11607994	0.66	3082484	0.58	3.77	2-Nonadecanone, O-methyloxime
43	14.366	131479805	7.52	32898477	6.23	4.00	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
44	14.502	16864748	0.96	5117665	0.97	3.30	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
45	14.630	6886800	0.39	1987119	0.38	3.47	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
46	14.716	19300939	1.10	5547909	1.05	3.48	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
47	14.795	3108932	0.18	1598631	0.30	1.94	11-Dodecyn-1-ol acetate
48	14.857	15436797	0.88	3984075	0.75	3.87	9,12-Octadecadienoic acid (Z,Z)-, 2,3-dihydro
49	14.955	2192488	0.13	793689	0.15	2.76	Octadecanoic acid, 2,3-dihydroxypropyl ester
50	15.128	51335001	2.94	27223374	5.15	1.89	Ethanol, 2-(9,12-octadecadienyloxy)-, (Z,Z)-
51	15.216	18301831	1.05	7038491	1.33	2.60	13-Docosenamide, (Z)-
52	15.368	3800562	0.22	1143500	0.22	3.32	9-Methyl-10,12-hexadecadien-1-ol acetate
53	15.453	9844390	0.56	4886605	0.92	2.01	9-Methyl-10,12-hexadecadien-1-ol acetate
54	15.515	2869027	0.16	1106635	0.21	2.59	11-Dodecyn-1-ol acetate
55	15.726	2618604	0.15	1620911	0.31	1.62	2,4-Hexadienedioic acid, 3,4-diethyl-, dimethy
56	16.309	3017496	0.17	2037157	0.39	1.48	.gamma.-Tocopherol
57	16.488	1518083	0.09	479585	0.09	3.17	Eicosyl pentafluoropropionate
58	16.683	24989884	1.43	14530385	2.75	1.72	Vitamin E
59	17.324	3412173	0.20	1909609	0.36	1.79	Ergost-5-en-3-ol, (3.beta.)-
60	17.471	14789030	0.85	7111622	1.35	2.08	Stigmasterol
61	17.860	36794937	2.10	15028157	2.84	2.45	.gamma.-Sitosterol
62	17.935	1436217	0.08	649654	0.12	2.21	Stigmastanol
63	18.472	4518952	0.26	1889684	0.36	2.39	dl-.alpha.-Tocopherol
64	18.789	3438797	0.20	1268955	0.24	2.71	17-Pentatriacontene
65	20.414	2432768	0.14	737053	0.14	3.30	9,19-Cyclolanost-23-ene-3,25-diol, 3-acetate,
		1749007657	100.00	528393262	100.00		

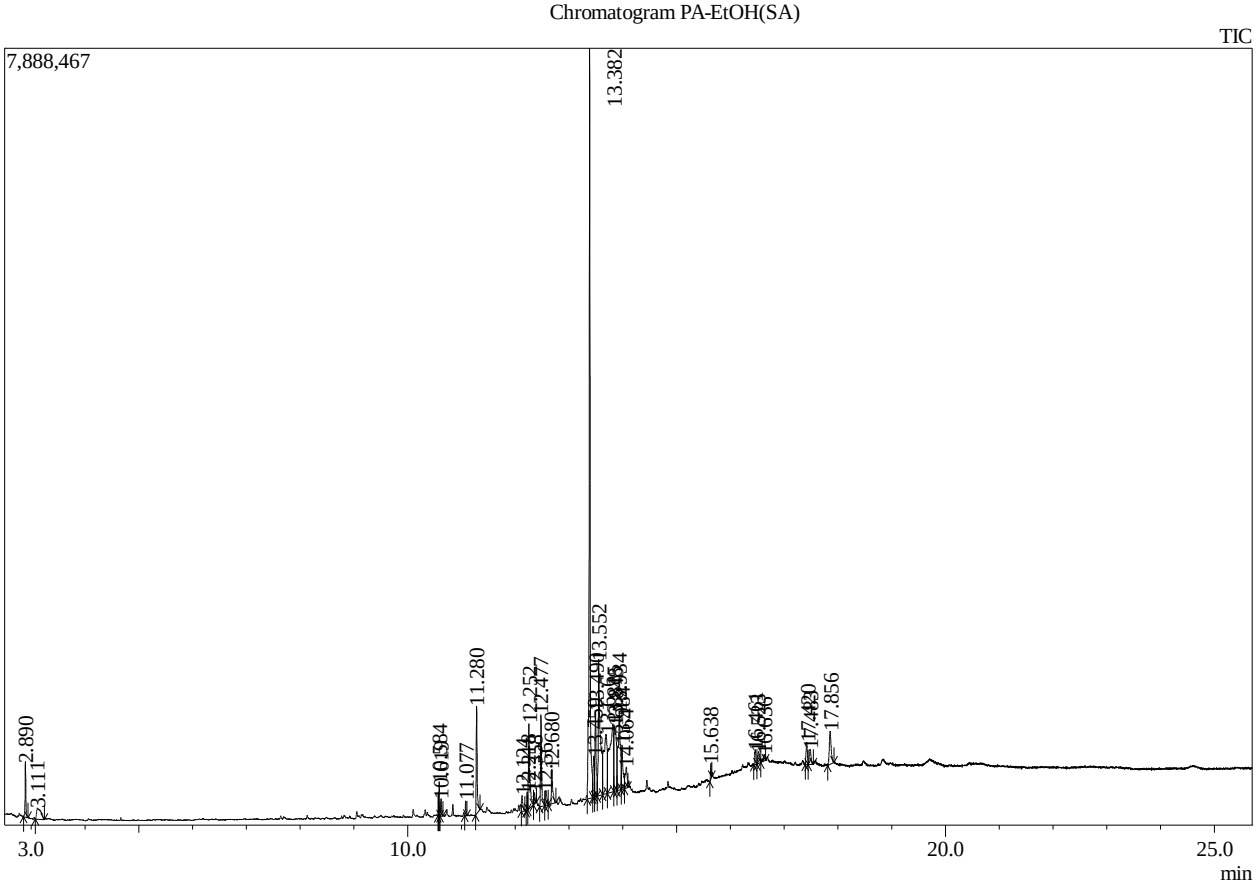
GC-MS ANALYSIS OF *PERSEA AMERICANA* (ETHANOL)- South Africa

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/08/02 3:51:29 AM
Sample Type	: Unknown
Sample Name	PA-: EtOH(SA)
Sample ID	: Akwasi
Vial #	: 84
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 07 03.qgt

Chromatogram PA-EtOH(SA)



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	2.890	517940	1.38	541658	2.73	0.96	Formamide, N-methoxy-
2	3.111	641988	1.71	113522	0.57	5.66	Benzene, chloro-
3	10.584	362727	0.97	347796	1.75	1.04	Phytol, acetate
4	10.619	201980	0.54	176335	0.89	1.15	2-Pentadecanone, 6,10,14-trimethyl-
5	11.077	145138	0.39	145594	0.73	1.00	1-Hexacosene
6	11.280	1314787	3.50	1064565	5.36	1.24	Pentadecanoic acid
7	12.124	269675	0.72	159516	0.80	1.69	Phytol
8	12.218	225919	0.60	179878	0.91	1.26	9,12-Octadecadienoic acid (Z,Z)-
9	12.252	1540140	4.10	855643	4.31	1.80	cis,cis,cis-7,10,13-Hexadecatrienal
10	12.358	139176	0.37	116112	0.58	1.20	Octadecanoic acid
11	12.477	1171102	3.12	890092	4.48	1.32	Hexadecanamide
12	12.566	288537	0.77	146551	0.74	1.97	Silane, dimethyl(2-decyloxy)propoxy-
13	12.680	558764	1.49	344112	1.73	1.62	cis-9-Hexadecenoic acid, trimethylsilyl ester
14	13.382	9873063	26.27	7531692	37.93	1.31	9-Octadecenamide, (Z)-
15	13.459	690727	1.84	417104	2.10	1.66	11-Dodecyn-1-ol acetate
16	13.490	1383822	3.68	858767	4.32	1.61	Octadecanamide
17	13.552	3759552	10.00	1376930	6.93	2.73	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
18	13.686	2655749	7.07	598958	3.02	4.43	2,3,4-Trimethyl-5-hexen-3-ol
19	13.815	3770070	10.03	689106	3.47	5.47	7-Octen-3-ol, 2,3,6-trimethyl-
20	13.848	2023221	5.38	665374	3.35	3.04	Geranyl acetate, 2,3-epoxy-
21	13.934	2307565	6.14	787517	3.97	2.93	10-Methyl-E-11-tridece-1-ol acetate
22	13.984	914342	2.43	450744	2.27	2.03	10-Methyl-E-11-tridece-1-ol acetate
23	14.064	513179	1.37	214303	1.08	2.39	10-Methyl-E-11-tridece-1-ol acetate
24	15.638	150090	0.40	149588	0.75	1.00	Heneicosane
25	16.461	198224	0.53	149773	0.75	1.32	Tetratetracontane
26	16.523	281126	0.75	118298	0.60	2.38	n-Tetracosanol-1

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	16.636	154062	0.41	62900	0.32	2.45	3.alpha.,7.beta.-Dihydroxy-5.beta.,6.beta.-epo
28	17.420	383201	1.02	222113	1.12	1.73	16-Hentriacontanone
29	17.483	341685	0.91	153202	0.77	2.23	Stigmasterol
30	17.856	810011	2.15	328834	1.66	2.46	.gamma.-Sitosterol
		37587562	100.00	19856577	100.00		

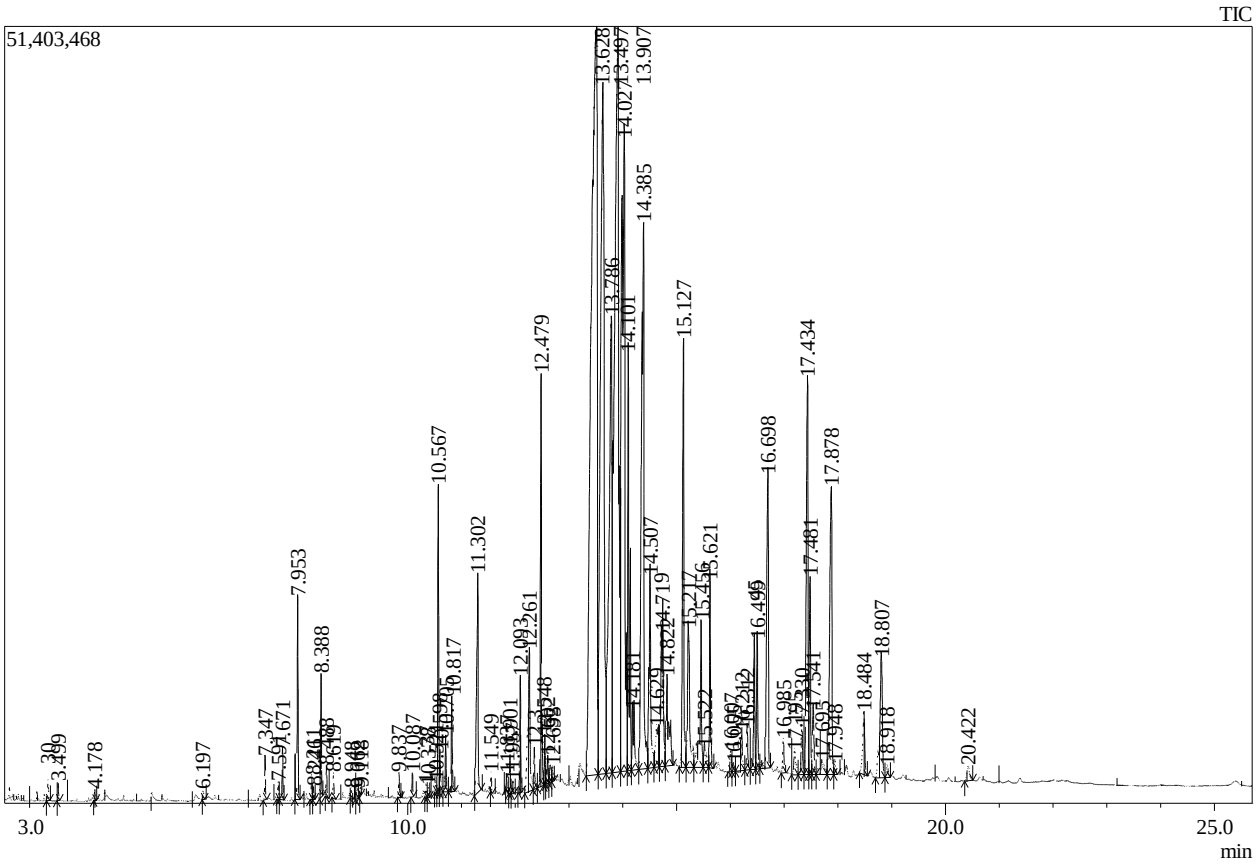
GC-MS ANALYSIS OF *PERSEA AMERICANA* (ETHYL ACETATE)- Ghana

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 7:50:57 PM
Sample Type	: Unknown
Sample Name	: PA-EtoAc-Ghana
Sample ID	: Akwasi
Vial #	: 39
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram PAEtoAc-Ghana



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	3.309	1463234	0.07	1306780	0.20	1.12	Benzene, 1,3-dimethyl-
2	3.499	1137389	0.05	1263210	0.19	0.90	Pentanoic acid, ethyl ester
3	4.178	594636	0.03	709981	0.11	0.84	Bicyclo[3.1.0]hexane, 4-methylene-1-(1-meth
4	6.197	716107	0.03	647465	0.10	1.11	1-Heptanol, 2-propyl-
5	7.347	2903220	0.14	2857081	0.44	1.02	.alpha.-Cubebene
6	7.591	1157941	0.06	1186778	0.18	0.98	.alfa.-Copaene
7	7.671	3584378	0.17	3499888	0.53	1.02	.beta.-copaene
8	7.953	15267414	0.73	13179090	2.01	1.16	Caryophyllene
9	8.211	1690749	0.08	1632907	0.25	1.04	Humulene
10	8.246	815511	0.04	824532	0.13	0.99	Aromandendrene
11	8.388	8361394	0.40	8177873	1.25	1.02	1,6-Cyclodecadiene, 1-methyl-5-methylene-8-
12	8.488	2184693	0.10	2076211	0.32	1.05	.gamma.-Elemene
13	8.619	1893668	0.09	1687055	0.26	1.12	cubedol
14	8.948	736613	0.04	728477	0.11	1.01	1,5-Cyclodecadiene, 1,5-dimethyl-8-(1-methyl
15	9.062	1128204	0.05	682331	0.10	1.65	(-)-Spathulenol
16	9.118	574864	0.03	591226	0.09	0.97	Caryophyllene oxide
17	9.837	1746620	0.08	1611926	0.25	1.08	Heptadecanal
18	10.087	1716303	0.08	1591913	0.24	1.08	Tetradecanoic acid
19	10.337	918534	0.04	861546	0.13	1.07	3,7,11,Trimethyl-8,10- dodecedienylacetate
20	10.378	829884	0.04	786788	0.12	1.05	Cyclohexane, 2-chloro-4-methyl-1-(1-methyle
21	10.521	944050	0.05	761052	0.12	1.24	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
22	10.567	22876198	1.10	19763105	3.02	1.16	Phytol, acetate
23	10.598	4476464	0.22	2783443	0.42	1.61	2-Hexadecene, 3,7,11,15-tetramethyl-, [R-[R*
24	10.705	5450925	0.26	3763695	0.57	1.45	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
25	10.817	7909229	0.38	6207771	0.95	1.27	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
26	11.302	28637545	1.38	14390521	2.20	1.99	Pentadecanoic acid

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	11.549	1263924	0.06	1346180	0.21	0.94	Cyclohexane, 2-chloro-4-methyl-1-(1-methyle
28	11.837	887411	0.04	1048540	0.16	0.85	9-Octadecen-1-ol, (Z)-
29	11.901	2329069	0.11	2511027	0.38	0.93	9,17-Octadecadienal, (Z)-
30	11.957	942904	0.05	816810	0.12	1.15	n-Heptadecanol-1
31	12.093	8319204	0.40	7574846	1.16	1.10	Phytol
32	12.261	25624392	1.23	9463973	1.44	2.71	cis,cis,cis-7,10,13-Hexadecatrienal
33	12.354	3921376	0.19	2754593	0.42	1.42	Octadecanoic acid
34	12.479	37672468	1.81	27013945	4.12	1.39	Di(1-decynyl)mercury
35	12.548	3997618	0.19	3332765	0.51	1.20	3-Methyl-1-dodecyn-3-ol
36	12.602	3167263	0.15	1873540	0.29	1.69	Phytol, acetate
37	12.652	2497343	0.12	1149402	0.18	2.17	2-Isopropenyl-5-methylhex-4-enal
38	12.695	1290391	0.06	1074079	0.16	1.20	3-Methyl-1-dodecyn-3-ol
39	13.497	377582906	18.15	48880256	7.46	7.72	2-Hydroxy-(Z)9-pentadecenyl propanoate
40	13.628	219442341	10.55	45308604	6.92	4.84	10,11-Epoxy-n-undecan-1-ol
41	13.786	115639063	5.56	29836183	4.56	3.88	11-Dodecyn-1-ol acetate
42	13.907	276529274	13.29	48817771	7.45	5.66	7-Octen-3-ol, 2,3,6-trimethyl-
43	14.027	188368481	9.05	42295280	6.46	4.45	7-Octen-3-ol, 2,3,6-trimethyl-
44	14.101	50240196	2.41	27472881	4.19	1.83	10-Methyl-E-11-tridece-1-ol acetate
45	14.181	12703959	0.61	4358603	0.67	2.91	Estran-3-one, 17-(acetyloxy)-2-methyl-, (2.alp
46	14.385	117777499	5.66	35094876	5.36	3.36	Cyclopropanecarboxylic acid, heptadecyl ester
47	14.507	30092269	1.45	12867687	1.96	2.34	9-Eicosyne
48	14.629	10929302	0.53	2738218	0.42	3.99	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
49	14.719	31901445	1.53	8984313	1.37	3.55	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
50	14.822	19700927	0.95	5815797	0.89	3.39	Tetratetracontane
51	15.127	50194113	2.41	27656444	4.22	1.81	Ethanol, 2-(9,12-octadecadienyloxy)-, (Z,Z)-
52	15.217	27628105	1.33	9366898	1.43	2.95	13-Docosenamide, (Z)-
53	15.456	21892190	1.05	9272032	1.42	2.36	9-Methyl-10,12-hexadecadien-1-ol acetate
54	15.522	4153297	0.20	1512019	0.23	2.75	9-Methyl-10,12-hexadecadien-1-ol acetate
55	15.621	16189044	0.78	12668084	1.93	1.28	Tetratetracontane
56	16.007	2288900	0.11	1353854	0.21	1.69	Tetratetracontane
57	16.055	999684	0.05	561595	0.09	1.78	Tetracosyl acetate
58	16.212	4068953	0.20	2669402	0.41	1.52	Hexadecanal
59	16.312	4660014	0.22	2812469	0.43	1.66	.gamma.-Tocopherol
60	16.445	14758858	0.71	8416794	1.29	1.75	Tetratetracontane
61	16.499	13976561	0.67	8586195	1.31	1.63	1-Heptacosanol
62	16.698	44928005	2.16	19030081	2.91	2.36	Vitamin E
63	16.985	3922266	0.19	2184012	0.33	1.80	Octacosyl acetate
64	17.195	4529638	0.22	1679060	0.26	2.70	Hexadecanal
65	17.330	6345268	0.30	3128750	0.48	2.03	Ergost-5-en-3-ol, (3.beta.)-
66	17.434	60094418	2.89	26075782	3.98	2.30	16-Hentriacontanone
67	17.481	23101647	1.11	12844920	1.96	1.80	Stigmasterol
68	17.541	9079967	0.44	4502793	0.69	2.02	Docosyl trifluoroacetate
69	17.695	4859996	0.23	945247	0.14	5.14	.alpha.-Tocopherol-.beta.-D-mannoside
70	17.878	55686461	2.68	18756136	2.86	2.97	.gamma.-Sitosterol
71	17.948	1790179	0.09	914033	0.14	1.96	Stigmastanol
72	18.484	11179558	0.54	4136550	0.63	2.70	dl-.alpha.-Tocopherol
73	18.807	26680906	1.28	8087298	1.23	3.30	Oleic acid, 3-(octadecyloxy)propyl ester
74	18.918	1978757	0.10	831064	0.13	2.38	Docosyl trifluoroacetate
75	20.422	3390943	0.16	998816	0.15	3.39	9,19-Cyclolanost-23-ene-3,25-diol, 3-acetate,
		2080914520	100.00	654993142	100.00		

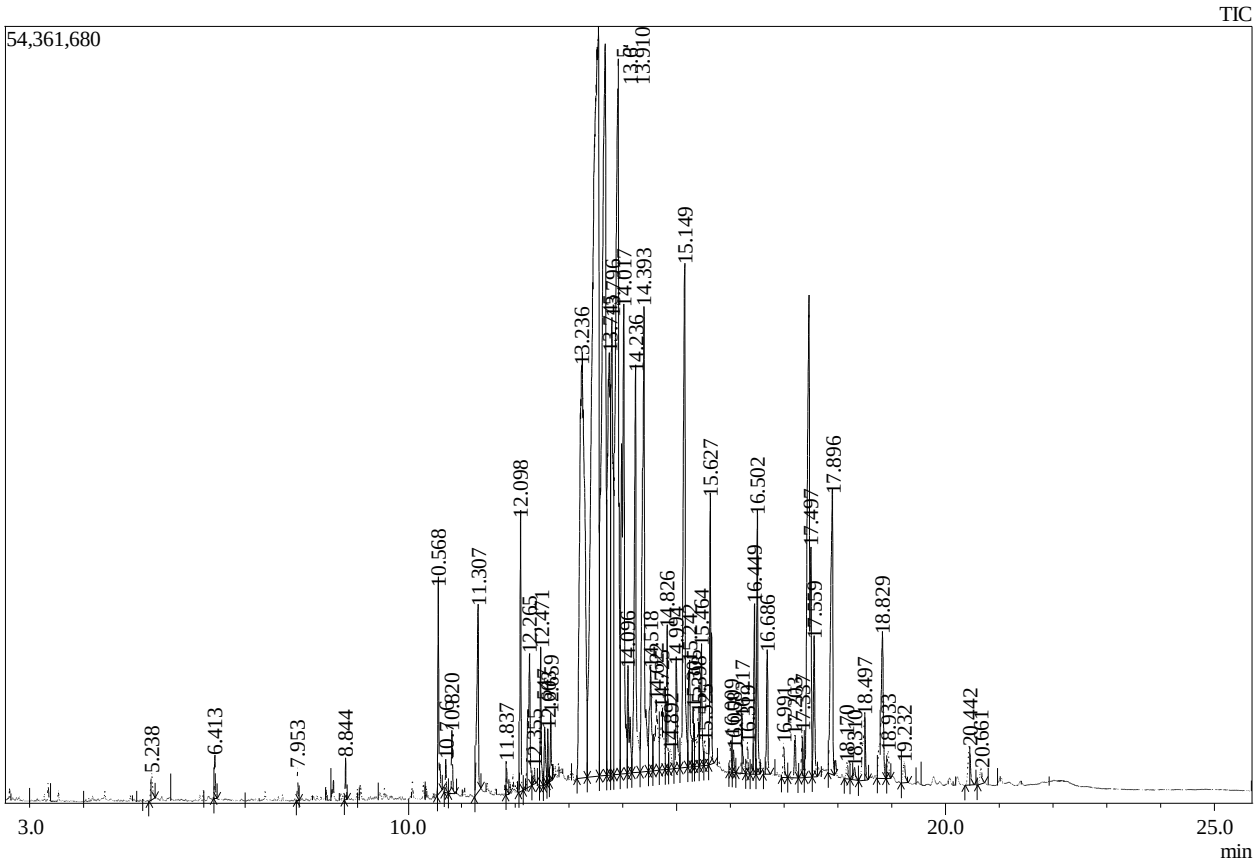
GC-MS ANALYSIS OF *PERSEA AMERICANA* (ETHYL ACETATE)- South Africa

Qualitative Analysis Report

Sample Information

Analyzed	: 2018/09/26 4:51:31 PM
Sample Type	: Unknown
Sample Name	: PA-EtoAc-SA
Sample ID	: Akwas
Vial #	: 34
Injection Volume	: 1.00
Method File	: C:\Shimadzu GCMS\Raw Data 2017-8\UKZN - M Nlooto\M Nlooto - split.qgm
Tuning File	: C:\GCMSsolution\System\Tune1\Normal Conc - 2018 09 25.qgt

Chromatogram PAEtoAc-SA



Peak Report TIC

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
1	5.238	4644587	0.19	1819623	0.27	2.55	1,2,3-Propanetriol, 1-acetate
2	6.413	3745409	0.16	2990744	0.44	1.25	1,2,3-Propanetriol, 1-acetate
3	7.953	2194774	0.09	2229885	0.33	0.98	Caryophyllene
4	8.844	2743610	0.11	2864013	0.42	0.96	1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl-, (E
5	10.568	18047398	0.75	14858294	2.20	1.21	Phytol, acetate
6	10.706	2707648	0.11	2282585	0.34	1.19	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
7	10.820	6189082	0.26	4278292	0.63	1.45	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
8	11.307	27930292	1.16	13067035	1.93	2.14	Pentadecanoic acid
9	11.837	2781237	0.12	2244637	0.33	1.24	9-Octadecen-1-ol, (Z)-
10	12.098	23016813	0.95	19414550	2.87	1.19	Phytol
11	12.265	25280444	1.05	9311098	1.38	2.72	7-Tetradecenal, (Z)-
12	12.355	1985424	0.08	1149204	0.17	1.73	Octadecanoic acid
13	12.471	11826112	0.49	9697162	1.44	1.22	Di(1-decynyl)mercury
14	12.547	7300053	0.30	4008733	0.59	1.82	3-Methyl-1-dodecyn-3-ol
15	12.603	3797774	0.16	3702795	0.55	1.03	Phytol, acetate
16	12.659	6507508	0.27	4690239	0.69	1.39	cis-9-Hexadecenoic acid, trimethylsilyl ester
17	13.236	196384386	8.14	28855140	4.27	6.81	3-Methyl-2-(2-methylene-cyclohexyl)-butan-2
18	13.547	440133256	18.24	51840431	7.67	8.49	2-Hydroxy-(Z)9-pentadecenyl propanoate
19	13.670	268530095	11.13	51030462	7.55	5.26	2-Hydroxy-(Z)9-pentadecenyl propanoate
20	13.745	112652140	4.67	28894981	4.28	3.90	(2-Methyl-[1,3]dioxolan-2-yl)-acetic acid, phe
21	13.796	92862799	3.85	31659274	4.69	2.93	(R)-(-)-14-Methyl-8-hexadecyn-1-ol
22	13.910	213451893	8.85	49445316	7.32	4.32	7-Octen-3-ol, 2,3,6-trimethyl-
23	14.017	108946365	4.52	32357425	4.79	3.37	7-Octen-3-ol, 2,3,6-trimethyl-
24	14.096	18071191	0.75	7297258	1.08	2.48	10-Methyl-E-11-tridece-1-ol acetate
25	14.236	68995595	2.86	27639788	4.09	2.50	2-Nonadecanone, O-methyloxime
26	14.393	98458053	4.08	31314524	4.64	3.14	Cyclopropanecarboxylic acid,pentadecyl ester

Peak#	R.Time	Area	Area%	Height	Height%	A/H	Name
27	14.518	20863562	0.86	7167885	1.06	2.91	E-11-Methyl-12-tetradecen-1-ol acetate
28	14.622	20729796	0.86	4684631	0.69	4.43	Acetic acid, 3-ethylpent-3-yl ester
29	14.725	18224696	0.76	4465098	0.66	4.08	Z-(13,14-Epoxy)tetradec-11-en-1-ol acetate
30	14.826	14389245	0.60	9705302	1.44	1.48	Tetratetracontane
31	14.892	4941710	0.20	1320877	0.20	3.74	11-Dodecyn-1-ol acetate
32	14.994	15384938	0.64	7285325	1.08	2.11	3-Methyl-2-(2-methylene-cyclohexyl)-butan-2
33	15.149	92186178	3.82	35088207	5.19	2.63	Ethanol, 2-(9,12-octadecadienyloxy)-, (Z,Z)-
34	15.242	20251398	0.84	6864908	1.02	2.95	Z,Z-5,16-Octadecadien-1-ol acetate
35	15.305	5617000	0.23	3863945	0.57	1.45	Squalene
36	15.398	10844969	0.45	3445635	0.51	3.15	Tetracosanal
37	15.464	17781444	0.74	8255994	1.22	2.15	9-Methyl-10,12-hexadecadien-1-ol acetate
38	15.525	4510671	0.19	1725181	0.26	2.61	Z-5,17-Octadecadien-1-ol acetate
39	15.627	32281610	1.34	17921658	2.65	1.80	Tetratetracontane
40	16.009	3822849	0.16	2475575	0.37	1.54	Tetratetracontane
41	16.058	2563531	0.11	1553354	0.23	1.65	Heptacosyl acetate
42	16.217	6683404	0.28	3965870	0.59	1.69	Hexadecanal
43	16.319	3000588	0.12	2136950	0.32	1.40	.gamma.-Tocopherol
44	16.449	18903828	0.78	11461822	1.70	1.65	Tetratetracontane
45	16.502	28212038	1.17	17201750	2.55	1.64	1-Heptacosanol
46	16.686	14195595	0.59	8433796	1.25	1.68	Vitamin E
47	16.991	4454121	0.18	2396675	0.35	1.86	Octacosyl acetate
48	17.203	5516430	0.23	3008849	0.45	1.83	Hexadecanal
49	17.337	6382344	0.26	3229303	0.48	1.98	Ergost-5-en-3-ol, (3.beta.)-
50	17.497	121049365	5.02	15852209	2.35	7.64	Stigmasterol
51	17.559	19233291	0.80	9780174	1.45	1.97	1-Heptacosanol
52	17.896	57193687	2.37	19655295	2.91	2.91	.gamma.-Sitosterol
53	18.170	2891343	0.12	1145364	0.17	2.52	Octacosyl acetate
54	18.310	2116966	0.09	868266	0.13	2.44	4,22-Stigmastadiene-3-one
55	18.497	15610069	0.65	4616902	0.68	3.38	dl-.alpha.-Tocopherol
56	18.829	33321008	1.38	9880098	1.46	3.37	Oleic acid, 3-(octadecyloxy)propyl ester
57	18.933	4288535	0.18	1929786	0.29	2.22	Tetracosyl pentafluoropropionate
58	19.232	4042036	0.17	1440873	0.21	2.81	Phytol, acetate
59	20.442	8637551	0.36	2664726	0.39	3.24	13,15-Octacosadiyne
60	20.661	3181617	0.13	1089933	0.16	2.92	9,19-Cyclolanostan-3-ol, 24-methylene-, (3.be
		2412491351	100.00	675525704	100.00		