8-6-2021

Phytochemical and biological strategies to improve essential oils content in lavender

Keefah AL-Garallaa
keefahalgarallaa@gmail.com

Phytochemical and biological strategies to improve essential oils content in lavender

By

Keefah AL-Garallaa

Approved by:

Richard L. Harkess (Major Professor)
Sorina Popescu (Minor Professor)
Guihong Bi
Tongyin Li
Jiaxu Li
Michael S. Cox (Graduate Coordinator)
Scott T. Willard (Dean, College of Agriculture and Life Sciences)

A Dissertation
Submitted to the Faculty of
Mississippi State University
in Partial Fulfillment of the Requirements
for the Degree of Doctor of Philosophy
in Horticulture
in the College of Agriculture and Life Sciences

Mississippi State, Mississippi

August 2021

Name: Keefah AL-Garallaa

Date of Degree: August 6, 2021

Institution: Mississippi State University

Major Field: Horticulture

Major Professor: Richard L. Harkess

Title of Study: Phytochemical and biological strategies to improve essential oils content in lavender

Pages in Study: 269

Candidate for Degree of Doctor of Philosophy

Lavender is an important medicinal plant that is sensitive to various environmental factors. Lavender essential oils have been shown to improve human health in response to many diseases. Lavender is grown all over the world, which indicates its ability to adapt to different climates. There are no reports of its commercial cultivation in Mississippi. It has been proven that there are many factors that directly affect the plant growth and concentration of essential oils in lavender. Studying these factors that affect plant growth and essential oils will be beneficial for lavender producers. The aim of this study is to increase the production of essential oil through cultural techniques and analyze the active compounds of three *Lavandula* species (*L. intermedia, L. angustifolia, L. stoechas)* using HPLC (High performance liquid chromatography) and GC-MS (Gas Chromatography, Mass spectrometer) technology. Applications of gibberellic acid, iron chelate, Mycorrhizae, humic acid, DAP, and potash fertilizer were studied to promote plant growth and production of essential oils. The essential oils were extracted using a Soxhlet distillation. The quantity and quality of the essential oils was also evaluated using HPLC and GC-MS. The linalool, linalyl acetate, and other compounds from *Lavandula* may be considered natural raw material source for pharmaceuticals and cosmetic products. These vital components

of the essential oil of lavender leaves, flowers, and stems (linalool, linalyl, and other compounds) were affected by chelated iron treatments, gibberellin, humic acid, and mycorrhizae. The essential oil compounds of *L. angustifolia* and *L.* x *intermedia* cultivars make them worth cultivating.

DEDICATION

For my mother. Thanks for always being with me. She left fingerprints of grace on our lives. She did not forget us.

For my father, I dedicate this dissertation to the soul of my beloved father.

For my husband Professor Rashad, where he was shadows of his soul with me.

For my sisters and my brothers Hayder, Sadiq, my brother-in-law Dr. Walid

my niece Zahraa, and my nephew Qassim for helping me survive all the stress and not allowing me to give up. The best part of life is sharing it with the ones you love.

I dedicate this dissertation to the soul of my friend Isaac Odisho.

I dedicate this dissertation to my supervisor Dr. Richard L. Harkess, who did not skimp on me with his beautiful support and help me with everything he has, and to my friend Bill Everett I dedicate this work to you, and I hope it will satisfy you.

ACKNOWLEDGEMENTS

Firstly, all praises are for God, the Exalted, Merciful and Magnificent. Many thanks and gratitude go only to God for the strength given to me to deal with the trials and tribulations during my studies. These trying moments could not have been endured without God. I thank all those who gave various kinds of support, advice and encouragement which enabled me to accomplish my research work.

I am grateful to my principal Professor and advisor Dr. Harkess who believed in my ability and capabilities. I express my sincere appreciation for his unwavering support, invaluable advice, suggestions, supervision, and crucial contributions; without which, this work would not have been possible His extensive knowledge, valuable advice and limitless enthusiasm has inspired me to produce high quality research as a researcher. His practical guidance, understanding and encouragement kept me enthusiastic throughout the research process. His sense of humor and practical advice helped keep me on the ground. I am eternally grateful for his constant encouragement throughout and his good counsel. He helped guide me while researching and writing this thesis. I could not imagine a better PhD advisor and mentor. Besides my advisor**,** I express my gratitude to Dr. Sorina Popescu, Co-advisor, for her treasured support, help, and encouragement which influenced the continuation of my research. Special thanks go to my committee members and special thanks for their advice to me and all the time they invested on commenting and improving my thesis, Drs. Guihong Bi, Tonyin Li, Jiaxu Li. In addition, without the financial support of the Iraqi Ministry of Higher Education and Scientific Research,

which offered me a scholarship for postgraduate studies, this work would not have been possible. I am also thankful to the staff. Thanks also go to the soul of my beloved mother, my father, and soul of my husband, sisters, and brothers for helping me survive all the stress and not allowing me to give up. The best part of life is sharing it with the ones you love. Thank you to all. Most of all none of this would have been possible without the love and encouragement of my family and friends. I am eternally grateful. Your faith in me humbles and inspires me to achieve more than I ever dreamed I could, thank you.

Special thanks go to Mr. Bill Everett for being a friend in every way.

I thank my esteemed brother-in-law, Dr Walid Al-Shahib Al- Hilli for the continuous support and encouragement of my PhD studies. Very strong thanks to him for facilitating procedures for obtaining the opportunity for funding to undertake my studies.

My gratitude extends to the Faculty of Agriculture and life science for undertaking my studies at the Department of Plant and Soil Sciences, Mississippi State University. Special thanks go to Dr. Amelia Fox for her support and encouragement. My sincere thanks also go to Dr. Hassan, and Dr. Islam who gave access to their laboratory and research facilities. Special thanks go to my small family (Waqar and Rabia) for their support and friendship over the year. Special thanks go to my family here in America (Melissa, Jenny, Maggie, Lauren, Jerry, Jay, and Jacob).

Special thanks go to Dr. Maher Al- Asady from AL-Qasim Green University- Iraq for his support and encouragement.

Special thanks go to Dr. Abdel Monnim Saadallah from Technical Agriculture College-Mosul - Iraq. Special thanks go to Dr. Marilyn and my American friends (Anna, Mathew, Hanna, Sarah, Jo Beth, Ashley, Nathan, and their families).

Thank you for everyone.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

INTRODUCTION

Lavender is cultivated for its aromatic flowers in the around the world, especially in the United State, Bulgaria, France, Spain, Britain, Australia, and Russia where large quantities of this herb are grown annually (Verma et al, 2010). Lavender belongs to the Lamiaceae and contains more than 25 to 30 species of flowering plants native to the Mediterranean region south to tropical Africa and to many regions of Asia. They are shrubs or herbs, perennial, or biennial in habit. The genus includes annuals, herbaceous plants, subshrubs, and small shrubs with a wide diversity in growth characteristics, leaf size, flower color, physical appearance, and aroma (Piccaglia et al., 1993). Lavender, also called Luanda, is known, and loved for its aromatic, blue flower spikes which can be more than 30 to 60 cm long.

Lavender falls into four main classes: *Lavandula latifolia*, a lavender like Mediterranean kelp; *Lavandula angustifolia*, a plant with full blooms, commonly known as English lavender (previously known as *L. vera* or *L. officinalis*); *Lavandula stoechas*, which has butterfly-like bracts above the flowers and is known as French lavender; and *Lavandula* x *intermedia*, which is a sterile cross between *L. latifolia* and *L. angustifolia*. The various lavenders have similar ethnobotanical properties and major chemical constituents (Agricultural Research Service, 2000). Lavender essential oil is an important raw material for perfumery and cosmetics (Webb et al., 1984). Essential oils from *L. angustifolia* are among the best and most sought-after lavender oils in the

beauty and aromatherapy industries. However, these valuable oils are produced in relatively low quantities (Lis-Balchin, 2002).

Aromatic plants contain mixtures of volatile compounds called essential oils which are a small part of the plant's composition (Buchbauer et al., 1993; Asbahani et al., 2015). These compounds are used in food, cosmetic and pharmaceutical applications. The composition of essential oils is complex and most of these components include terpenes, mostly monoterpenes and sesquiterpenes. However, there are allyl and propenylphenol (phenylpropanoid) components of some essential oils (Zakaria,1991; Hay and Waterman, 1993; Bilia et al., 2014; Hay, 1998). Essential oils are composed mainly of terpenes and phenylpropenes with most essential oils containing predominantly (90%) terpenes. These terpenes contain mono terpenes with ten carbons, sesquiterpenes with fifteen carbons and diterpenes with twenty carbons (Hay and Waterman, 1993; Zakaria 1991). Terpenoids are the main ingredients found in Lavender (*Lavandula dentata*) and Nutmeg (*Myristica fragrans*) essential oils. These oils contain compounds containing antioxidants, antimicrobials, anti-inflammatories, anti-tumors, hypoglycemia, anticonvulsants, and pesticides (Aprotosoaie et al., 2017; Cossetin et al., 2018). Since the base oils are hydrophobic, this often limits production, viability and biological activity, and the low water solubility of these compounds shows high oil / water separation coefficients (Prakash et al., 2015).

Essential oils are found in different parts of the plant and when exposed to the air at normal temperatures they evaporate easily, so they are called volatile oils or etheric essential oils because they represent the "essence". (Tanker et al., 1990). Many essential oils contain powerful and active anti-microbial agents (Deans and Balchin, 1997). Some have pharmacological properties (Lis Balchin and Hart, 1999; 1995). Lavender has been very popular since ancient times being traced

back from the ancient Romans and Greeks to the present day as an aromatic plant as well as a medicinal agent. Lavender's importance was confirmed recently when it was named "The herb of the year 1999" by Herb Growing and Marketing Network in the United States (Evelegh, 1996; Anonymous, 1999). Lavender essential oil is extracted usually by steam distillation from both flower heads and foliage, but the chemical composition varies greatly with sweetest essential oil derived from flowers (McGimpsey and Porter, 1999).

The main chemical ingredients of lavender include linalool, linalyl, eucalyptol, geraniol, limonin, cineol, coumarins, flavonoids and triterpenoids (Prashar et al, 2004; Hajhashemi et al, 2003). Linalool and linalyl are the main components of lavender oil (Buchbauer et al., 1991). Linalool forms 70% of terpenoids of floral scents. Linalool also accounts for 60 - 90% of cosmetics as a component of the top perfumes (Cal and Krzyzaniak, 2006). Lavender is a perennial plant of high medicinal use in the European Pharmacopoeia (2008). Medicinal and aromatic plants are used by 80% of the world's population for therapeutic, pharmacological effects (WHO, 2008). The top 150 prescription drugs are based on natural sources derived from plants (74%), fungi (18%), bacteria (5%), and vertebrate species such as snakes and frogs (3%). Many of these plants manufacture many secondary metabolites, of which no less than 12,000 have been isolated which are beneficial for maintaining health in humans and animals, and most of which are phenols, alkaloids, glycosides, and saponins (Lai, 2004; Tapsell, 2006).

The essential oils in lavender depend on the original habitat and the environmental factors where grown as well as the stage of plant growth. The extraction method also affects the quantity of oil produced (Lakusić et al., 2014). *Lavandula angustifolia* essential oil contains linalool and linalyl acetate. Linalyl acetate is the main compound found in flowers. The plant also contains

rosmarinic acid and coumarin (Lis Balchin, 2002) as well as carvacrol (26.2%), limonene (19.6%), 1,8-cineole (11.8%), terpene-4-ol (7.6%), spathiolenol (4.9%), alpha pinene (4.2%), p-cimin (4.2%).), caryophyllene oxide (2.7%) and terpinolene (2.6%) (Bakhsha et al., 2014).

Lavender varieties are characterized by having the same ethnobotanical characteristics. French lavender (*Lavandula stoechas*) is characterized by the presence of bracts resembling butterflies at the top of the brightly colored flowers often used to treat headaches, while the English lavender (*Lavandula angustifolia*) is used as a diuretic. The species *L.* x *intermedia* is a sterile cross between *L. angustifolia* and *L. latifolia* (Agriculture Research Service, 2000). *Lavandula* x *intermedia* is characterized by high production of essential oils up to 120 kg per hectare. Other species and hybrids of lavender are not used in the perfume industry because they contain high amounts of camphor, but they are used for their antifungal and antiseptic properties. Essential oils are produced in the lavender plant from glands located on the surface of flowers and leaves (Lis-Balchin, 2002). Lavender has been an important source of medical materials since ancient times and a major source of many modern medicines. In its different growth stages, it builds up a range of metabolic materials developing terpenes, phenols, and alkaloids (secondary metabolites in the plant). These compounds are important for the survival of the plant in the natural environment. Some of the compounds constitute defensive mechanisms against attack from pathogens.

These secondary metabolites can be pharmaceutical substances, food flavors, pigments, fragrances, or pesticides (Tripathi and Tripathi, 2003). The process of detecting the presence of these compounds and extracting them to identify their precise chemical composition is a complex process and requires advanced equipment and techniques such as multiple column chromatography (CC), Mass spectroscopy (MS), Nuclear Magnetic Resonance (NMR), Gas

Chromatograph - Mass Spectrometry (GC – MS), MS - matrix-assisted laser desorption /ionization (MALDI), and high-performance liquid chromatography (HPLC) (Eja et al., 2011; Nascimento et al., 2000).

Lavender plants need a limited amount of fertilizer necessary for the development of flowers rich in oil (Lis-Balchin, 2002). Organic fertilizers improve biodiversity)Enwall et al, 2005), increase the abundance of soil microorganisms, and provide organic matter and micro-nutrients to organisms such as fungi (Pimentel et al, 2005), which help plants absorb nutrients (Mäder et al., 2002(. There is much research on medicinal and aromatic plants. Some of the research explains the positive effect of chemical fertilizers and organic fertilizers on growth, as well as the promotion of the content of active ingredients at certain rates of supplementation.

Recently, there has been much interest in using environmentally friendly stimuli in promoting plant growth (Zhu, 2001; Akula and Ravishankar, 2011; Gupta et al., 2015). It has been shown that the use of plant growth regulators (plant hormones) is an advanced way to nourish plants and increase their production due to the important role these hormones play in plant growth (Forde and Lorenzo, 2001; Meixner et al., 2005). These hormones are organic materials mainly produced in active plant tissues and they have a great influence on some physiological processes. This is because plant hormones move from their sites of production to sites of influence (Weyers and Paterson, 2001; Davies, 2010; Wani et al., 2016). Plant hormones play a great role in the growth and development of various plant structures. In addition to its effect on metabolic processes, it also affects many specialized physiological processes (Davies, 2010; Keurentjes et al., 2011). The application of gibberellic acid affects plant growth and development by increasing plant height, number of leaves and number of branches (Roy et al., 2010; Qureshi et al., 2013).

Research has shown that GA_3 promotes growth and photosynthesis (Rood et al., 1990; Khan and Samiullah, 2003).

There has been great interest in using natural organic matter and substances, such as humic acid, that act as powerful fertilizers. Humic acid has hormone-like activity that stimulates plant growth and development (Ferrara and Brunetti, 2010). It has been observed that humic substances have a positive effect on vegetative properties, root, leaf and vegetative growth, and germination of different crop species (Piccolo et al., 1993). The chemical composition of plants, such as bean, grape, and tomato, was also improved significantly by using humic substances (Ferrara and Brunetti, 2010; Abbas, 2013). Humic acid is the main ingredient for humic substances, which are the main organic matter (humus). Humic substances have many beneficial effects on the physical composition of the soil as well as in stimulating plant and cell growth, nutrient absorption, and yield increase (Cimrin and Yilmaz, 2005; Asik et al., 2009). Foliar spray of humic acid works to increase photosynthesis and antioxidant metabolism (Fujiu et al., 1995).

Iron (Fe) plays a major role in the regulation of many cellular processes, including chlorophyll biosynthesis, photosynthesis, and mitochondrial respiration and is an essential nutrient for plants (Ghasemi et al., 2014).

There has been increased interest in beneficial soil microbes including arbuscular mycorrhizal (AMF) fungi. Notably, under natural conditions, plants often interact with microbes, with which some micro-plant interactions mitigate stress-related damage and improve plant tolerance to stressful conditions (Ngumbi and Kloepper, 2016). Arbuscular mycorrhizal fungi (AMF) are soil microorganisms which can form a symbiotic bond with terrestrial plants. These beneficial microbes offer many benefits to the host plants (Bonfante and Genre, 2010). During

mycorrhization, besides improving plant nutritional status, AMF can enhance plant performance to withstand many environmental stresses (Balestrini and Lumini, 2018). Previous studies showed the great contributions of the coexistence of AMF (Arbuscular mycorrhizal fungi) including various innate benefits such as nutrient absorption, modifications in host physiology, photosynthesis, plant hormones, and more efficient antioxidant systems (Ruiz et al., 2015; Duc et al., 2018).

Mycorrhizae also play an important role in enhancing the survival and development rate of seedlings, standardizing crops, and reproductive capacity (Lu and Koide., 1994). Mycorrhizae can reduce inputs of P and N fertilizers and increase environmental stress tolerance (Birhane et al., 2012; Duc and Posta, 2018). Trichoderma is a mycorrhizal biological agent used to stimulate plant growth (Oliveira et al., 2016, Roese et al., 2017). Growth-promoting mechanisms in plants are related to microorganisms through metabolism, plant hormone production or nitrogen fixation (Carvalho et al., 2011; Péres-Montaño et al., 2014). Trichoderma spp. are widely used as biological control agents (BCAs) due to their ability to reduce the incidence of fungal diseases (Cooney and Lauren, 1998).

This study explores the use of gibberellic acid, iron chelate, humic acid, and mycorrhiza to enhance essential oil production of lavender in cultivation.

Problem Statement

With over 45 species in the *Lavandula* (lavender) genus and over 450 named varieties or cultivars grown as annuals, short-lived perennials, or perennials, it may be a challenge to identify specific species with the highest production of essential oils. There are three species which predominate at plant nurseries in the United States: *L. angustifolia*, *L.* x *intermedia*, and *L.*

stoechas. Each species has its own unique traits and not all lavender is created equal. In addition, there is insufficient published research on biological fertilizers in the production of lavender and the effects on plant growth and yield of essential oils.

Justification of the study

One of the essential oil components of lavender, eucalyptol, has been shown to significantly improve memory and performance on cognitive tasks. Lavender is considered an adaptogen helping the body deal with stress or imbalances and helping promote calmness and wellness (Cavanagh and Wilkinson, 2005). With increased demand for use in natural remedies, perfumes, food items and traditional herbal medicines, there is a need for increasing essential oil yield in lavender production.

Research Objectives

The general objective of this research was to investigate the effects of production practices on essential oil yield in three lavender species: *L. angustifulia*, *L.* x *intermedia*, and *L. stoechas*. Specifically, the objectives were to 1) investigate the effects of gibberellic acid, iron chelate, humic acid, mycorrhiza, Trichoderma, diammonium phosphate (DAP), and potash on plant growth and essential oil yield and 2) determine the essential oils chemical composition for three lavender species using GC-MS, and HPLC.

Significance of the study

This study yielded data and information useful in producing essential oil of lavender. The study findings and recommendations will be beneficial to lavender growers, herbalists, and vendors of essential oils. This study aims to rely on research data about how essential oils are produced and extracted.

Scope and limitations of the study

This study focused on three species of lavender plants and the research was conducted in the Department of Plant and Soil Sciences greenhouses on the Mississippi State University campus. This study investigated the species' richness, characteristics, abundance of essential oils, and the influence of production factors on production of essential oils. Of over 45 species of lavender, only three species were investigated due to available time and resources.

CHAPTER II

LITERATURE REVIEW

General Characteristics, Classification and Distribution of lavender

The lavender plant (*Lavandula* sp.) is a medicinal herb consumed by people around the world since antiquity. Today it is regarded as one of the most important aromatic herbs **(**Barata et al., 2011; Duke, 1985). The Latin name *Lavandula* is from the ancient use of this plant to scent shower water, and it is derived from the Latin word *lavare* which means to be washed (Lis-Balchin, 2002). Lavender, also known as medicinal lavender, true lavender, or common lavender (*Lavandula angustifolia, L. officinalis, L. vera*), is an evergreen perennial plant. It is native to the Mediterranean region (France, Spain, Andorra, and Italy).

Lavender is a perennial shrub plant of the mint family (Lamiaceae). The genus *Lavandula* includes more than 30 species. The three primary commercial species are lavender (*Lavandula angustifolia* mill), lavender spike (*Lavandula latifolia* L.) and lavandin - a sterile hybrid developed by crossbreeding *L. angustifolia* × *L. latifolia*. Lavender plants can be propagated from seeds, from cuttings, or through tissue culture. Lavender in production is pruned regularly to stimulate plant growth and promote flowering. The flowering period lasts from July to August. Harvesting should take place on dry, sunny days. The flowers are collected before opening and are dried in bundles in shaded, well-ventilated places. There are two types of products harvested, flowers (*Flos Lavandulae*) or the aerial parts (*Herba Lavandulae*) of the flowering plant (Góra and Lis, 2005). Lavender is cultivated across the world with Bulgaria, France, United Kingdom,

China, and Spain the major producers of lavender oil. It is also cultivated in other countries, including Poland (Boelens, 1995; Śmigielski et al., 2009). Lavandin plants are mainly grown in France which produces 90% of the world's production of lavender oil. Lavender is the primary species grown for essential oil in Spain.

Lavender has been cultivated on a large scale since ancient times. The increased cultivation of lavender and the production of essential oils in Europe during the first half of the last century stimulated the improvement of lavender in France and the Soviet Union (Russia, Ukraine, and Moldova). Improvement programs have focused on a wide range of lavender properties, including flower yield, essential oils compounds, quality, and content of linalyl acetate and linalool (Romanenko, 1973; Singh et al., 1989).

Lavender has been used as a therapeutic, essential oil for centuries. For medicinal products it is stated the extract of the flowers or the leaves, the essential oil, has curative properties, including antiseptic, anti-inflammatory, anti-fungal, analgesic, cleansing, balance, and calming affects (Kasper et al., 2010; Woelk and Schlafke 2010). There is evidence *Lavandula angustifolia* extract protects neurons from glutamate toxicity (Büyükokuroğlu et al., 2003; Kaka et al. 2016). Recent studies have proven *L. angustifolia* extract works as an improvement in treating spinal cord injury by reducing nerve function damage (Kaka et al. 2016; Yaghoobi et al., 2016).

Lavender oil is of higher quality than lavandin oil. Lavebder 0-0.6% camphor *L. angustifolia* while lavandin 6-10% camphor *L. intermedia*. Both lavender and lavandin essential oils are widely used in various industrial products including perfumes, pharmaceuticals, cosmetics, personal care, and home maintenance products (Cavanagh and Wilkinson, 2005; Lesage-Meessen et al., 2015). Additionally, lavender oil has been widely used in aromatherapy

and integrative medicine. It has a positive effect in treating anxiety and insomnia (Kashani et al., 2011; Sienkiewicz et al., 2011; Soltani. et al., 2013).

Lavandula cultivars in America

This study focused on three species of lavender: *L. angustifulia*, *L.* x *intermedia*, and *L. stoechas*. These species differ in terms of morphology, habitat, and chemical composition (Lesage-Meessen et al., 2015). Because of their essential oils (EOs) content they have been cultivated in various regions of the world as in Europe, Canary Islands, Madeira, North Africa, Southwest Asia, the Arabian Peninsula, India and North and South America (Lis-Balchin 2002; Benabdelkader et al., 2015). Lavender is grown in many of the United States of America, including Washington, which contains large lavender farms around Sequim, WA.

Lavender farms around Sequim provide great support for the local economy and form a part of the local tourist industry. There are lavender festivals, tours featuring lavender farms and a lavender street fair showcasing lavender products and services from the Sequim-Dungeness area, as well as regional garden products and natural crafts. There is an outdoor market, lavender crafting demonstrations, food, and aromatherapy. Olympic Lavender Farm, located near Scheme, WA, has 1,200 lavender plants on a five-acre area and provides a viewing tour of lavender oil distillation. The processes of developing the essential oils industry began in the United States of America about 1985 with the oils being used in perfumery and aromatherapy (Adam, 2006).

Foliar Nutrition

Foliar feeding means spraying nutrients on the vegetative or foliar portions of plants. It can result in higher returns as well as higher quality products. Foliar fertilizers allow direct nutrient

supply to the leaves within the period needed (Fageria et al., 2009; Kostadinov and Kostadinova, 2014). It is a quick response feeding system to meet the requirements of the plant because transporting nutrients through the roots requires a long time. The plant nutrition influences many physiological and biochemical processes that affect growth, development, and yield (Stojanova et al., 2016).

Some researchers, such as Bozorgi (2012), Sadeghzade et al. (2012) and Saykhul et al. (2014), consider foliar feeding more efficient than ground feeding if applied according to crop requirements. It is not a substitute for root absorption as it is somewhat specialized and requires little energy to transport the elements to the action centers such as chlorophyll. It is a good way to supply plants with nutrients, especially micro-nutrients, to meet their needs faster compared to soil applied fertilization. This is especially true when used in accordance with the requirements of the crop, considering the nature of the fertilizer, the concentration of the active ingredient, the number of applications, the time of application, and the type of crop. The absorption of nutrients through the leaves occurs in two ways:

1-Apoplast: Through stomata and interstitial spaces between the leaf cells until they reach the transporting vessels and then to other parts of the plant.

2- Symplast: Through cytoplasmic bridges or tubes located under the cuticle layer of the epidermal cells of the leaves and then through the cytoplasm and from it to other parts of the plant (Fernandez et al., 2013; Buckley, 2015).

There are many factors affecting the absorption of nutrients through the leaves that must be considered such as the type of plant, the thickness of the cuticle layer and the surface area of the leaves. The nutritional status of the plant, the nature of the nutrient in the solution, and its

concentration will also affect efficacy of foliar applications of nutrients. In addition to the environmental factors surrounding plant growth, the rate of absorption of nutrients is affected by leaf age and physiological state as the penetration of ions is faster in new leaves compared to mature leaves which have a thick cuticle layer (Wojcik, 2004; Li et al., 2017).

The addition of nutrients to the plant by foliar spray can be very effective under certain conditions, some of which are related to the soil. There are many reasons preventing the plant being provided with the nutrients it needs. Some are related to the plant as some plants have a weak root system or sparse branching limited to the surface layer of the soil. This does not enable the plant to absorb nutrients in the amounts it needs which allows foliar feeding to provide these nutrients to the plant (El-Fouly et al., 1995). It appears the importance of foliar feeding emerged from the possibility of mixing fertilizers with pesticides, which provides an opportunity to reduce the energy consumption required for the transport of elemental ions within the plant (Akanbi et al., 2007). This secures the plant's nutrient needs during the critical and sensitive stages of its growth that the roots are unable to provide (Khan et al., 2009).

Choosing the right time for foliar feeding has a major role in increasing spraying efficiency. It is important when spraying to avoid times when evaporation is high so the leaf can absorb the largest possible amount of solution from its surface. The more time the nutrients remain in a solution on the surface of the leaf the greater the absorption of the nutrients (Raafat and Tharwat, 2011). Evening and early morning are also good times for foliar fertilization due to low temperatures (Saeed et al., 2012).

Gibberellic Acid (GA_3)

Gibberellic acid is a plant hormone that stimulates growth through cell enlargement. Gibberellins are naturally found in all plant kingdoms with growing peaks, fresh leaves, fruit, and newly grown seed embryos primary sources of these compounds (Taiz and Zager, 2010). The response of different plants to gibberellins varies according to the types of plants, their stage of growth, the amount and quality of internally formed gibberellin, the length of the light period to which the plant is exposed, and other factors (Kariali and Mohapatra, 2007). Gibberellins affect the promotion of stem elongation by stimulating cell elongation, overcoming genetic dwarfism, and encouraging cell division and expansion, as young cells respond by dividing, while older cells respond only with expansion (Shani et al., 2013). Gibberellins work to regulate the permeability of cellular membranes and encourage plant transition from vegetative to the flowering stage. Gibberellins also regulate plant growth and development in response to environmental conditions by modifying the production, distribution, or transmission of the signal from these hormones (Colebrook et al., 2014). The physiological effect of gibberellin is due to its control of enzymatic activity and its activation of metabolism processes (AL-Rumaih, 2007).

Gibberellins activate the formation of nucleic acids and treatment with gibberellin further directs nutrients toward growth sites (Iqbal et al., 2011). The plant's content of gibberellins is usually associated with the growth and development of the plant. Roots also produce types of gibberellins that travel to the stems and the rest of the plant. The movement of the gibberellins is free within the plant as it moves down and to the top of the plant without hindrance (Tanimoto, 2012). The biosynthesis of gibberellins inside a plant can modify the plant's tolerance to the surrounding environmental conditions (Iqbal and Ashraf, 2013). The use of plant growth regulators can improve growth, yield, and flower quality (Nuvale et al., 2010). Plant growth

regulators are of two types: Biotic inhibitors such as methylamine and (ABA) and inducers such as gibberellins, auxins, and cytokinin's (Giannakoula et al., 2012). Growth regulators can control biochemical and physiological processes in plants such as controlling the chemical composition of plants, dormancy, amount of mineral absorption from soil, flowering, fruit set, and crop development (Sarkar et al., 2002). Application of growth regulator substances to regulate endogenous hormones and inhibit or stimulate flowering depends on the concentration and time of application (Shakarami et al., 2013).

The phytohormone gibberellic acid regulates nearly all plant development and growth processes, including seed growth, germination, stem and root growth, cell division, and flowering time. GA, when first discovered, was named after the fungus *Gibberella fujikuroi* that causes uncontrolled cell elongation and various pathological symptoms in rice (Davière and Achard, 2013). GA has essential functions in, pollen formation, and pollen tube development (Kwon et al., 2015). Gibberellic acid has many functions, such as promoting seed germination and seed development, promoting leaf and flower growth, promoting stem elongation, increasing pollen production, tolerating salinity, preventing chlorophyll breakdown, increasing plant life, and preventing plant aging (Rosenvasser et al., 2006; Rieu et al., 2008; Tuna et al., 2008).

Gibberellin, as a phytohormone, can modify the growth of plants throughout their life cycle. This is achieved through the signaling and biosynthesis pathway (Fleet and Sun, 2005). It is noteworthy that this high complementarity of signaling pathways is derived from the combined action of gibberellins and other plant hormones and other regulatory factors (Bottini et al., 2004). The function of gibberellic acid signaling is de-suppression, being responsible for

transcription regulators known as DELLA domain proteins that inhibit GA responses (Fleet and Sun, 2005). Basically, the activation or suppression of processes associated with GA signaling (growth and development) depends on the absence or presence of DELLA protein inhibitors (Schwechheimer and Willige, 2009).

Effect of gibberellin on the characteristics of growth and essential oil:

The effect of GA on plants has been demonstrated by many researchers (Brian and Hemming, 1955; Marth et al., 1956; Chrispeels and Varner, 1967; Roy et al., 2010; Saptari and Dewi, 2013). Gibberellins primarily affect cell elongation, cell division, or both. Gibberellin affects the plant life cycle, such as germination, seed set, flowering, leaf stretching, and fruit development (Phillips et al., 1998; Gomi and Matsuoka, 2003). It has been reported the application of GA_3 is an important growth-stimulant for *L. angustifolia* with GA treatments increasing growth, rapid fruiting, and plant improvement (Heller et al., 2006).

Al-Rawi et al. (2016) concluded that foliar spraying of gibberellic acid at a concentration of 100 mg L^{-1} on peach trees (*Prunus persica L.*) increased the leafy area of the plant and the content of the chlorophyll and carbohydrates, as well as increasing nitrogen and zinc. Sardoei and Shahdadneghad, (2014) found the use of gibberellin at 250 mg L^{-1} significantly increased leaf chlorophyll content in calendula (*Calendula officinalis* L.). Singh et al. (2017a) found a significant increase in chlorophyll in *Capsicum annum* (L.) 'Indra' when using 30 mg L^{-1} GA. In a study by Neware et al. (2017) on the orange plant (*Citrus sinensis L.* Osbeck), the treatment of the plant with GA at 20 mg L^{-1} resulted in a significant increase in leaf chlorophyll content.

Gibberellins are involved in the growth of buds and meristems. They often work to break bud dormancy (Machado et al., 2011). Gibberellins affect many development processes and control various aspects of seed germination, including dormancy, moreover, GAs influence the

transition from juvenile to maturity stage and flowering stimulation (Taiz and Zeiger, 2004). Plant hormones are modified and can increase in response to environmental factors including photosynthesis and biotic stresses. In fact, due to some environmental changes, hormone-like messages are generated, and these hormonal messages are recycled between the roots and buds as the roots import hormones and the buds act as active hormonal pools (Jackson, 1993).

Gibberellins have been shown to affect oil content in plants. Sofy et al. (2016) in their study on *Chenopodium quinoa* showed a significant increase in the percentage of fats in seeds when using gibberellin at 50 mg L^{-1}. Selim et al. (2017) in their study on *Polianthes tuberosa* 'Double' found the use of GA at 150 mg L^{-1} caused a significant increase in plant height and an increase in the percentages of oil components. Chetouani et al. (2017) showed the most significant effects were obtained at 50 ppm and 1000 ppm of GA for each of *Thymus satureioides* and *Lavandula dentata*. In addition, GA_3 foliar spray application increased height and oil production in *Mentha arvensis* L. (Bose, 2013). In research on lavender grown in the greenhouse, Hassanpouraghdam et al. (2011) assessed the effects of applying foliar GA_3 on growth characteristics, essential oil content, yield, and some macronutrients content of French lavender (*Lavandula officinalis* Chaix.). The greatest amounts of lavender essential oil content and yield were obtained from plants treated with 300 mg L^{-1} GA_3 with concentrations of more than 300 mg L^{-1} GA_3 likely to be the preferred foliar application level to enhance the growth and production of the essential oil from lavender. Hajisamadi et al. (2011) investigated the influence of foliar applied GA_3 on growth, essential oil content, and lavender yield. The highest content and yield of essential oils for both offshoots and flowers was obtained using 300 mg L^{-1} GA_3. Fresh and dry weight of leaves and stems were highest when 300 mg L^{-1} GA_3 was applied. Application of GA_3 at 300 mg L^{-1} significantly increased flower numbers. As with other traits, this concentration also achieved maximum plant height. GA_3 at 300

mg L^{-1} was the most effective foliar spray treatment for lavender plants in terms of growth characteristics, essential oil content and yield.

The importance of iron for the plant

Plants need iron in small quantities, and it is one of 16 essential elements for plant growth and reproduction. Most plants need about 100-500 mg iron Kg^{-1} leaf dry weight. Iron toxicity appears when it is more than 500 Mg Kg^{-1} dry weight. Iron deficiency appears when its content in the leaves is about 50-100 mg Kg^{-1} dry weight (Barker and Stratton, 2015). Iron is found in the soil in several forms, and the most abundant form of iron is Fe_2O_3 or hematite, which is insoluble and gives a red color to the soil (Journet et al., 2014). Iron influences increasing the yield and quality of various agricultural crops (Imtiaz et al., 2010). Among the most important symptoms of iron deficiency are yellowing of newly formed leaves and the edges of leaves turning brown and, in severe cases, burn the entire leaf. The appearance of symptoms on new leaves is due to the difficulty of the transfer of the element in the plant. The yellowing of leaves is due to the breakdown and loss of chlorophyll (Taiz and Zeiger, 2010).

Iron is important in the plant in several ways:

- It is included in the synthesis of some enzymes such as catalase and peroxidase (as it is found in the porphyrin ring complex), as the catalase enzyme stimulates hydrogen peroxide hydrolysis into water and oxygen, and peroxidase is a family of enzymes that lead to metabolism of reactive oxygen species.
- It is part of the structure of Ferredoxin which acts as an electron transporter in the processes of photosynthesis, nitrate reduction, sulfite reduction and nitrogen fixation.
- It is included in the composition of Cytochromes which act as the oxygen transporter for photosynthesis.
- It is included in leghemoglobin synthesis and works as an oxygen carrier in nitrogen fixation, and iron is an important part of the nitrogenase enzyme that affects nitrogen fixation in nitrogen fixing plants.

- Iron is the electron donor to the NADPH energy compound in the first photovoltaic system (Barker and Stratton, 2015).

Effect Iron Chelate Fertilizer on Essential Oils

Chelated iron fertilizer is a plant source rich in divalent iron and is effective because of its high stability and gradual release of iron in a wide range of pH (3-11). The advantage of chelating iron is the increase in the ratio of ferrous iron ion to ferric iron ion in the chelating surface which leads to increased chlorophyll production in the plant (Roosta et al., 2015).

Iron chelate has been shown to increase the yield and quality of lavender essential oils (Badri et al, 2015). In addition to the elements zinc and manganese (Kaviani et al., 2014), iron helps to increase photosynthesis, thereby increasing the production of active substances in the plant (Singh et al., 2016). Farahani et al (2015), indicated the use of nano iron fertilizer increased flower number and wet flower yield of *Crocus sativus* by 10 kg. But the application of 5 kg nano-chelate increased dry stigma and dry leaf yield, leaf iron and total iron concentrations. Another study reported that the effect of the significant interaction between the time of iron spraying and concentration on the flower yield of pot marigold, the highest flower yield of pot marigold *(Calendula officinalis)* was obtained at the first, second and third harvest. The highest yield of essential oil (2.397 kg / ha) was obtained with application of humic acid (Amuamuha et al., 2012). Abdel-Wahab (2008) reported that micronutrients, such as iron, play important roles in plants. Growth and yield of aromatic and medicinal plants, such as *Mentha pipperita*, have apocrine glands in the leaves containing the essential oils. Oil content was increased in the plant due to the addition of nutrients. Nasiri et al. (2010) show the percentage of essential oils, the essential oil yield and the flower yield of *M. chamomile* increased by

adding iron. Misra et al. (2006) declared that the biosynthesis of essential oils is strongly influenced by iron in basil plants.

Effect of humic acid on the growth and production of essential oils:

Organic acids, including humic acid, Fulvic acid, and humin play an active role in plant growth and plant nutrient uptake. Seen and Kingman (1998) explained the acid enters the plant to increase the vitality of the plant as a complementary source of phenol which acts as a chemical intermediary increasing enzymatic efficacy and increasing the division of cells and development of the root system thus increasing the production of dry matter. Mataroiev (2002) also pointed out that humic acid has an effective role in improving soil and chemical properties through its interaction with soil minerals and its effect on improving the air and water properties of soil while increasing the absorption capacity of nutrients. Humic acid inhibits IAA Oxidase activity which stimulates IAA activity. IAA plays an important role in stimulating plant and root growth (Wandruszka et al., 1999).

Humic acids improve the capacity of nutrient retention in the soil by its association with sodium, which helps the plant tolerate high concentrations of this element and protects against toxicity and osmotic problems (Stevenson,1994). Piccolo et al. (1993) also found alumic acid improves plant growth and increases its resistance to stress conditions by acting as a regulator of the concentration of plant hormones within the plant. Humic acid has a direct effect through the development of roots and nutrient intake by the plant and an indirect effect by increasing the absorption of both water and nutrients by plant roots (Lobartini et al., 1997). Hunic acid it increases the nutrient uptake by the plant and promotes absorption of calcium, potassium, phosphorus, and magnesium from the soil. It also plays an important role in minimizing the

harmful effect of adding foliar fertilizers to the soil (Hartwigson and Vans, 2000; Turkman et al., 2004).

The high acidity of humic acid increases the efficiency of the soil to retain ketones and reduce the loss of nitrogen (Tan, 2003). Adding humic acid to the soil and plants leads to an increase in the percentage of nutrients absorbed by the decomposition of organic matter, which in turn affects the acidic nature of the soil (pH) (Harpe et al., 2000). The addition of humic acid activates the growth of the plant by increasing the permeability of the cellular wall and thus increases the absorption of water, nutrients, and oxygen, especially in the roots and increases the expansion of stem cells and their division and thus increases the activity of photosynthesis and absorption of phosphorus (Faust, 1998). Humic acid has an important role in stimulating the activity of microorganisms in soil improving the soil and increasing the efficiency of roots in the absorption of water and nutrients (Phelpstek, 2002). The addition of humic acid to the soil helps to reduce the loss of nutrients and leaching in the soil as it acts as a claw compound to reduce loss (Leonardo, 2008). Badri et al. (2015) indicated humic acid concentrations of 2500 mg resulted in the highest shoot dry weight and improvement in plant height, dry weight, plant fresh weight, number of leaves and increased lavender essential oils yield and quality.

Humic acid increases biomass and essential oil production. Concentrations of humic acid at 300 mL L^{-1} resulted in greater production of biomass and essential oils. Hyssop (*Hyssopus officinalis*) produced greater biomass and oil with 300 ml L^{-1} humic acid in the second year of the experiment compared to the first year (Khazaie et al., 2011). Humic acid is a natural, bioavailable organic matte.r It has a great effect on plant growth and quality. However, the enhanced effect of humic acid was largely on the volatile oils in thyme (Noroozisharaf and

Kaviani, 2018). Humic acid has been widely considered a natural chelate for cationic micronutrients (Varanini and Pinton, 1995). Sanchez-Sanchez et al. (2006) reported the addition of humic acid might ameliorate Fe uptake by chelating free Fe and so hamper its immobilization. Humic acid is a stimulant of bacterial activity. This biologically increased activity enhanced P and total N content (Busato et al., 2012). Different levels of humic acid formulation significantly increased all quantitative and qualitative characteristics of Rosemary (Jalayerinia et al., 2017). In a study conducted by on potato plants, spraying with humic acid increasing the growth of shoots, increasing photosynthesis and leaf area activity (Ghorbani et al., 2010).

Biological Fertilizers

The use of organic and bio fertilizers has a long history in agriculture and specifically sustainable and organic agriculture (Gryndler et al., 2006). Organic and Biodynamic agriculture has spread rapidly around the world helped by the rapid demand for organic farming products (Yasseen, 2002; Abd El-Malek, 2005). Biofertilizers are produced after isolating fungi or bacteria from soil. The type of microorganism is dependent on the purpose for which it is propagated. Then it is developed in an appropriate environment under sterile conditions. It is then joined to suitable special carriers to prevent contamination with other organisms that may be a competition or antagonist to the original organism (Alexander, 1971). The carriers are usually various organic materials (Lucy et al., 2004). The fertilizer is kept under appropriate conditions until it is used as a soil inoculant. It is added to the soil in various ways, either near the roots of plants, directly to the permanent field, by fogging the roots of the seedlings before transplanting them, or by mixing the seeds with them before planting (Lakshmana, 2000).

Vermicompost has been considered a biological fertilizer. Microbiologically, vermicompost is an organic compound rich in macronutrients and microelements also produced by the interaction of earthworms and microorganisms that decompose organic matter (Yanga et al, 2015). Fernandez et al. (2009), also mentioned that using vermicompost in sustainable agriculture helps improve soil porosity and availability of nutrients. Different microorganisms release acids such as oxalic acid and humic acid, which helps to increase the solubility of nutrients in the soil (Adak et al., 2014). In this line, Blaize et al. (2005), mentioned that animal fertilizers are also considered an organic source of nutrients for sustainable plant production which contributes to an increase in soil organic matter, thus increasing the percentage of seed germination and the growth and development of roots and stems.

Effect of Mycorrhizae fungus on the growth and essential oil

The symbiotic association between fungi and plant roots is called mycorrhizae, which is a compound word of two words: Mycoss, which in Greek means fungus, and rhizae, which means microscopic plant roots (Hamel et al., 1991). Mycorrhizae work to improve the properties and composition of soils by causing physical, chemical, and biological changes by secreting glomalin, a protein compound that binds soil molecules between them (Wright and Upadhyaya 1996). Mycorrhizae have a positive role in several physiological processes of plants, especially those related to plant nutrition (Gianinazzi, 1983). Mycorrhizal fungi may have a positive effect on the structure of plant and bacterial community environmental stability (Diagne et al., 2020). Arbuscular mycorrhizal fungal (AMF) symbiosis is known as bio enhancers of plants. These root colonizers influence the growth and productivity of plants (Habibzadeh et al., 2013; Begum et al., 2019). Baslam and Goicoechea (2012(indicated mycorrhiza improves photosynthesis efficiency, strengthening and regulating the antioxidant system and plant osmotic balance. The

mycorrhiza helps plants absorb nutrients. These processes increase growth and productivity under stress conditions (Abdul Latif and Miransari 2014; Habeeb et al., 2020). Plant responses to fungal symbiosis differ depending on the species of plants and the type of fungi (Murphy et al., 2015; Zhang et al., 2013).

Mycorrhizal fungi are divided into two types, external and internal fungi, the last one is considered the most important in plant growth as they form a symbiotic relationship with most plant species (i.e., about 80% of the plant species). These fungi grow in live media and do not have the ability to grow in artificial media. Mycorrhizae are found in the soil, but they are in the form of dormant spores or live inside the roots of dead plants before entering any symbiotic relationship with plants as these parts tolerate unfavorable weather conditions such as drought and high temperatures (Orcutt and Nilsen, 2000). However, when the appropriate moisture is available, the process of coexistence between plants and fungi begins with the hydration phase. Then, the activity of enzymes and metabolic activities that play a role in the formation of the germ tube increases. As this tube grows to connect to the root of the plant host, it secretes the chemical compounds that make contact between the hypha that was formed from the germ tube and the root surface by a structure called Apperssorenum. New structures are then formed that transfer nutrients from the soil to the plant (Fig.2.1). Mycorrhizae have a positive role in several phylogenetic processes of the plant as it enables plants to absorb phosphorous many times greater than plants not inoculated (Gianinazzi, and Gianinazzi,1983). There is sharing of the nutritional benefits between the symbiotic associates (Chen et al.,2018).

In mycorrhizal plants the nutrition is transferred to the leaf of lavender much more than in non-mycorrhizal associated plants (Armada and Azcon, 2014). In dry soil, plants depend on

mycorrhizal activity to increase nutrients and absorb water. Lavender, rosemary, and thyme were inoculated with fungi forming successful colonization (Pirzad and Muhammadzadeh, 2018). Ouahmane et al. (2006), indicated mycorrhizal inoculation with *Glomus intraradices* had a significant effect on increases in root growth and shoots of *Lavandula* species. While mycorrhizal dependency of *L. stoechas* was much less than that recorded for other *Lavandula* species. Differences in mycorrhizal colonization has been tested among Lavandula species. Binet et al. (2020), reported that in lavender, the mycorrhizal root colonization was significantly higher in the healthy plant compared to the diseased plant. Azcón, and Barea, (1997) reported lavender response to acid phosphatase activity. However, mycorrhizal inoculation largely improved P, N, and K uptake thereby restoring the biochemical cycling of plant nutrients. There are several factors which may explain the implementation of symbiosis between AMF and host plants such as the root exudates (Hugoni et al., 2018). The mycobacterial associations communities' range along a continuum of parasitism to mutualism (Johnson et al., 1997). Li et al. (2013) indicated AMF regulation of plant aquaporin genes helps improves plant water status and drought tolerance. Mycorrhiza caused Fe chelating and increased its uptake and mobilization in peanuts and sorghum by exuding different kinds of siderophores (Caris et al., 1998).

In a study to investigate the effect of adding three types of mycorrhizae fungi on the growth and yield of potatoes, *Glomus mossea* led to an increase in plant height, fresh and dry weight of the shoots, and an increase in the number of tubers with an increase in the absorption of nitrogen and phosphorous (Gong et al., 2001). In a study to determine the effect of AMF on pollinating tomato plants , it was found that fungi had a significant effect in increasing the carbohydrate content of leaves, total yield, fruit weight, and fruit size (Abdel Latef and Hamed, 2010). Most importantly, Svenningsen et al. (2018) demonstrated suppressive activity incurred

upon AMF by the surrounding microbiota. AMF are well known to colonize many plants. The activity mycorrhiza induces in plants is expected to be under the control of plant hormones, and these hormones may be jasmonic acid (JA), salicylic acid (SA), ethylene and abscisic acid (ABA) which also play an important role as signal compounds in biotic or abiotic reactions (Nair et al., 2015; Pozo et al., 2015). Herrera et al. (2008), also indicated that AMF colonization in tomato is strongly controlled by the JA signaling pathway. Moreover, JA has been shown to contribute to the susceptibility of tomato plants to infection with AMF and play a regulatory role in the development of mycorrhizal colonization (Foo et al., 2013).

Mycorrhizal fungi were investigated for their role in the production of auxin-, gibberellin- and cytokinin-like substances that stimulated the growth of plants (Barea and Azcon-Aguilar, 1982). Moreover, higher concentrations of chlorophyll have been indicated in AMF-associated plants with increased chlorophyll level helping increase photosynthetic rates (Mathur and Vyas, 1995). The specialized microorganism competes with symbionts to colonize the root surface due to its inability to reproduce significantly in the soil, its dependence on sugars, amino acids, and organic acids, released by the plant and which are short-lived (Garbaye, 1991) (Fig2.1).

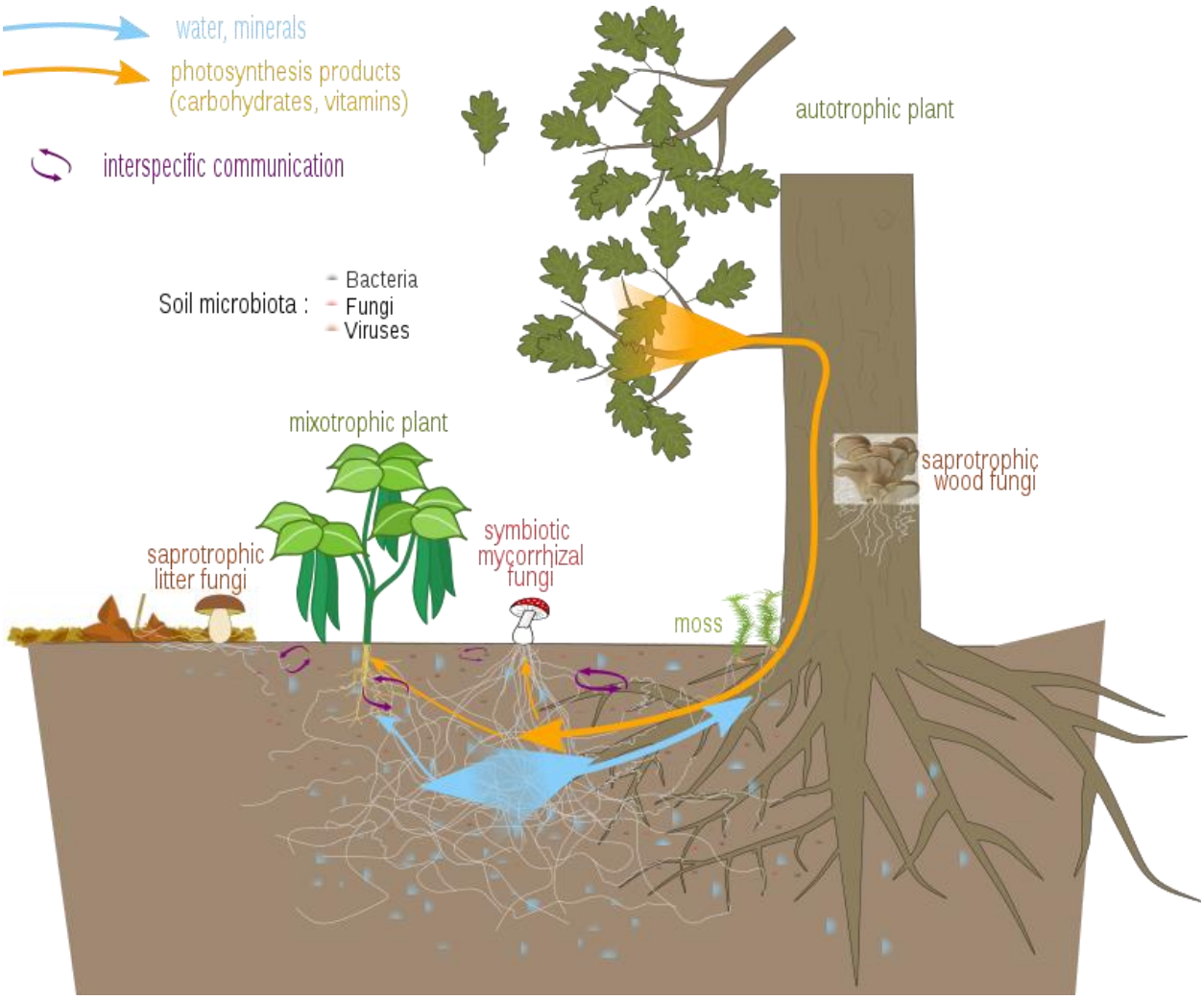

Figure 2.1 Nutrient exchanges and communication between a mycorrhizal fungus and plants. Within mutualistic mycorrhiza, the plant gives carbohydrates (products of photosynthesis) to the fungus, while the fungus gives the plant water and minerals in exchange, (https://en.wikipedia.org/wiki/Mycorrhiza).

Effect of Trichoderma harzianum on growth and yield of essential oils of lavender.

The use of fungal species against plant pathogens has received a great deal of interest from researchers worldwide as potential biocontrol agents in many crops, and among the most well-studied fungal genera is *Trichoderma* (syn. *Hypocrea*) (Yacoub et al., 2017). Qi and Zhao (2013) indicated that use of *Trichoderma* spp. is considered a new technology that has received

great attention as it plays a pivotal role in helping to improve the growth and development of plants. *Trichoderma* is a genus of fungi in the Hypocreaceae family found in all types of soils where it is considered the most widespread cultured fungus (Harman et al., 2004). This genus can form mutual internal relationships with many plant species (Bae et al., 2011). *Trichoderma* are beneficial microorganisms associated with plant roots and can enhance the absorption and use of nutrients promoting plant growth and stimulate plant defenses and the inhibitory effects of plant pathogens through the synergistic activity of antibiotics and cell wall-degrading enzymes (Abdul Wahid et al., 2007; Aleandri et al., 2015). *Trichoderma* colonizes the roots of the plant which promotes growth and enhances resistance to systemic diseases in the plant by endophytes (Doley et al., 2019). *Trichoderma* is very important in the fight against many pathogens because of its ability to release toxic substances that inhibit or limit the activity of these pathogens (Naseby et al., 2000). Inoculating fluorescent *Pseudomonas* mycorrhizal fungi with *Trichoderma spp*. significantly improved plant height and reduced Panama wilt in banana plants (Mohandas et al., 2010). *Trichoderma* as a biological control agent has been widely used to control different *Pythium* species.

Trichoderma has many important activities such as breaking down cell walls, producing hydrogen cyanide and indole acetic acid, and dissolving phosphate (Vainayarani and Prakash, 2018. Some studies have indicated the use of applying different types of Trichoderma spp. to broccoli, sugar beets, tobacco, hot peppers, cucumbers, and tomato plants (Muthukumar et al., 2011; Mbarga et al., 2012; Kipngeno et al., 2015). A study of applying the fungus *Piriformospora indica* (Pi) and *Trichoderma harzianum* (Th) on the morphology, yield and essential oils of mint showed positive effects on plant morphology and essential oil ratio. *Trichoderma harzianum* inoculated compost-modified growth medium showed suitable physical

properties and similar growth in the production of lavender and rosemary plants compared to the commercial substrate (Agulló et al., 2011). The inoculation with Th improved plant productivity and quality. The highest yield of essential oil was observed with the combined application of two fungi (Vafayi et al., 2019). A study was performed using *Piriformospora indica* and *Trichoderma virens* to assess the growth, morphological and physiological parameters of mung bean (*Vigna radiate* L.) (Tamalla et al., 2014). Only *P. indica* increased plant length and root length. Additionally, the fresh and dry bean weights were increased as was stem, bud, and whole plant dry weights when the bean seed were inoculated.

Habitat and cultivation of Lavandula plants

The genus *Lavandula* includes species of short-lived to annual shrubs which vary in their aroma and tolerance to drought and high temperature (Upson, 2004). Lavender originates in the Mediterranean basin and grows in limestone rocky areas (Swanepoel and Alberts, 2009). Lavender is found in North Africa, the Mediterranean, Europe, and western India (Knosravi and Sendi, 2013). It has been cultivated since the time of the ancient Greeks and Romans. The species *L. angustifolia* is the most important commercial species of lavender. Other important species are *L.* x *intermedia* and *L. spica*. All types of lavender are used in medicinal preparations, although they have different medicinal properties. Lavender can be grown in fields, gardens, or containers. It is cultivated commercially all over the world mainly in France, the United Kingdom, Bulgaria, Italy, Romania, Australia, China, Russia, and India (Lawrence,2004).

Lavender needs the maximum amount of sunlight and heat, so it is best to plant it in full sun. Seedlings are planted either in the fall or in the spring. Spring cultivars are smaller and maintain a healthy rooting system. Lavender is planted at 120 cm between rows and 30 cm to 40

cm between plants. This distance between plants depends on the available moisture, size, and variety of cultivar as well as mechanical cultivation and harvest time (Kimbrough and Swift, 2009). Lavender needs abundant watering for the first two years to help the plant grow well. Irrigation should also be done regularly in the various stages of plant growth, especially in the stage of flower formation, which is the stage in the life of the plant on which the percentage of essential oils obtained from the plant depends. Watering may increase oil production in mature plantings. Drip irrigation is recommended as it helps control weed growth (Ernst, 2017). There are very few pests or disease problems at flowering time. Excessive watering during summer, poor drainage and excessive moisture are the main causes of fungal diseases in lavender (Adam, 2006).

The genus Lavandula

The genus *Lavandula* contains many different species (Fig. 8). It belongs to the Lamiaceae (syn. Labiatae) family and is of the Mediterranean countries of origin (Baytop, 1984). Varieties of this family are distinguished by their multiple pharmacological effects, such as anticonvulsants, calming, antispasmodic, analgesic, antioxidant, and topical anesthetics (Hosseinzadeh et al., 2004; Kovacheva et al., 2001). *Lavandula* species are distinguished by their high content of essential oil (EO) productivity which has economic value in the fragrance industry (Kovatcheva et al., 2001). The native species of lavender *L. angustifolia* Mill. (syn. *L. officinalis* Chaix.; *L. vera* DC); spike lavender, *L. spica* DC (syn. *L. latifolia* Mill.), and lavandin, *L.* x *intermedia* Emeric (syn. *L. hybrida* Revr.) are consider the most economically important (Gamez et al, 1990). Lavender is propagated mainly by seed, cuttings, layers, tissues culture, and division of roots (Hassiotis et al., 2010). Varieties derived from *Lavandula angustifolia* produce different types of lavender essential oil and are widely distributed throughout the world. The

different species of lavender differ in genetics and cultivation, or the conditions of growth and climatic factors, which have a broad influence on the chemical composition of essential oils (Gouyon et al., 1986; Vokou et al., 1993). Different lavender species have unique botanical properties and chemical ingredients with differences in the reported therapeutic uses for the different species. (Agricultural Research Service, 2000) (Fig.2.2).

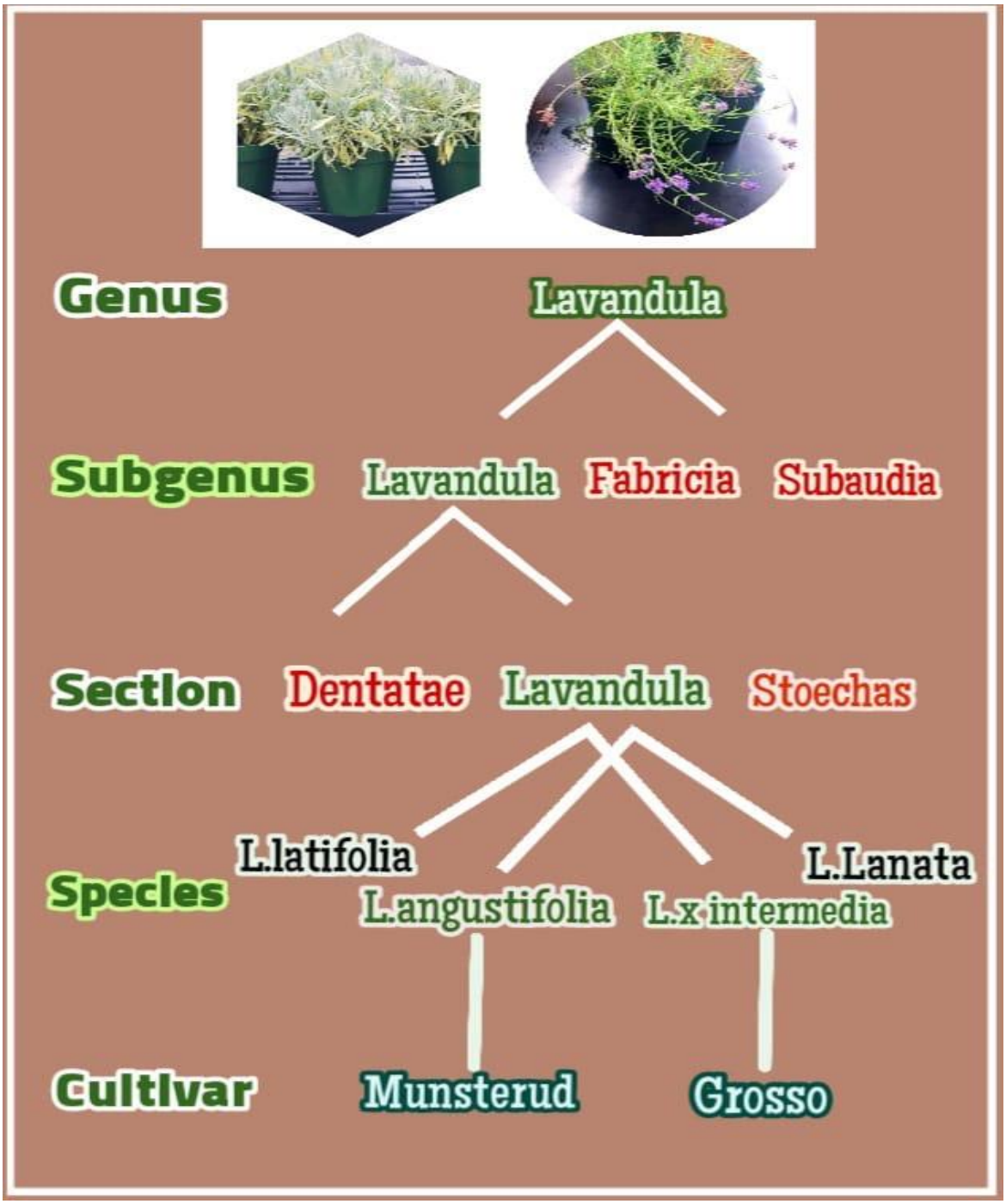

Figure 2.2 Subgenus, sections, species, and cultivars within the genus Lavandula (Credit by AL-Garallaa) (https://www.perennials.com/plants/lavandula-angustifolia-platinum-blonde.html).

Description of Lavandula species used in this study:

According to the morphological characteristics, the genus *Lavandula* is divided into three subgenera: Lavandula, Fabricia and Sabaudia. Each subgenera is diversified into sections which are divided into species. For example, *Lavandula* includes the Divisions Lavandula, Dentatae and Stoechas; *L. angustifolia*, *L. latifolia* and *L. lanata* are species of *Lavandula* (Fig.2.2). Moreover, there are also interspecific hybrids which arise from the natural or artificial crossing of two species. Lavandin (*Lavandula* x *intermedia*) is derived from the cross of *L. latifolia* x *L. angustifolia*. It is an important type of lavender known worldwide (Upson and Andrews, 2004), **(**Fig. 2.2).

Lavandula angustifolia

Lavandula angustifolia is also known as true lavender and has many varieties such as 'Hidcote Superior', which has a dark purple flower color, green foliage, stem length 6-8 inches, and blooms once a year in spring. The plant height is 20 inches. *L. angustifolia* contains over 51 varieties that differ according to the original habitat and flower color (Bader, 2012). The flower spike consists of cymes, a branching determinate inflorescence with a flower at the end of each branch (Fig.2.3) either in an opposite decussate or alternate spiral arrangement, which are subtended by bracts (Lis-Balchin, 2002). *L. angustifolia* is considered the most important commercial species of lavender. The plants of this species are grown all over the world. This species has a sweeter aromatic scent that differs from other lavender species (Bader, 2012). *L. angustifolia* is also characterized by rapid growth and high yields of oil from accumulated monoterpenes. *L. angustifolia* is widely cultivated in the Mediterranean regions to produce basic materials essential oils used in perfumes, cosmetics, and flavorings (Segura and Calvo, 1991). *L. angustifolia* tolerates high humidity and creates a beautiful round shape in greenery. It has many

important varieties including: ‘Hidcote Superior’, ‘Dwarf Blue’, ‘Thumbelina Leigh’, and ‘Blue River’.

Figure 2.3 *Lavandula angustifolia* “ Hidcote”

Lavandula x intermedia

Lavandula x *intermedia* varieties or lavandins are sterile hybrids of *L. angustifolia and L. latifolia* (spiked lavender). They produce more spikes than other lavenders. It has an acquired camphor odor somewhat like the smell of wood. Moreover, it is characterized by the high production of essential oils that are superior by up to five times that of *L. angustifolia* varieties. It is also distinguished by its beauty in the gardens. *Lavandula* x *intermedia* has several varieties, the

variety used in this study was ‘Provence”, which is characterized by light purple flower color, green foliage color, stem length 24-30 inches, and blooming once a year in early summer . The plant height is 48-60 inches. *Lavandula* x intermedia is resistant to fungi (Bader, 2012) (Fig.2.4).

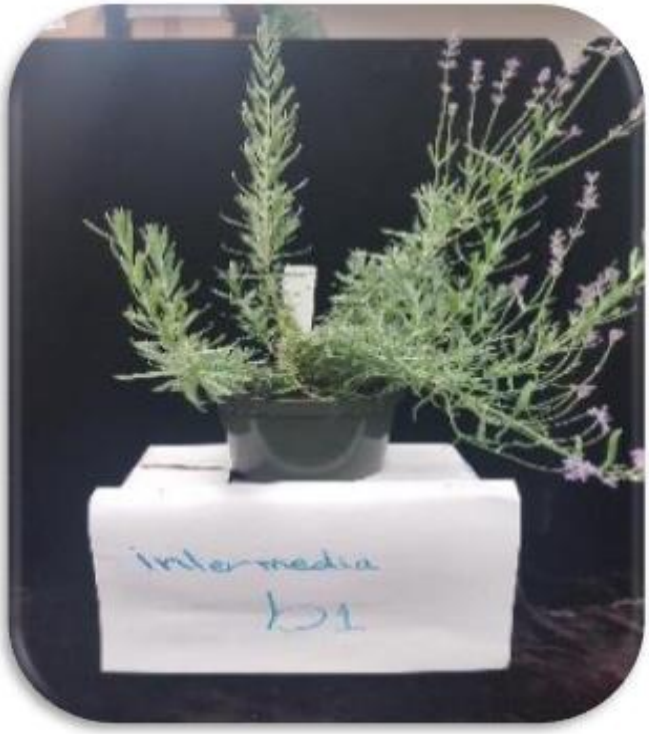

Figure 2.4 *Lavandula* x *intermedia* ‘Provence”

Lavandula stoechas

Lavandula stoechas can be easily identified by its cylindrical, rolled-up flowers resembling rabbit ears or butterfly wings. It is characterized by plum flower color, moss green foliage, floral stem length of 2-4 inches, and it blooms continuously starting in early spring. It is known as a healing plant natively growing on the islands of Stoechades (now known as Hyeres) (Festing, 1989). The plant height is 25-30 inches (Bader, 2012). *Lavandula stoechas* is an

evergreen shrub growing up to one meter (Benabdelkader et al., 2011). This species was named *stoechas* relative to a former name of the island group off the coast of France. *Lavandula stoechas,* is also known as 'cantueso', French lavender, Spanish lavender, or topped lavender (Da Porto and Decorti, 2008). Varieties of this species are distinguished by their tolerance of high humidity. The species often grows as small evergreen shrubs having aromatic foliage and flowers (Allaby, 1992). *Lavandula stoechas* is cultivated to extract essential oils on a small scale mainly in Spain for use in air fresheners and deodorants. It is widely cultivated as an ornamental shrub. This species has been classified as an invasive weed in a part of Australia and therefore strictly controlled. It is also known as French or Italian lavender (Lis-Balchin, 2002) (Fig.2.5).

Figure 2.5 Lavandula stoechas 'Otto Quast'

***Lavandula angustifolium* 'platinum blonde'**

'Momparler' lavender or 'platinum blonde' (*Lavandula agustifolia*) is an aromatic herb, characterized by its beautiful flowers and attractive gray leaves marked with a cream color. It is considered an evergreen plant but does not tolerate extreme cold in the winter. The plant needs pruning after flowering in early spring before new growth begins. Flower color is light blue blooming in early Summer, mid-Summer, and late Summer. Foliage color is grey green, variegated white with the plant growing to 16-23 inches (40-60 cm). 'Platinum Blonde' is a

multiple award winner including a nomination for Plant of the Year at the Chelsea Flower Show in 2013 (http://www.perennialresource.com/variety.php?ID=LAVPB) (Fig. 2.6).

Figure 2.6 *Lavandula angustifolia* 'Platinum Blonde' (English Lavender)

Chemical composition of lavender essential oils

Natural Products

Natural products are of natural origin and includes organisms, whether a plant or an animal, which have not undergone any kind of treatment other than simple preservation, such as drying. Natural products can be dried leaves or flowers of plants, an isolated animal part, secretions, or pure compounds, such as alkaloids, coumarins, flavonoids, glycosides, peels, steroids, polysaccharides, terpenoids, etc., isolated from plants, animals, or microorganisms (Samuelsson, 1999). However, in most cases the term natural products refer to secondary metabolites. Natural products can also be sourced from land or marine (Sarker and Nahar,

2012). The current increased interest in natural products is attributed to the need for new therapeutic compounds. The distinguished diversity in the chemical composition and the biological activities of the natural secondary metabolites along with the development of modern and sensitive technologies for the identification of bioactive natural products and improvement of techniques for isolation, purification, and chemical composition of these active compounds has increased interest and availability of natural products (Rout et al., 2009).

Classification of natural products:

Natural products are classified into two large sections (Mullen et al., 2006; Harborne1973). The first section contains the compounds involved in the initial reactions resulting in carboxylic and amino acids, sugars, fats, proteins, and nucleic acids. The second section contains secondary biological compounds, which may be classified according to the natural sources from which they are produced, or according to their physiological effects or structure, the most important of which are terpenes, steroids, alkaloids, and flavonoids. There are three main substances: shikimic acid, acetate, and amino acids which are the building blocks of secondary metabolism. The products of secondary metabolism are classified into terpenes and their derivatives, phenolic compounds, alkaloids and their equivalents, and antibiotics and vitamins.

Methods for identifying the structural composition of natural products:

- Physical methods: used to determine the melting point, boiling point, solubility, aroma, and activity.
- Chemical methods: used to identify the active groups present in the natural compound followed by the preparation of derivatives of the same compound.

■ Spectroscopy methods: used to identify the structure of organic compounds and have facilitated identifying the natural compounds isolated.

■ Ultraviolet spectrum: enables identification of organic compounds containing active groups with free electrons, or compounds containing reciprocal double bonds like turbines, as the wavelength containing reciprocal double bonds.

■ Infrared spectrum: gives information about the compound and the functional groups present.

■ Nuclear magnetic resonance spectrum provides information on the structure of organic molecules and the structural structure of organic compounds.

■ Mass spectroscopy: to determine the molecular weights and identity of molecular components of a substance (Harborne, 1998).

Active compounds

Terpenes

Terpenes act in plant defense as a deterrent against feeding on herbivores (Langenheim, 1994) and as attractants for pollinators. They are widely found in nature. More than 22,000 unique terpenoids are known today, making terpenoids the largest group of natural products. Among its properties are its usefulness in the storage of agricultural products, such as germination inhibitors in potatoes. The biotransformation of terpenes is important by producing enantiomerically pure flavors and fragrances under mild reaction conditions, terpenes constitute the largest class of plant secondary metabolites (Dudareva et al., 2004). Terpenes volatilize easily at room temperature and are considered major components of floral scent and essential oils in herbs, vegetables, and fruits (Dudareva et al., 2004; Dudareva and Pichersky, 2008).

All plant terpenes consist of the carbon precursor 5-isopentenyl diphosphate, and dimethylallyl diphosphate, both of which are derived from two alternative pathways, the mevalonate pathway in the cytosol or the methyl erythritol phosphate pathway in the plastids. Then they are converted into monoterpenes, sesquiterpenes, and diterpenes through the activity of terpene synthase enzymes (TPS) (Tholl, 2006). The components of the essential oil of lavender consist mainly of monoterpenoids and sesquiterpenoids (Shellie et al., 2002; Fakhari et al., 2005; Jung et al., 2005). The percentages of these components vary greatly, with (R) -linalool and (R) -linalyl acetate being the most abundant compounds. Other ingredients identified are (R) -limonene, 1,8-cineol, camphor, terpinen-4-ol, lavandulol, lavandulyl acetate and α-terpineol (Kreis and Mosandl, 1992; Jung et al., 2005). Landmann et al. (2007) noted the cloning and biochemical characterization of two monoterpene synthases and a sesquiterpene synthase have been used to make significant contributions to the flavor of *L. angustifolia* essential oil and to improve essential oil production through molecular biological techniques or conventional breeding of economically important lavender.

Terpene synthesis in lavender

Lavender essential oil is a complex blend of mainly monoterpenes, for example, linalool, camphor, and 1,8-cineole (Lawrence 2004). Several terpenes have been isolated and cloned in relatives of lavender including mint and sage (Darshan and Doreswamy, 2004; Huber et al., 2004). The functional roles of terpenoids are characterized as hormones (gibberellins), photosynthetic pigments (phytol, carotenoids), electron carriers (ubiquinone, plastoquinone), as well as communication and defense mechanisms (Langenheim, 1994). The lipophilic nature of many monoterpenes and sesquiterpenes coupled with the wide and high structural diversity

calculate its role as chemical carriers of information (Williams et al., 1989; Croteau et al., 2000). The essential oils in lavender species primarily contain volatile monoterpenes and sesquiterpenes which are linear or monocyclic or dicyclic hydrocarbons, alcohols, or ketones (Upson and Andrews, 2004). Vranova et al. (2013) noted the relative contribution of each pathway to the biosynthesis of different classes of terpenes or even a particular terpenoid is not certain and may depend on specific plant species or different environmental conditions. In this line, research has been conducted to understand the biosynthetic pathways of essential oils. Several genes that encode terpene synthases (TPSs) and contribute to the production of essential oils have been described in different species of lavender, including D-xylulose-5-phosphate synthase DXS, reductoisomerase (DXR), (S) -linalool synthas (sLiS), (R) -linalool synthase (rLiR) and alcohol acetyltransferase (AAT) (Sarker et al., 2013; Jullien et al., 2014; Benabdelkader et a.l., 2015; Adal et al., 2017). Li et al., (2017) it reported that essential oils from *Lavandula angustifolia* consist mainly of monoterpenoids and sesquiterpenoids, however, the fluctuations of the volatile terpenoids showed a regular change in each axis. The results of the research also indicated the molecular mechanism underlying the regulation of volatile terpenoids during the flowering process is poorly understood in lavender. However, lavender blossoms are typically used for studying regulation of terpenoid synthesis in molecular and cellular environmental levels (Adal et al., 2019). Only 15 lavender-derived chains were revealed to terpenoid biosynthesis, which is considered insufficient compared to the frequency of identification of volatile compounds in lavender (Guitton et al., 2010).

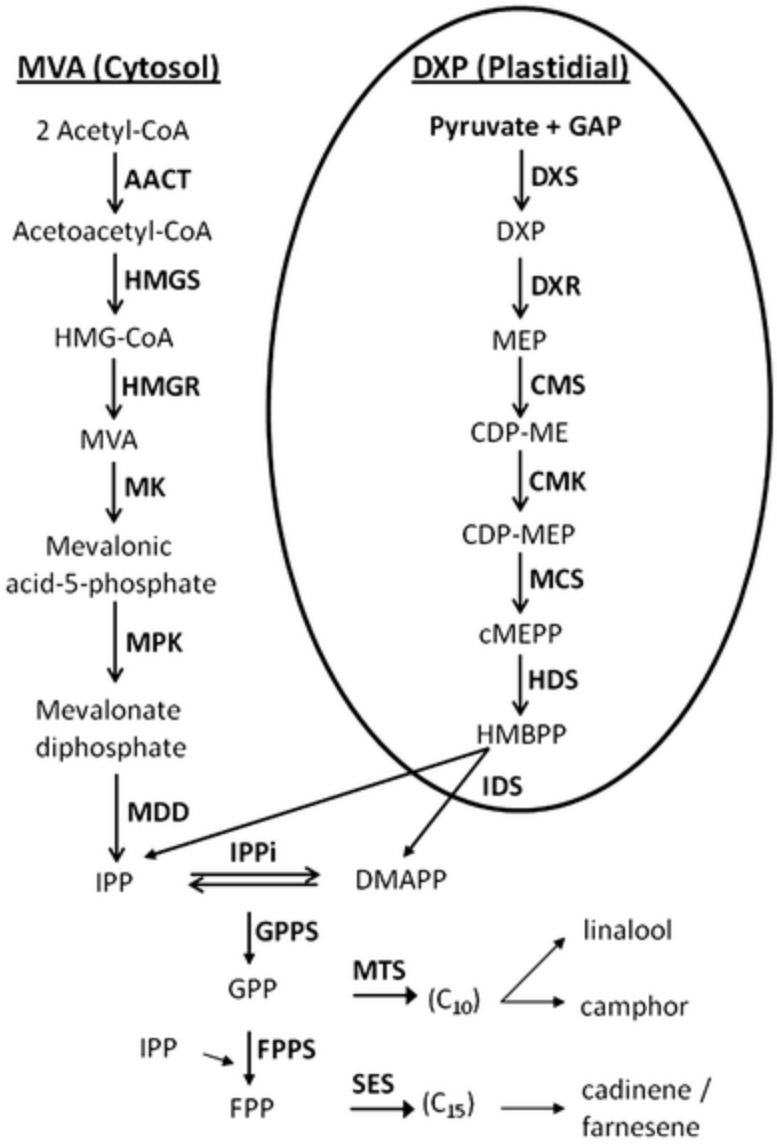

Figure 2.7 The mono- and sesquiterpenes are derived through the condensation of the universal isoprenoid precursor isopentenyl diphosphate (IPP) and its allylic isomer, dimethyl allyl diphosphate (DMAPP) (Lane et al., 2010).

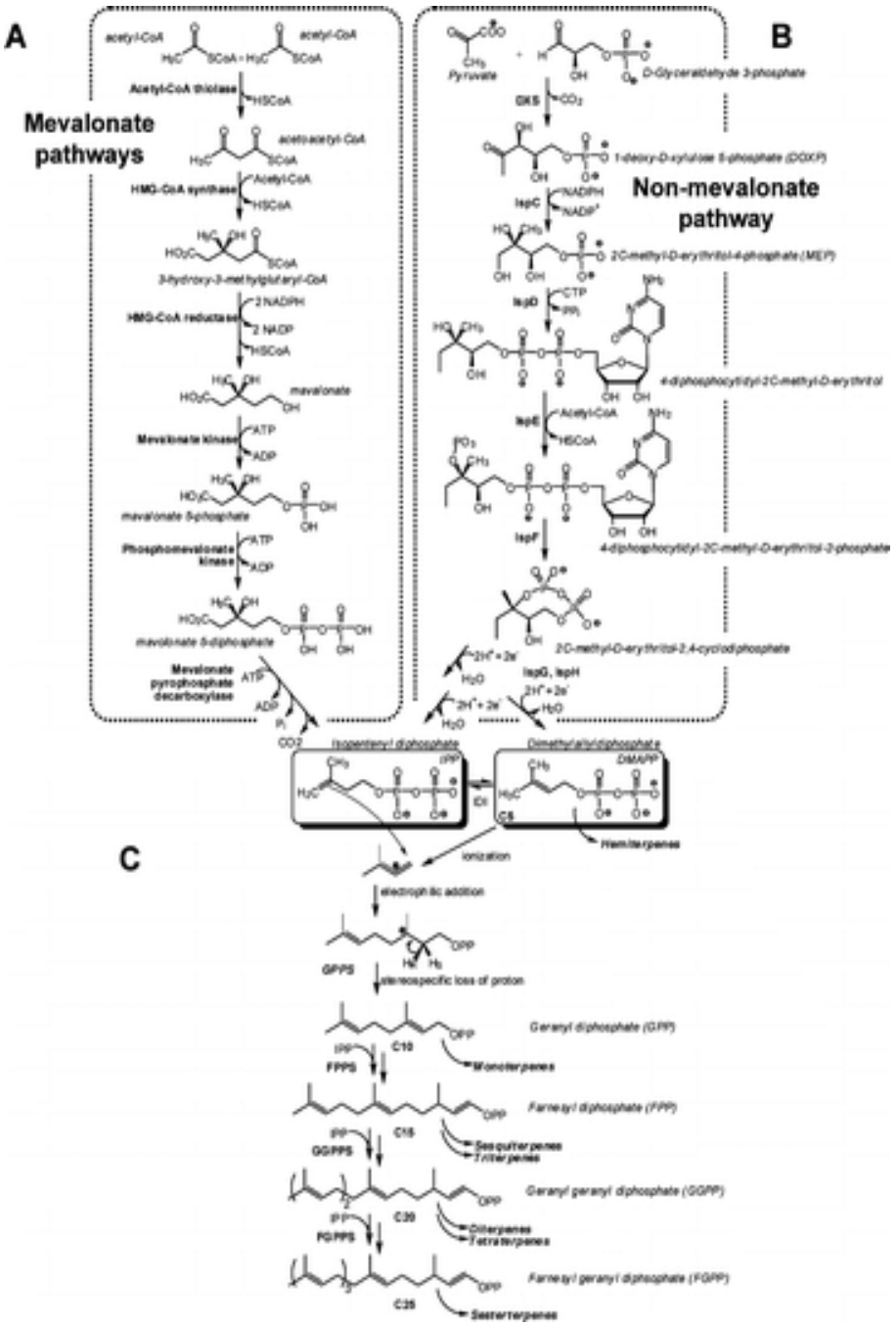

Figure 2.8 Schematic illustration of biosynthetic pathways to terpenoids (Ajikumar et al., 2008). In higher plants, the MVA and MEP pathways are effective. Many mono- and diterpenes are produced in the plastid compartment by the non-mevalonate pathway. The picture is complicated by the exchange of certain terpene precursors between the two compartments. Cross-linking of the two pathways leads to the synthesis of terpenes in plastids that can be partially derived from the mevalonate pathway in the cytoplasm and vice versa (Bick and Lange, 2003; Laule et al., 2003)

Table 1.1 Major mono- and sesquiterpenes in English lavender, lavandin and spike lavender (Lis-Balchin, 2002).

Terpine	English lavender	Lavandin	Spike lavender
Camphor	0.5-1%	4-11%	10-20%
Caryophyllene	3-12%	n.d.	n.d.
1,8-Cineole	1-2%	5-10%	20-30%
Linalool	30-49%	30-40%	40-50%
Linalool acetate	30-45%	20-30%	<1%
Ocimene	2.5-6%	n.d.	n.d.
Pinene (α and β)	n.d.	n.d.	1-3%

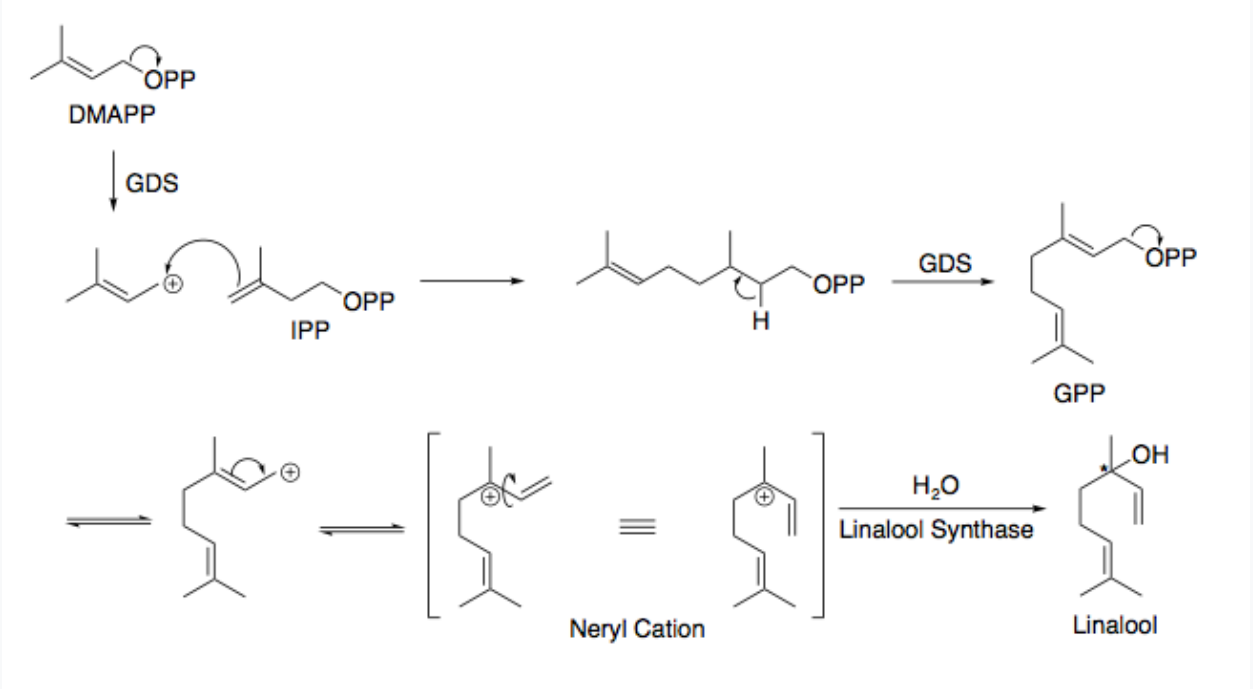

Figure 2.9 Linalool biosynthesis pathway.

Abbreviations used: geranyl diphosphate synthase (GDS), pyrophosphate ester (OPP), isopentenyl pyrophosphate (IPP), dimethylallyl pyrophosphate (DMAPP), geranyl pyrophosphate (GPP). Stereogenic centers are indicated by an asterisk (Woronuk et al., 2011.; Dewick, 2002; Cseke et al., 1998; Iijima et al., 2004).

The history of lavender use:

The common name lavender is derived from the Latin larvae. It is believed the first use of essential oils in folk medicine was by Ibn al-Baitar (1188-1248) an Andalusian doctor and pharmacist (Houtsma, 1993). Lavender is perhaps best known for its common use in perfumery and has been used medicinally throughout history. Many types of lavender are grown around the world. It is believed each species has different properties. Today, interest in lavender has increased for aromatherapy. Flowers and lavender leaves were used in the ancient Roman and Carthaginian washrooms to give an aromatic aroma to the water of the bathrooms where they were washing. They found, through experience, that lavender was good for washing their heads

and that it appeared to be effective in treating mental illness**.** It was said that Mary Magdalene had an alabaster box containing lavender oil which she opened and wiped off Jesus' feet. Queen Elizabeth owned a farm of lavender flowers and was even named after the queen plant. Shakespeare also had a share of this plant where he mentioned it in a tale of the middle ages. Lavender was sold in the city of Neruda in Syria in the time of the Greeks where its blossoms were sold for 100 dinars per pound, then a very large sum.

The Romans called it the name of girlfriends, believing there was a viper living among the lavender and to be careful when picking the plant as most of them thought the plant was closely related to ginger. In England, the prevailing belief was that it kept the clothing white, pure, and free of insects when washed with lavender and was called Lavyndull. Commercially, lavender has been grown in England for centuries. Prices of lavender grown in England were much higher than lavender grown in France since it contained the best oils, and the varieties of English lavender produced the most beautiful smells of all varieties. The use of lavender is attributed to the Romans and the ancient Greeks as the most famous aromatic and medicinal plant (Cavanagh and Wilkinson, 2002). The traditional uses of lavender range from its use as a fragrance to an antimicrobial agent. This herb with its powerful and effective compounds has been used throughout ancient times and is still considered a popular household item today.

Recent studies have found that lavender essential oils were historically used to preserve potato tubers in storage instead of chemical methods (Vokou, 1993). Medicinal remedies of lavender were used in the 1st century AD and were mentioned by the Greek naturalist, Dioscorides. Lavender was used as a perfume and as a main ingredient for incense in ancient Egypt. It was also known as the love herb in the Middle Ages. Lavender in China was named

white flower oil. Its oil has been used to embalm corpses, treat animals for lice, and tame lions and tigers throughout historical ages (Joe et al., 1999). Flowers were used in ancient Rome and North Africa to smell good in public baths and were carried Roman army for use as an anti-infectious substance (Barrett et al.,1996). Lavender was also used in the Middle Ages and Renaissance to store laundry. It was also said that the ancient Egyptians used lavender flowers in the mummification process (Duke, 1985). Lavender has also been used in Chinese medicine to treat many conditions including infertility, infection, anxiety, and fever (Kenner, 1998). Lavender has long been used in Arabic medicine to treat stomach pain and kidney problems (Ghazanfar, 1994). Lavender has been used in various folk traditions to treat dizziness and hair loss (Hay, 1998; Duke, 1985). Lavender was recognized as an herb in 1999 by Herb Growing and Network Marketing in the United States of America (Evelegh, 1996; Anonymous, 1999(.

Morphology and growth habit of Lavandula:

Lavender is an herbaceous plant (Fig. 1) commercially cultivated for the extraction of essential oils in France, Spain, America, Bulgaria, and England (Cawthorn 1995; Vakili et al.,2014). It is also cultivated in India as well as other parts of the world because of its characteristic aromatic oil with medicinal properties found in its flowers (Basch et al., 2004; Naðalin, 2014). The plant height is between 50 to 70 cm, a shrub that blooms in mid-July and prefers to grow in sunny areas (Upson, 2002) with leaves that are different by species. In general, the leaves are simple, covered with soft hair and small glands containing essential oils. The leaves have curly edges and are colored in silver and green and covered with indumentum that protects them from strong sunlight and desiccation (Upson and Anderson, 2004). The flowers are carried on a petiole where the flower rises above the leaves. In some species, the bracts of bright colors may be blue or purple and sometimes a yellowish purple color. The bracts

are tubular with five lobes (Upson and Anderson, 2004). Lavender is suited to warm conditions, temperatures of 26°C, and develops best under long days in sun and needs dry, well-drained soil with a 6.0-8.0 pH (Grieve, 1971; Ernst, 2017). The harvest is done manually or mechanically according to the required use (Ernst, 2017).

Figure 2.10 Lavender plants in the field.

(https://www.google.com/search?q=picture+of+lavender&sxsrf=ALeKk01keTVtO8tBcPLedbdPlVZ5p-m2oQ:1623011650120&tbm=isch&source)

L. intermedia ***L. stoeches*** ***L. angustifolia***

Figure 2.11 The three species of lavender used in the current study (photo credit : Keefah Al Garallaa).

Figure 2.12 *Lavandula angustifolia* 'Platinum Blonde'.
(https://www.easytogrowbulbs.com/products/lavender-platinum-blonde?variant)

The stages of maturity

Lavender inflorescences are spikey, that is, inflorescences of an indeterminate main axis (central flower stalk) arranged in circles of opposite pairs (Fig.2.11). *Lavandula angustifolia* is characterized by a perpendicular remote control at the base of its non-branching center stem and all vertical columns are equal in size to the top of the inflorescence, while the *L.* x *mediator*

produces a branched inflorescence (Figure 2.13) lateral inflorescences, and an unlimited number of vertical columns of equal size at first and then at an increasingly smaller size towards the top of the inflorescence. In lavender species, VOCs are produced and accumulate in specialized trichomes scattered on the surface of all green tissues such as leaves, calyces, stems and bracts (Schmiderer et al., 2008). The stage of development of inflorescences is known to affect the nature of the volatile terpenes that accumulate in the leaves of many plant species including members of the *Lamiaceae* such as lavender, sage, and mint (Sangwan et al., 2001). No study has evaluated the effect of flora development on biosynthesis and accumulation of volatile terpenes in lavender.

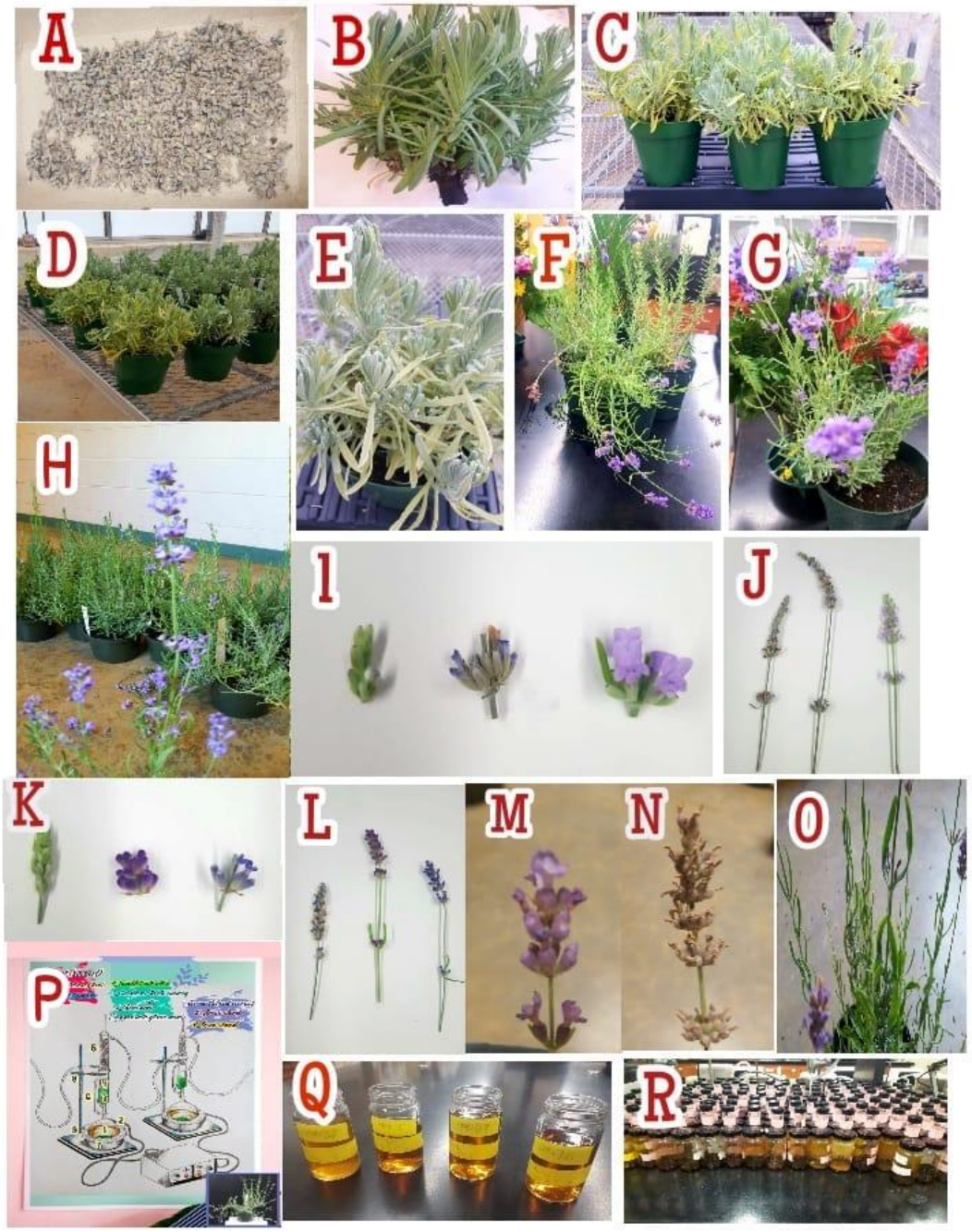

Figure 2.13 A. Lavender seeds, B. *Lavandula stoechas* growing in the greenhouse, C and E *L. angustifolia* 'Platinum Blonde', D. experiment in the greenhouse, Fand G. *L angustifulia*, H. *L.* x *intermedia*, I. development of flowers, J, K, L, flowering for three species of lavender, M, N, and O. flowering of *L. angustifulia* and *L.* x *intermedia*, P. the Soxhlet used to extract essential oils of lavender, Q and R. the samples after the extraction. (photo credit: Keefah Al Garallaa)

Description of flowering and flowering areas

Lavender inflorescences are spikey, i.e., inflorescences of an indeterminate main axis (Central flower stalk) which are organized in rings of opposite pairs. Lavender blossoms are distinguished by their floral arrangement's different stages of ripening, from immature to pale colored, while open flowers are distinguished by being scattered on different columns during the stages of flowering development (Guitton et al., 2010). Lavandula essential oils are stored in glandular hair distributed over the aerial parts of the plant, especially in flowers (Sudria et al., 1999). The oils from Lavandula species are easily released from glandular filaments in flowers during the distillation process (Werker, 1993). The essence accumulates in an area under the skin and then the essence is released by tearing of the epidermis (Figueiredo et al., 1994). Bloom time of lavender depends on the variety and your growing conditions particularly sun and warmth. Some lavender blooms once a season for 3-6 weeks while a few blooms continuously all summer. Others can bloom twice and even three times if the finished flowers and stems are removed promptly)https://empressofdirt.net/grow-lavender/).

Oil provenance

It could be only climatic factors that affect volatile oil production and chemical composition (Elena and Daniela, 2017). The accumulation of terpenoids in plants determines their synthesis according to different temperatures in the soil, air, rain, and hours of sunshine. Perhaps the most important factor is the distribution of sunlight hours over the volatile portion of plant matter. Fresh lavender flowers are the basis of volatile oil and fresh flowers have more complex formation while the dried flowers change their odor and pharmacokinetics.

Lavandula harvest

The best time to harvest lavender is early morning. Harvesting in early spring gives the plant enough time to produce another crop of flowers again in late summer through fall. This is typical of a short summer growing season. In frost-free climates, some types of lavender may bloom year-round, so that small crops may be harvested over and over throughout the year **(**Deanna, 2020**).** Wahyuni et al. (2021) mentioned that post-harvest drying affects the volatile composition of flavor components. Erbas and Baydar (2008) indicated the fresh yield of flowering stems harvested by hand was about 2.5 tons/hectare and about 1-2% oil extracted using steam distillation after air drying in the sun. The harvest method has a great influence on the essential oil yield (Çiğdem et al., 2021).

Environmental conditions and their effect on lavender oil:

One of the important factors contributing to increased production of essential oils with good quality is the length of the day (Ellis, 1960). Low temperatures adversely affect the increase in production of essential oils (Sangwan et al., 2001). Environmental conditions, genetic characteristics, harvest time and plant fraction used (flowers vs. foliage) have a direct impact on content of essential oils (Marotti et al., 1996; Munoz-Bertaneu et al., 2009). The method of planting and cultivation may affect the production of oils (Atalay et al., 2008). There is also an impact on essential oil production due to distillation and drying methods of plant parts (Pinto et al., 2007; Baydar and Erbas, 2007). Production of linalool in lavender depends on temperature, flower growth and rainfall (Prusinowska and Emigielski, 2014; Hassiotis et al., 2014). Environmental factors such as temperature, precipitation, moisture, and minerals affect the productivity of secondary metabolites of plants as they affect gene expression pathways (Nair et al., 2000). There is a reverse relationship between the production of essential oils and

temperature on lavender *(L. angustifolia)* during the flowering period (Silva et al., 2013). As in lavender, the production of aromatic oil in mint increased with high temperatures and sunny days, although excessive temperatures lead to a loss of essential oils (Evans, 2002). The oil content of the leaves in aromatic plants may decrease when exposed to low temperatures and rainfall (Hassiotis et al., 2014). However, low soil moisture may lead to an increase in essential oils (Sonmez and Bayram, 2017). Hassiotis et al. (2014) pointed out the essential oils of lavender may be affected by climatic conditions when harvested during the day further explaining why the aromatic oil content and quality of lavender may change after harvest. In addition, soil nutrients may have a significant effect on the production of essential oils in medicinal plants (Tayade et al., 2013).

Baydar, H.A.S.A.N. and Erbaş, S., 2007, April. Effects of harvest time and drying on essential oil properties in lavandin (lavandula× intermedia emeric ex loisel.). In *I International Medicinal and Aromatic Plants Conference on Culinary Herbs 826* (pp. 377-382).

Essential oil compounds:

Essential oils are oils that contain the "essence" of the plant, such as its smell or flavor. Essential oil compounds in plants are secondary products. Only 20-30% of all plants have been researched so far for their essential oils (Wink, 1999; Ceylan, 1997). Some of these secondary compounds are malodorous while others are very pleasant. They are produced in small quantities, from 0.01- 20% of the plant, based on tissue fresh weight (Rumbangh, 1999). Essential oils are used in aromatic products and to add flavor to food and beverages. A primary use is for aromatherapy. Some oils, such as tea tree oil, are prescribed as a home remedy for a variety of different diseases from acne to dandruff. Tea tree oil makes up 36 percent of the global

essential oil market, more than any other type of essential oil. Another popular essential oil, lavender oil, is expected to grow in market value. According to Runham (1998), the global trade in essential oils increased by 40% from 1984 to 1998. The global demand for essential oils is expected to increase dramatically between 2018 and 2025; From about 226.9 kt to 404.2 kt (Shahbandeh, 2019) (Fig.2.14).

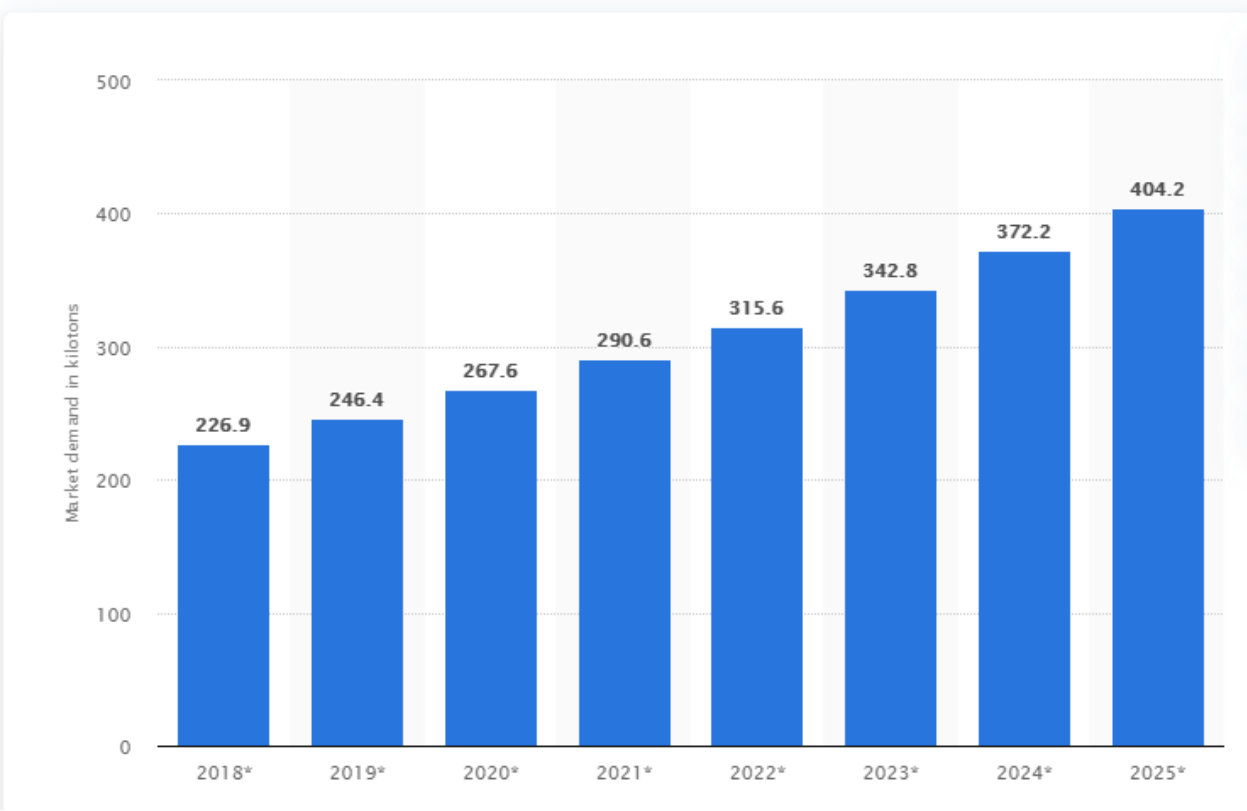

Figure 2.14 Market demand of essential oils worldwide from 2018-2025 (Source: *Global demand of essential oil 2018-2025*; Shahbandeh, 2019).

Volatile oils are one of the primary products produced by plants, especially medicinal and aromatic plants. These oils are a mixture of various organic substances of varying composition such as hydrocarbons, aldehydes, ketones, alcohols, esters, and others which are volatile at

normal temperatures. Most oils exist in a free, liquid state while a few are solids or resins. Essential oils oxidize when exposed to light and air and turn into solid resins. The volatile oils are single terpenes and sesquiterpenes. Volatile oils evaporate or volatilize without decomposing distinguishing them from fixed oils. Volatile oils do not become rancid with exposure to direct light and air (Hariharan et al., 2009).

The volatile oils are divided into two main groups:

Oleoptenes (syn: eleoptene)

Oleoptenes are the liquid part of the essential oil. These hydrocarbons are made up of isotopes of terpene hydrocarbons with each isoprene unit consisting of 5 carbon atoms and their chemical isoprene unit. The isoprene units are combined with the formation of volatile oils in the plant.

Stearoptenes

Stearoptenes are the oxygenated solid parts of an essential oil. The medicinal effect of oil is usually attributed to oxygenated compounds which have the odor and taste of the essential oil. Most of the important essential oil components may be oxygenated compounds (Tyler et al., 1988).

Lavender Essential oil compounds

Lavender oil has been used in folk medicine as an aromatherapy treatment to relieve pain, relieve anxiety and promote energy (Lahlou, 2004; Kako et al., 2008; Kiecott-Glase et al., 2008). Lavender oil is added to cleaning materials, furniture cleaning materials and to food and drinks. It gives flavor and fragrance. The chemical composition of essential oils in lavender has been investigated since the 1930s. Aromas extracted from flowers used in the perfume industry are

made up of 70% terpenoids (Stashenko and Marlinez, 2008). Lavender flowers are the main source of lavender oil for perfumes (d'Acampora Zellner et al., 2006). Acyclic monoterpene compounds are essential oils used in the composition of flavors and perfumes (King and Dickins, 2003). Linalool is an acyclic monoterpene alcohol and an important component of the essential oil in lavender. Linalool is found in more than 200 species of plants belonging to different families (Stashenko and Martinez, 2008). Linalool and linalyl are considered the most essential compounds in lavender oil (Fig.2.15) (Buchbauer et al., 1993).

As a result of environmental influences, lavender oil undergoes many chemical modifications (Hagvall et al., 2008). Oxolinalod, 8-hydroxylinalool, and 8- carboxylinalool are derivatives of linalool (Aprotosoaie et al., 2017). Studies have also shown lavender leaves contain cytochromes that generate linalool metabolites (Boachon et al, 2015). The medicinal and olfactory properties of lavender oil are mainly attributed to monoterpenes, which are the group of volatile organic compounds that make up the essential oil of lavender and are responsible for lavender's distinctive aroma. There are forty to fifty different monoterpenes in lavender essential oil, with linalool, linalool acetate, 1,8-cineole, beta-osmine, terpinene-4-ol and camphor the main ingredients (Kreis and Mosandl, 1992; Flores et al., 2005). The quality of the oil used medicinally is determined by the ratio of monoterpenes with the required biological activity. For example, linalool acetate and linalool, which are present in large quantities in *L. angustifolia*, have sedative and local anesthetic effects, however the use of the variety in disease treatment depends on the type of compounds of the essential oils present in it (Chu and Kemper, 2001). There are three types of lavender, *Lavandula angustifolia* (formerly *L. officinalis*, English lavender), *L. latifolia* (Spike lavender) and *L.* x *intermedia* (Lavandin), primarily used for

producing quality lavender oil (Table. 2.2). Each species of lavender has a unique chemical composition as follows:

- *Lavandula angustifolia* – predominately esters and alcohols
- *Lavandula stoechas* – predominately ketones
- *Lavandula latifolia* – predominately oxides, alcohols, followed by ketones and monoterpenes.

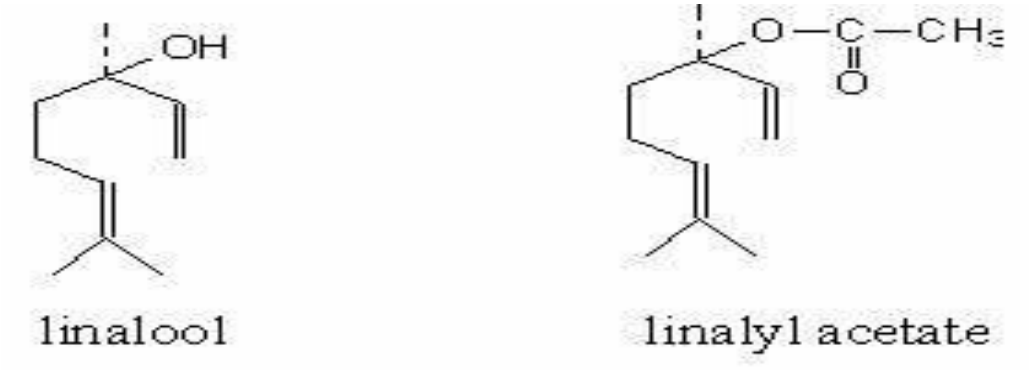

Figure 2.15 structures of linalool and linalyl acetate

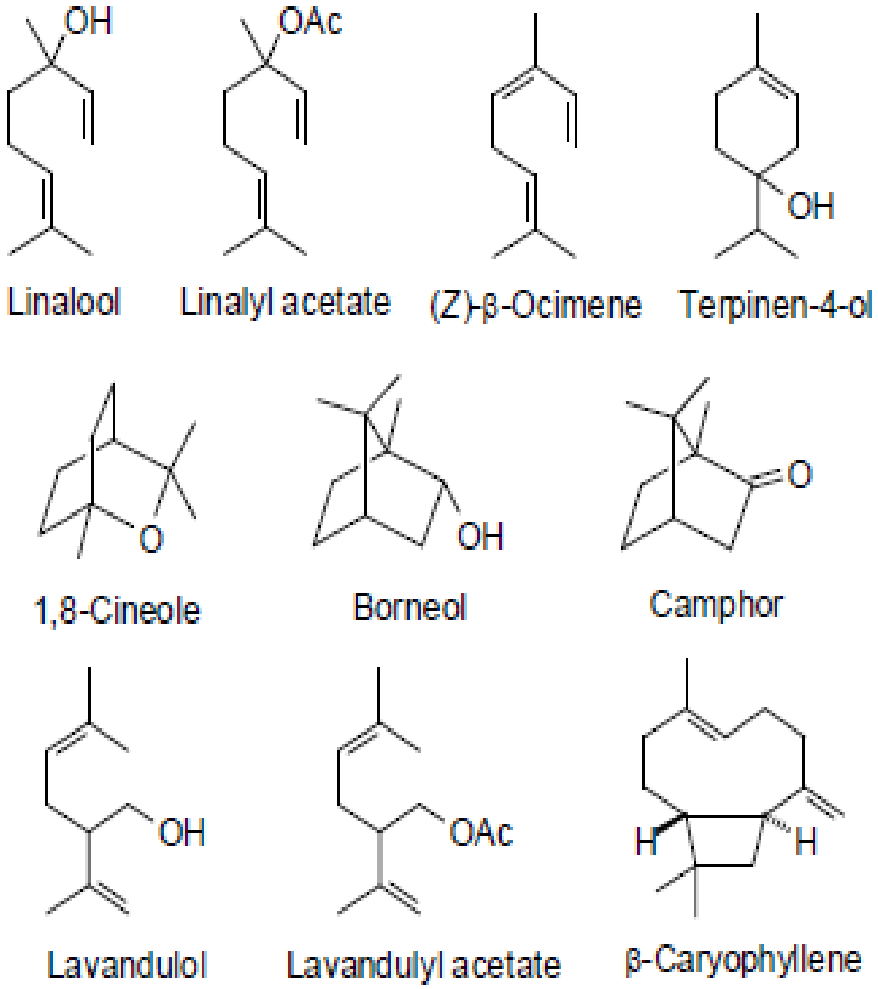

Figure 2.16 The Structure of major chemical components of *Lavandula angustifolia* (Salehi et al., 2018).

Many compounds, the most important of which are linalool, linalyl and camphor, can be obtained from the process of distillation of lavender flowers or leaves (Table.2.2). These compounds are responsible for the medicinal properties of lavender oil (Umezu et al., 2006; Dupuy et al., 2014). (Miri et al., 2015). Lavender oil containing high amounts of linalool and linalyl acetate and a low quantity of camphor is considered positive for the pharmaceutical and cosmetic

industry (Cavanagh and Wilkinson, 2002; Shellie et al., 2002). The aromatic oil in lavender may vary between 2.19-4.45% (Hassiotis et al., 2014).

Table. 2.2 Chemical components of lavender essential oil (Hui et al, 2010).

NO.	Compound	%
1	a-Phellandrene	0.09
2	1S-_-Pinene	0.3
3	Camphene	0.27
4	1-Octen-3-ol	0.17
5	Bicyclo[3.1.0]hexane, 4-methylene- 1-(1-methylethyl)-	0.15
6	β -Pinene	0.43
7	Bicyclo[3.1.0]hex-2-ene, 4-methyl- 1-(1-methylethyl)-	0.39
8	Acetic acid, hexyl ester	0.25
9	3-Carene	0.16
10	Benzene, 1-methyl-4-(1-methylethyl)-	0.04
11	Benzene, 1-methyl-2-(1-methylethyl)-	0.19
12	D-Limonene	0.55
13	Eucalyptol	7.32
14	1,3,7-Octatriene, 3,7-dimethyl-	0.99
15	1,4-Cyclohexadiene, 1-methyl-4-(1- methyl ethyl)-	0.05
16	cis-_-Terpineol	0.13
17	**Linalool oxide trans**	0.30
18	1S-_-Pinene	0.34
19	1,3,7-Octatriene, 3,7-dimethyl-	25.10
20	Octen-1-ol, acetate	0.62
21	Butanoic acid, hexyl ester	0.12
22	**Camphor**	3.79
23	4-Hexen-1-ol, 5-methyl-2-(1-methylethenyl)-, (R)-	0.22
24	Borneol	1.54
25	3-Cyclohexen-1-ol, 4-methyl-1-(1-methylethyl)-	1.56
26	Butanoic acid, octyl ester	0.48
27	p-menth-1-en-8-ol	0.98
28	1,5-Dimethyl-1-vinyl-4-hexenyl butyrate	43.73
29	Bornyl acetate	0.17
30	2,6-Octadien-1-ol, 3,7-dimethyl-, acetate	1.42
31	4-Hexen-1-ol, 5-methyl-2-(1-methylethenyl)-, acetate	0.27
32	1H-Benzoimidazole, 5-amino-1-ethyl	0.07
33	4-Hexen-1-ol, 5-methyl-2-(1-methylethenyl)-, acetate	0.40
34	1,3,6,10-Dodecatetraene, 3,7,11-trimethyl-, (Z, E)-	0.06
35	Caryophyllene	4.38
36	Di-epi-. alpha. -cedrene	0.20

Table 2.2 (continued)

37	Isocaryophillene	0.05
38	a-Caryophyllene	0.11
39	1,6,10-Dodecatriene, 7,11-dimethyl -3-methylene-, (Z)-	0.58
40	1,6-Cyclodecadiene, 1-methyl-5-met hylene-8-(1-methylethyl)-, [s-(E, E)]-	0.68
41	1,6,10-Dodecatriene, 7,11-dimethyl -3-methylene-, (E)-	0.07
42	Spiro [5.5] undec-2-ene, 3,7,7-trime thyl-11-methylene-, (-)-	0.07
43	2,6-Octadien-1-ol, 3,7-dimethyl-, acetate, (E)-	0.20
44	Bicyclo[4.4.0]dec-1-ene, 2-isopropyl-5-methyl-9-methylene-	0.20
45	1-Methylene-2-vinylcyclopentane	0.05
46	Caryophyllene oxide	0.61
47	Copaene	0.17

Use of lavender oil:

Lavender oil has been used in the treatment of sedative disorders, antidepressants, infections, antivirals, and antibiotics, as well as gastrointestinal, systemic, and rheumatic disorders (Danh et al., 2012; Wilkinson, 2002) (Fig.2.17). Lavender oil has been shown to improve sleep in older people (Hudson, 1996) and in infants (Field et al, 2008). It increases deep sleep when inhaled during sleep (Goel et al., 2005). Lavender oil also works to reduce anxiety by improving the nervous system receptors of amino butanoic acid (Kessler et al., 2012). It was also found lavender oil vapor works to remove the odors found in some hospitals (Millot and Brand, 2001). Lavender oil is associated with emotion such as happiness and joy and has been classified as one of the oils most associated with the autonomic nervous system. (Ismaili et al., 1997). Walsh and Wilson (1999) found mixing lavender oil with rosemary increased the improvement of patients in intensive care with no negative results. They also found that people who inhaled lavender oil for a period of 3 minutes were more relaxed, less worried and increased their sleepiness (Diego et al., 1998). The use of lavender oil for a period of 10 to 15 minutes significantly improved cognition in people with dementia (Kilstoff and Chenoweth, 1998).

Lavender oil is thought to have a function like benzodiazepines (Tesserand, 1988). Benzodiazepines are the therapeutic agents most often used in the treatment of generalized anxiety disorder and insomnia and they also find utility in the treatment of panic disorder (Beaumont, et al., 1990; Uhlenhuth, et al., 1999). They can act as anticonvulsants, sedatives / hypnotics, anxiolytics, and muscle relaxants. They produce heterogeneous memory loss and are used as anesthesia or anesthesia aid (Ashton,et al., 1994). It is also prescribed to treat sleep or anxiety disorders. Lavender oil is quickly absorbed through the skin by nerves and muscles within 19 minutes after topical application with a massage (Jager et al., 1992). Linalyl acetate has been used in traditional medicine because of its anesthetic effect (Tisserand and Balacs, 1999). Lavender oil from *L. stoechas* is characterized by high content of camphor, which leads to convulsions when used at high concentrations (Tisserand and Balacs, 1999). Linalool also affects brain activity (Yamada et al., 1994). Lavender was officially recognized as a treatment when it was named by Herb Growing and Network as the United States of America Herb of the year 1999 (Evelegh, 1996). The plant is also used for decorations spices and disinfectant against germs and fungi (Hanamanthagouda et al., 2010). Lavender oil has also been used as an anti-inflammatory and anti-cramp and to reduce anxiety and stress (Hajhashemi et al., 2003; Gilani et al., 2000; Field et al., 2008). It is also an antidepressant, sedative, and immune stimulant (Cavanagh and Wilkinso, 2002; Yang et al., 2010).

As studies suggest, the use of essential oil of lavender may be safer and more effective since it has low toxicity and few side effects in the treatment of inflammatory diseases (Gosselin et al., 2011). Lavender oil was used as a pain reliever and for rheumatism (Hajhashemi et al., 2003). There are also studies on the use of lavender oil in the treatment of the heart muscle for problems caused by isoproterenol (Singal et al., 1981). Lavender oil is also used in the manufacture of

sweets, drinks, ice cream and chewing gum (Hassiottis et al., 2010). The large quantities of the residues from oil extraction from the lavender plant have also been used as fertilizer (Lubbe and Verpoorte, 2011; Schmidt, 2010).

There are three ways in which essential oils can be absorbed in the human body: 1) through the respiratory system, 2) transdermal via direct contact, and 3) oral ingestion (Perry, 2006). Some evidence indicates the absorption of essential oil has a greater effect in the bloodstream, whether by inhalation or by skin contact, in the treatment of pain, anxiety, sleep disturbances and depression (Heuberger et al., 2001). Frey et al. (2002) showed the therapeutic agents could bypass the blood-brain barrier by absorbing the oil through the sense of smell in mice. This was later confirmed in humans (Born et al., 2002; Hallschmid et al., 2004). Lavender is commonly used today to make perfumes, soaps, bath powders, talcum, and candles, scented sachets and small quantities used to flavor teas and foods, as in the French *Herbs de Provence* (Szejtli et al., 1986). The oil extracted from flowers, is considered one of the most beneficial and versatile essential oils commonly used in aromatherapy (Welsh, 1995). Lavender essential oil has also been used to treat a wide range of ailments including stress, anxiety, depression, fatigue, motion sickness and high blood pressure. Treating colic and stimulating the appetite (Duke, 1985). A mixture of lavender and peppermint essential oils is also recommended to relieve tension headache (Anonymous,1998).

Figure 2.17 Benefits of lavender essential oils (created by Keefah Al Garallaa).

General qualities of essential oils

1- Odor: Essential oils are characterized by a distinctive aromatic smell, as each oil has its own distinctive aroma.
2- Texture: All essential oils are liquid at normal temperature.
3- Color: The essential oils are primarily colorless, but some have a pale-yellow color, and some have a slight redness. This is when the oil is fresh and has not undergone oxidation or decomposition factor and has not been subjected to any abnormal factors during the process of extraction.
4- Volatilization: Essential oils are volatile substances when at normal temperatures. This distinguishes them from fixed oils that do not volatilize at normal temperatures.
5- Solubility: Essential oils melt in most organic solvents such as ether, absolute alcohol, and petroleum ether, but have low solubility in water.
6- Refractive index: Essential oils are known for their high refractive index.
7- Specific gravity: Most essential oils are lighter than water, except for some oils such as cinnamon oil and clove oil.
8- Optical rotation: Essential oils have the properties of optical rotation, through which it is possible to identify the quality of oil and its detection (Langeveld et al., 2014; Reeds et al., 2000).

Biological activity of *Lavandula* essential oil

Lavender is one of the most popular perfumes used in commercial aromatherapy (Burnett et al., 2004). However, the bioactive compounds of *Lavandula* species can be used as novel drug treatments. Antioxidant, anti-inflammatory, antibacterial, anti-fungal, antiseptic, antibacterial, anticholinergic, antiepileptic, antispasmodic, neuroprotective, anxiolytic, sedative, analgesic, hyaluronidase, and lipid-inhibitors are the most common biological activities of *Lavandula* spp. These activities have been examined *in-vitro*, and *in-vivo* in clinical trials in numerous studies (Başer and Buchbauer, 2016**).**

Table 2.3 Chemical description of Linalool.

Molecular Formula:	$C_{10}H_{18}O$
Synonyms:	(-)-Linalool 126-91-0 L-Linalool (3R)-Linalool (R)-(-)-Linalool
Molecular Weight:	154.25 g/mol

(https://pubchem.ncbi.nlm.nih.gov/compound/Linalool).

Linalool acetate belongs to the class of organic compounds known as cyclic monoterpenoids (Table. 2.3). These monoterpenes are cycle-free. Linalool acetate is characterized as a solid and is considered practically insoluble (in water), Linalool acetate is mainly located in the cytoplasm. Linalool acetate is found in all eukaryotes, from yeast to humans. Linalool acetate is a potentially toxic compound (https://pubchem.ncbi.nlm.nih.gov/compound/Linalyl-acetate).

Table 2.4 Chemical and Physical Properties of linalool

Property Name	**Property Value**
Molecular Weight	154.25 g/mol
XLogP3-AA	2.7
Hydrogen Bond Donor Count	1
Hydrogen Bond Acceptor Count	1
Rotatable Bond Count	4
Exact Mass	154.135765 g/mol
Monoisotopic Mass	154.135765 g/mol
Topological Polar Surface Area	20.2 Å²
Heavy Atom Count	11
Formal Charge	0
Complexity	154
Isotope Atom Count	0
Defined Atom Stereocenter Count	1
Undefined Atom Stereocenter Count	0
Defined Bond Stereocenter Count	0
Undefined Bond Stereocenter Count	0
Covalently Bonded Unit Count	1
Compound Is Canonicalized	Yes

(https://pubchem.ncbi.nlm.nih.gov/compound/3R_-3_7-dimethylocta-1_6-dien-3-ol.2020).

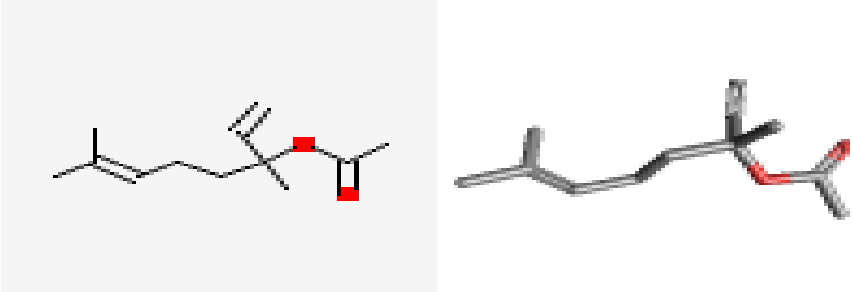

Figure 2.18 Chemical structure depiction linalyl ($C_{12}H_{20}O_2$)

Table 2.5 Chemical description of Linalyl

Molecular Formula:	$C_{12}H_{20}O_2$ or $CH_3COOC_{10}H_{17}$
Synonyms:	LINALYL ACETATE 115-95-7 Linalyl acetate Bergamiol 3,7-dimethylocta-1,6-dien-3-yl acetate
Molecular Weight:	196.29 g/mol

(https://pubchem.ncbi.nlm.nih.gov/compound/Linalyl-acetate).

Table. 2.6 Chemical and physical properties of linalyl

Property	Value
Molecular Weight	**196.29 g/mol**
XLogP3-AA	**3.3**
Hydrogen Bond Donor Count	**0**
Hydrogen Bond Acceptor Count	**2**
Rotatable Bond Count	**6**
Exact Mass	**196.14633 g/mol**
Monoisotopic Mass	**196.14633 g/mol**
Topological Polar Surface Area	**26.3 Å²**
Heavy Atom Count	**14**
Formal Charge	**0**
Complexity	**237**
Isotope Atom Count	**0**
Defined Atom Stereocenter Count	**1**
Undefined Atom Stereocenter Count	**0**
Defined Bond Stereocenter Count	**0**
Undefined Bond Stereocenter Count	**0**
Covalently Bonded Unit Count	**1**
Compound is canonicalized	**Yes**

(https://pubchem.ncbi.nlm.nih.gov/compound/3R_-3_7-dimethylocta-1_6-dien-3-ol.2020).

Literature Cited

Abbas, S. 2013. The influence of biostimulants on the growth and on the biochemical composition of *Vicia faba* cv. Giza 3 beans. Rom. Biotechnol. Lett. 18(2): 8061-8068.

Abdel-Latef, A., and A. Hamed. 2010. Effect of arabuscular mycorrhizal fungi on growth, mineral nutrition , antioxidant enzymes activity and fruit yield of tomato grown under salinity stress 127(3): 228-223.

Abdel-Latef, A.A.H.A., and M. Miransari. 2014. The role of arbuscular mycorrhizal fungi in alleviation of salt stress. In: Use of microbes for the alleviation of soil stresses. Springer, New York, NY. 23-38.

Abd-El-Malek, M., G.R. Ganger, G.R. Goodson, M.K. Reiter, and J.J. Wylie. 2005. Fault-scalable Byzantine fault-tolerant services. ACM SIGOPS Operating Systems Review. 39(5): 59-74.

Abdel-Wahab, B.F., H.A. Abdel-Aziz. and E.M. Ahmed. 2008. Convenient Synthesis and Antimicrobial Activity of New 3-Substituted 5-(Benzofuran-2-yl)-pyrazole Derivatives. Int. J. Pharm. Medicinal Chem. 341(11):734-739.

Abdul Wahid O.A., A. Mustafa, and R. Mohamed-Metwally. 2007. Enhancement of plant growth through implementation of different Trichoderma species. Proceed. Second Scientific Environ. Conf. Zagaaig Uni. 12:43-59.

Adak, T., A. Singha, K. Kumar, S.K. Shukla, A. Singh, and V. Kumar Singh. 2014. Soil organic carbon. dehydrogenase activity, nutrient availability, and leaf nutrient content as a_ected by organic and inorganic source of nutrient in mango orchard soil. J. Soil. Sci. Plant Nutr., 2: 394-406.

Adal, A.M., L.S. Sarker, A.D. Lemke, and S.S. Mahmoud. 2017. Isolation and functional characterization of a methyl jasmonate-responsive 3-carene synthase from *Lavandula* x *intermedia*. Plant Mol. Biol. 93: 641-657.

Adal, A.M., L.S. Sarker, R.P.N. Malli, P. Liang, and S.S. Mahmoud. 2019. RNA-Seq in the discovery of a sparsely expressed scent-determining monoterpene synthase in lavender (*Lavandula*). Planta. 249: 271-90.

Adam, K.L. 2006. Lavender production, products, markets, and entertainment farms. – Nat. Sustainable Agri. Inf. Service 1: 1-12.

Agriculture Research Service. 2000. Dr Duke's phytochemical and ethnobotanical databases. Duke, J. A. 2000. Dr. Duke's phytochemical and ethnobotanical databases, Phytochemical database. USDAARS-NGRL, Beltsville Agricultural Research Centre, Maryland, USA.

Agulló, E., M.A. Bustamante, C. Paredes, R. Moral, J.A. Pascual, and F. Suárez-Estrella. 2011. October. Use of functional biosolids-derived composts as growing media to produce lavender (*Lavandula dentate*) and rosemary (*Rosmarinus officinalis*). Acta Hortic. 1013: 351-357.

Ajikumar, P.K., K. Tyo, S. Carlsen, O. Mucha, T.H. Phon, and G. Stephanopoulos. 2008. Terpenoids: opportunities for biosynthesis of natural product drugs using engineered microorganisms. Mol. Pharm. 5(2): 167-190.

Akanbi, W.B., T.A. Adebayo, O.A. Togun, A.S. Adeyeye, and O.A. Olaniran. 2007. The use of compost extract as foliar spray nutrient source and botanical insecticide in *Telfairia occidentalis*. World J. Agric. Sci. 3(5): 642-652.

Akula, R., and G.A. Ravishankar. 2011. Influence of abiotic stress signals on secondary metabolites in plants. Plant signal. Behav. 6(11): 1720-1731.

Aleandri, S., D. Bandera, R. Mezzenga, and E.M. Landau. 2015. Biotinylated cubosomes: a versatile tool for active targeting and codelivery of paclitaxel and a fluorescein-based lipid dye. Langmuir. 31(46): 12770-12776.

Alexander, M. 1971**.** Microbial Ecology. Eds: John Wiley and Sons. 2000. Azotobacter Inoculation and Crop Productivity. Azotobacter in sustainable Agriculture. Ch.11 (ed.) Neeru Narula. India.

Allaby, M. 1992. The concise Oxford dictionary of zoology. New York: Oxford University Press. 340

Al-Rawi, W.A.A., Al-Hadethi, M.E.A. and Abdul-Kareem, A.A., 2016. Effect of foliar application of gibberellic acid and seaweed extract spray on growth and leaf mineral content on peach trees. The Iraqi J. Agric. Sci. 47: 98-105.

AL-Rumaih, M.M. 2007. Interactive effect of gamma radiation and gibberellic acid on soluble carbohydrate metabolism in three Plantago species. J. Food Agric. Environ. 5(3): 399-402.

Amuamuha, L., A. Pirzad, and H. Hadi. 2012. Effect of varying concentrations and time of Nanoiron foliar application on the yield and essential oil of Pot marigold. Int. Res. J. Appl. Basic Sci. 3(10): 2085-2090.

Anonymous. 1999. Lavender to be promoted in '99. Country. Side small stock J 83:102.

Anonymous. Staying ahead of headaches. 1998. new drugs and approaches offer new ways to manage pain. Drug Topics. 6: 46-51.

Aprotosoaie, A.C., E. Gille, A. Trifan, V.S. Luca, and A. Miron. 2017. Essential oils of *Lavandula* genus: a systematic review of their chemistry. Phytochem. Rev. 16: 761-799.

Armada, E., A. Roldan, and R. Azcon. 2014. “Differential Activity of Autochthonous Bacteria in Controlling Drought Stress in Native *Lavandula* and *Salvia* Plants Species under Drought Conditions in Natural Arid Soil.” Microb. Ecol. 67(2): 410-420.

Asbahani, A.E., K. Miladi, W. Badri, M. Sala, E.H. Aït-Addi, H. Casabianca, A.E. Mousadik, D. Hartmann, A. Jilale, F.N.R. Renaud, and A. Elaissari. 2015. Essential oils: from extraction to encapsulation. Int. J. Pharm. 483: 220-243.

Ashton, I.K., D.A.Walsh, J.M. Polak, and S.M. Eisenstein. 1994. Substance P in intervertebral discs: binding sites on vascular endothelium of the human annulus fibrosus. Acta Orthopaedica Scandinavica. 65(6): 635-639.

Asik, B.B., M.A. Turan, H. Celik, and AV. Katkat. 2009. Effects of humic substances on plant growth and mineral nutrients uptake of wheat (*Triticum durum* cv. Salihli) under conditions of salinity" Asian J. Crop Sci. 1(2): 87-95.

Atalay, Y., D. Avcı, and A. Başoğlu. 2008. Linear and non-linear optical properties of some donor–acceptor oxadiazoles by ab initio Hartree-Fock calculations. Struct. Chem. 19(2): 239-246.

Azcón, R., and J. Barea. 1997. Mycorrhizal dependency of a representative plant species in Mediterranean shrublands (*Lavandula spica L.*) as a key factor to its use for revegetation strategies in desertification-threatened areas. Appl. Soil Ecol. 7(1): 83-92.

Bader, B. 2012. The lavender lover’s handbook. 978(1):221-231.

Badri, M.S., A. Hani, and L. Hakimi. 2015. Effect OF Humic Acid on Performance and Nano Iron Chelated Herbs and Essential Oil of Lavender (*Lavandula angustifolia)*. Enviro. Res. 45:12-23.

Bick, J.A., and B.M. Lange. 2003. Metabolic cross talk between cytosolic and plastidial pathways of isoprenoid biosynthesis: unidirectional transport of intermediates across the chloroplast envelope membrane. Archives of biochemistry and biophysics, 415(2), pp.146-154.

Bae, H., D.P. Roberts, H.S. Lim, M.D. Strem, S.C. Park, C.M. Ryu, R.L. Melnick, and B.A. Bailey. 2011. Endophytic Trichoderma isolates from tropical environments delay disease onset and induce resistance against Phytophthora capsici in hot pepper using multiple mechanisms. Mol. Plant Microbe Interact. 24(3): 336-351.

Bakhsha, F., M. Mazandarani, M. Aryaei, and S.Y. Jafari. 2014. Phytochemical and antioxidant activity of lavandula angustifolia mill. Essential oil on preoperative anxiety in patients undergoing diagnostic curettage. Int. J. Women Health Reprod. Sci. 2(4): 268-271.

Balestrini, R., and E. Lumini. 2018. Focus on mycorrhizal symbioses. Appl. Soil Ecol. 123: 299-304.

Barata, A.M., F. Rocha, V. Lopes, E. Bettencourt, and A.C. Figueiredo. 2011. Medicinal and aromatic plants Portugal. Medicinal and aromatic plants of The World, Encyclopedia of Life Support Systems (EOLSS). Agri. Biotechnol. 45:45-59.

Barea, J.M., and C. Azcón-Aguilar. 1982. Production of plant growth-regulating substances by the vesicular-arbuscular mycorrhizal fungus Glomus mosseae. Appl. Environ. Microbiol. 43(4):8 10-813.

Barker, A.V. and M.L. Stratton. 2015. Iron. Chapter 11. In: A.V. Barker and D.J. Pilbeam. (eds): Handbook of Plant Nutrition. Second Edition. CRC Press Taylor and Francis Group. London. New York, pp: 399-426.

Barrett, S.C.H., L.D. Harder, and A.C. Worley. 1996. The comparative biology of pollination and mating in flowering plants. Philos. Trans. R. Soc. Lon. B. Biol. Sci. 351(1345): 1271-1280.

Basch, E., I. Foppa, R. Liebowitz, J. Nelson, M. Smith, D. Sollars, and C. Ulbricht. 2004. Lavender (*Lavandula angustifolia* Miller). J. Herb. Pharmacother. 4(2): 63-78.

Başer, K.H.C., and G. Buchbauer. 2016. Handbook of Essential Oils. Science, Technology and Applications, Second Edition, CRC Press, Boca Raton, Florida.

Baslam, M., and N. Goicoechea. 2012. "Water Deficit Improved the Capacity of Arbuscular Mycorrhizal Fungi (AMF) for Inducing the Accumulation of Antioxidant Compounds in Lettuce Leaves." Mycorrhiza 22 (5): 347–359.

Baydar, H., and S. Erbaş. 2007, April. Effects of harvest time and drying on essential oil properties in lavandin (*lavandula× intermedia* emeric ex loisel.). In I International Medicinal and Aromatic Plants Conference on Culinary Herbs. 826: 377-382.

Baytop, T. 1984. Phytotherapy in Turkey, past and present. Phytotherapy in Turkey (Eds. T. Baytop) Istanbul University publications, 3255: 194-195.

Beaumont, F., Jouvec, H.M., Gagnon, J., Gaillard, J. and Pelmont, J., 1990. Purification and properties of a catalase from potato tubers (*Solanum tuberosum*). Plant Sci. 72(1): 19-26.

Begum, N., C. Qin, M.A. Ahanger, S. Raza, M.I. Khan, M. Ashraf, N. Ahmed, and L. Zhang. 2019. "Role of Arbuscular Mycorrhizal Fungi in Plant Growth Regulation: Implications in Abiotic Stress Tolerance." Front. Plant Sci. 10: 1068.

Benabdelkader T, Guitton Y, Pasquier B, Magnard JL, Jullien F, Kameli A, Legendre L (2015) Functional characterization of terpene synthases and chemotypic variation in three lavender species of section *stoechas*. Physiol Plant 153:43–57

Benabdelkader, T. 2015. Functional characterization of terpene synthases and chemotypic variation in three lavender species of section *stoechas*. Physiol. Plant. 153(1): 43-57.

Benabdelkader, T., A. Zitouni, Y. Guitton, F. Jullien, D. Maitre, H. Casabianca, L. Legendre, and A. Kameli. 2011. Essential oils from wild populations of Algerian *Lavandula stoechas L.*: composition, chemical variability, and in vitro biological properties. Chem. Biodiverse. 8(5): 937-953.

Berringer Bader S (2012). The lavender lover's handbook, Timber Press. London.

Bilia, A.R., C. Guccione, B. Isacchi, C. Righeschi, F. Firenzuoli, and M.C. Bergonzi. 2014. Essential oils loaded in nanosystems: a developing strategy for a successful therapeutic approach. Evid. Complement. Alternat. Med, 1–14.

Binet, F., G. Cagnone, S. Crespo-Garcia, M. Hata, M. Neault, A. Dejda, A.M. Wilson, M. Buscarlet, G.T. Mawambo, J.P. Howard, and R. Diaz-Marin. 2020. Neutrophil extracellular traps target senescent vasculature for tissue remodeling in retinopathy. Sci. 369: 6506.

Birhane, E., F.J. Sterck, M. Fetene, F. Bongers, and T.W. Kuyper. 2012. Arbuscular mycorrhizal fungi enhance. photosynthesis, water use efficiency, and growth of frankincense seedlings under pulsed water availability conditions. Oecologia. 169(4): 895-904.

Blaize, D., J.V. Singh, A.N. Bonde, K.V. Tekale, and C.D. Mayee. 2005. Effects of farmyardmanure and fertilizers on yield, fiber quality and nutrient balance of rainfield cotton (*Gossypium hirsutum*). Biores. Technol., 96(3): 345-349.

Boachon, B., R.R. Junker, L. Miesch, J.E. Bassard, R. Höfer, R. Caillieaudeaux, D.E. Seidel, A. Lesot, C. Heinrich, J.F. Ginglinger, and L. Allouche. 2015. CYP76C1 (Cytochrome P450)-mediated linalool metabolism and the formation of volatile and soluble linalool oxides in Arabidopsis flowers: a strategy for defense against floral antagonists. The Plant Cell, 27(10): 2972-2990.

Boelens, M.H. 1995. Chemical and sensory evaluation of *Lavandula* oils. Perf. Flav. 20(3): 23 25.

Bonfante, P., and A. Genre. 2010. Mechanisms underlying beneficial plant-fungus interactions in mycorrhizal symbiosis. Nat. Commun. 1(1): 1-11.

Born, J., T. Lange, W. Kern, G.P. McGregor, U. Bickel, and H.L. Fehm. 2002. Sniffing neuropeptides: a trans nasal approach to the human brain. Nat Neurosci; 5(6): 514-516

Bose, S.K., R.K. Yadav, S. Mishra, R.S. Sangwan, A.K. Singh, B. Mishra, A.K. Srivastava, and N.S. Sangwan. 2013. Effect of gibberellic acid and calliterpenone on plant growth attributes, trichomes, essential oil biosynthesis and pathway gene expression in differential manner in Mentha arvensis L. Plant physiol. Biochem. 66: 150-158.

Bottini, R., F. Cassán, and P. Piccoli. 2004. Gibberellin production by bacteria and its involvement in plant growth promotion and yield increase. Appl. Microbiol. Biotechnol. 65(5): 497-503.

Bozorgi, H.R. 2012. Study effects of nitrogen fertilizer management under nano iron chelate foliar spraying on yield and yield components of eggplant (*Solanum melongena* L.). J. Agric. Biol. Sci. 7(4): 233-237.

Brian, P.W., H.G. Hemming, and M. Radley. 1955. A physiological comparison of gibberellic acid with some auxins. Physiol. Plant. 8(4): 899-912.

Buchbauer, G., L. Jirovetz, and W. Jäger. 1991. Aromatherapy: evidence for sedative effects of the essential oil of lavender after inhalation. Z. Naturforsch. C. 46(11-12): 1067-1072.

Buchbauer, G.W., L. Jager, J. Jirovetz, J. Ilmberger and H. 1993. Dietrich, in Bioactive Volatile Compounds from Plants, ed. R. G. Buttery and H. Sugisawa, p. 159.

Buckley, T.N. 2015. The contributions of apoplastic, symplastic and gas phase pathways for water transport outside the bundle sheath in leaves. Plant, Cell Environ. 38(1): 7-22.

Burnett, K.M., L.A. Solterbeck, and C.M. Strapp. 2004. Scent and mood state following an anxiety-provoking task. Psychol. Rep. 95(2): 707-722.

Busato, J.G., L.S. Lima, N.P. Aguiar, L.P. Canellas, and F.L. Olivares. 2012. Changes in labile phosphorus forms during maturation of vermicompost enriched with phosphorus-solubilizing and diazotrophic bacteria. Bioresour. Technol. 110: 390-395

Büyükokuroğlu, M.E., A. Gepdiremen, A. Hacimüftüoğlu, and M. Oktay. 2003. The effects of aqueous extract of Lavandula angustifolia flowers in glutamate-induced neurotoxicity of cerebellar granular cell culture of rat pups. J. Ethnopharmacol. 84: 91-94.

Cal, K., and K. Krzyzaniak. 2006. Stratum corneum absorption and retention of linalool and catecholamine induced cardiomyopathy. Can. J. Physiol. Pharmacol. 42(3): 265-267.

Caris, C., W. Hordt, H.J. Hawkins, V. Romhel, and G. Eckhard. 1998. Studies of iron transport by arbuscular mycorrhizal hyphae from soil to peanut and sorghum plants. Mycorrhiza. 8(1): 35-39.

Carvalho, D. 2011. Controle de *Fusarium oxysporum* f.sp. phaseoli in vitro e em sementes, epromoção do crescimento inicial do feijoeiro comum por Trichoderma harzianum. Trop. Plant Pathol. 36(1): 28-34.

Cavanagh, H., and J. Wilkinson. 2002. Biological activities of lavender essential oil. Phytother. Res. 16(4): 301-308.

Cavanagh, H.M., and J.M. Wilkinson. 2005. Lavender essential oil: a review. Australian Infection Control, 10(1): 35-37.

Cawthon. 1995. A review of the literature surrounding the research into aromatherapy Complement. Ther. Nurs. Midwifery, 1(4): 118-120.

Ceylan, A. 1997. Medicinal Plant II (essential oil plants). Euphtical Fac. Agri. Pub. 481: 306.

Chen, M., M. Arato, L. Borghi, E. Nouri, and D. Reinhardt. 2018. Beneficial Services of Arbuscular Mycorrhizal Fungi - from Ecology to Application. Front. Plant Sci. 9: 1270.

Chetouani, M., I. Mzabri, A. Aamar, A. Boukroute, N. Kouddane, and A. Berrichi. 2017. Effect of gibberellic acid (AG3) on the germination of seeds of *Thymus satureioides* L and *Lavandula dentate*. J. Mat. Environ. Sci. 8(3): 942-948.

Chrispeels, M.J., and J.E. Varner. 1967. Gibberellic acid-enhanced synthesis and release of α-amylase and ribonuclease by isolated barley and aleurone layers. Plant Physiol. 42(3): 398-406.

Çiğdem, K.U.Ş., and M.E. Duru. 2021. Effects of post-harvest drying times of Lavandula angustifolia and L. intermedia species on chemical components of their essential oils. Avrupa Bilim ve Teknoloji Dergisi. 21: 501-505.

Cimrin, K.M., and I. Yilmaz. 2005. Humic acid applications to lettuce do not improve yield but do improve phosphorus availability. Acta Agric. Scandin. Sec. B. Soil Plant Sci. 55(1): 58-63.

Colebrook, E.H., S.G. Thomas, A.L. Phillips, and P. Hedden. 2014. The role of gibberellin signalling in plant responses to abiotic stress. J. Exp. Biol. 217(1): 67-75.

Cooney, J., and D. Lauren. 1998.Trichoderma/pathogen interactions: Measurement of antagonistic chemicals produced at the antagonist/pathogen interface using a tubular bioassay. Lett. Appl. Microbiol., 27(5), 283-286.

Cossetin, L.F., E.M.T. Santi, J.F. Cossetin, J.B. Dillmann, M.D. Baldissera, Q.I. Garlet, T.P. De Souza, L. Loebens, B.M. Heinzmann, M.M. Machado, and S.G. Monteiro. 2018. *In vitro* safety and efficacy of lavender essential oil (Lamiales: lamiaceae) as an insecticide against houseflies (Diptera: muscidae) and blowflies (Diptera: calliphoridae). J. Econ. Entomol. 111(4): 1974-1982.

Croteau, R., T.M. Kutchan, and L.G. Norman. 2000. Natural Products (Secondary Metabolites). Biochem. Mol. Biol. Plants, 24: 1250–1318.

Cseke, L., N. Dudareva, and E. Pichersky. 1998. Structure and evolution of linalool synthase. Mol. Biol. Evol. 15(11): 1491-1498.

Da Porto, C., and D. Decorti. 2008. Analysis of the volatile compounds of flowers and essential oils from *Lavandula angustifolia* cultivated in northeastern Italy by headspace solid-phase microextraction coupled to gas chromatography-mass spectrometry. Planta. medica. 74(2): 182-187.

Danh, L.T., N.D.A. Triet, L.T.N. Han, J. Zhao, R. Mammucari, and N. Foster. 2012. Antioxidant activity yield and chemical composition of lavender essential oil extracted by supercritical CO_2. J. of Supercrit. Fluids. 70: 27-34.

Darshan, S., and R. Doreswamy. 2004. Patented anti-inflammatory plant drug development from traditional medicine. Phytother. Res. 18(5): 343-357.

Davière, J.M., and P. Achard. 2013. Gibberellin signaling in plants. Development. 140(6): 1147–1151.

Davies, J., and D. Davies. 2010. Origins and evolution of antibiotic resistance. Microbiol. Mol. Biol. Rev. 74(3): 417-433.

Deanna. 2020. Harvesting Fresh Lavender: How to Harvest, Prune and Dry Lavender Flowers, Creator Plant 14:1-9.

Deans, S.G., and M. Lis-Balchin. 1997. Food Microbiol. Int. J. 82. 759-762. (In print, 1997).

Dewick, P.M. 2002. Medicinal natural products: a biosynthetic approach. John Wiley and Sons.

Diagne, C., J.A. Catford, F. Essl, M.A. Nuñez, and F. Courchamp. 2020. What are the economic costs of biological invasions? A complex topic requiring international and interdisciplinary expertise. NeoBiota. 63: 25.

Diego, M.A., N.A. Jones, T. Field, M. Hernandez-Reif, S. Schanberg, C. Kuhn. 1998. Aromatherapy positively affects mood, EEG patterns of alertness and math computations. Int. J. Neurosci. 96(3-4): 217-224.

Doley, K., M. Borde, and M. Kulkarni. 2019. AM fungi and Trichoderma interaction for biological control of soilborne plant pathogen *Fusarium oxysporum*. Plant Microbe Interface. 95-128.

Duc, N.H., and K. Posta. 2018. Mycorrhiza induced alleviation of plant disease caused by *Clavibacter michiganensis* subsp. *michiganensis* and role of ethylene in mycorrhiza-induced resistance in tomato. Acta Biol. Hung. 69(2): 170-181.

Duc, N.H., Z. Csintalan, and K. Posta. 2018. Arbuscular mycorrhizal fungi mitigate negative effects of combined drought and heat stress on tomato plants. Plant Physiol. Biochem. 132: 297-307.

Dudareva, N., and E. Pichersky. 2008. Metabolic engineering of plant volatiles. Curr. Opin. Biotechnol. 19(2): 181-189.

Dudareva, N., E. Pichersky, and J. Gershenzon. 2004. Biochemistry of plant volatiles. Plant Physiol. 135: 1893-1902.

Duke, J., and M.J. Bogenschutz. 1994. Dr. Duke's phytochemical and ethnobotanical databases. USDA, Agricultural Research Service. 1-8.

Duke, J.A. 1985. Handbook of medicinal herbs. Boca Raton: CRC Press.

Dupuy, N., V. Gaydou, and J. Kister. 2014. Quantitative analysis of lavender (*Lavandula angustifolia*) essential oil using multiblock data from infrared spectroscopy. Am. J. Analyt. Chem., 5(10): 633-645.

Eja, M.E., G.E. Arikpo, K.H. Enyi-Idoh, and E.M. Ikpeme. 2011. An evaluation of the antimicrobial synergy of Garlic (*Allium sativum*) and Utazi (*Gongronema latifolium*) on *Escherichia coli* and *Staphylococcus aureus*. Malays. J. Microbiol. 7(1): 49-53.

Elena, B., and H.E. Daniela. 2017. Research on volatile oil for lavender species. J. Hortic. Fores. Biotechnol. 21(3): 62-67.

El-Fouly, A.F., A.F. Fawzi, and A.A. El-Sayed. 1995. March. Optimizing fertilizer use in oranges through fertigation. A case studies. In Proc. Dahlia Greidinger Int. Symp. on fertigation. 26: 303-312.

Ellis, M.B. 1960. Mycological Papers, Vol. 76 pp. 36 pp. "essential oil", Oxford English Dictionary (online, American English ed,), Retrieved 2014-07-21.

Enwall, K., Philippot, L. and Hallin, S., 2005. Activity and composition of the denitrifying bacterial community respond differently to long-term fertilization. Appl Environ. Microbiol. 71(12): 8335-8343.

Erbas, S., and H. Baydar. 2008. Effects of harvest time and drying temperature on essential oil content and composition in lavandin (*Lavandula x intermedia* Emerice x Loisel.). Turk. J. Field Crops. 13: 24-31.

Ernst, M. 2017. Lavender. In *Lavender*, Center for Crop Diversification, University of Kentucky College of Agriculture, Food and Environment, Lexington, KY.

Evans, K. 2002. Taking control of their lives? Agency in young adult transitions in England and the New Germany. J. youth stud. 5(3): 245-269.

Evelegh, T. 1996. Lavender. Lorenz Books: Sydney. extracts. J. Appl. Microbiol. 86(6): 985-990.

Fageria, N.K., F.M.P. Barbosa, A. Moreira, and C.M. Guimarães. 2009. Foliar fertilization of crop plants. J. Plant Nutr. 32(6): 1044-1064.

Fakhari, A.R., P. Salehi, R. Heydari, S.N. Ebrahimi, and P.R. Haddad. 2005. Hydrodistillation-headspace solvent microextraction, a new method for analysis of the essential oil components of *Lavandula angustifolia* Mill. J. Chromatogr. A. 1098(1-2): 14-18.

Farahani, S.M., A. Khalesi, and Y. Sharghi. 2015. Effect of nano iron chelate fertilizer on iron absorption and saffron (*Crocus sativus* L.) quantitative and qualitative characteristics. Asian J. of Biol. Sci. 45:1-9

Faust, J. 1998, December. The robustness of identified VAR conclusions about money. In Carnegie-Rochester conference series on public policy. North-Holland. 49: 207-244.

Fernandez, V., T. Sotiropoulos and P. Brown. 2013. Foliar Fertilization: Scientific Principles and Field Practices. First ed., International Fertilizer Industry Association (IFA), Paris, France. G. Zheng. Patent. A.

Fernandez-Bayo, J.D., R. Nogales, and E. Romero. 2009. Assessment of three vermicomposts as organic amendments used to enhance diuron sorption in soils with low organic carbon content. Eur. J. Soil Sci., 60(6): 935-944.

Ferrara, G., and G. Brunetti. 2010. Effects of the times of application of a soil humic acid on berry quality of table grape (*Vitis vinifera* L.) cv Italia. Span. J. Agric. Res. 3: 817-822.

Festing, S.1989. The Story of Lavender, 2nd edn, Heritage in Sutton Leisure: London.

Field T., T. Field, C. Cullen, S. Largie, M. Diego, and S. Schanberg. 2008. Lavender bath oil formation of volatile and soluble linalool oxides in Arabidopsis flowers: a strategy for from a massage oil. J. Soc. Cosmetic Chem. 43: 49-54.

Figueiredo-Pereira, M.E., K.A. Berg, and S. Wilk. 1994. A new inhibitor of the chymotrypsin-like activity of the multicatalytic proteinase complex (20S proteasome) induces accumulation of ubiquitin-protein conjugates in a neuronal cell. J. neurochem. 63(4): 1578-1581.

Fleet, C.M. and T.P. Sun. 2005. A DELLAcate balance: the role of gibberellin in plant morphogenesis. Curr. Opin. in plant biol. 8(1): 77-85.

Flores, G., G.P. Blanch, M.L. Ruiz-del-Castillo, and M. Herraiz. 2005. Enantiomeric composition studies in *Lavandula* species using supercritical fluids. J. Sep. Sci. 28(17): 2333-2338.

Foo, K.Y., L.K. Lee, and B.H. Hameed. 2013. Preparation of activated carbon from sugarcane bagasse by microwave assisted activation for the remediation of semi-aerobic landfill leachate. Bioresour. Technol. 134: 166-172.

FuJiu, C. and W. Qingsheng, W. 1995. Physiological effects of humic acids on drought resistance of wheat. Ying Yong Sheng tai xue bao, J. Appl. Ecol. 6(4): 363-367.

Fujiu, C., Y. Daoqi, and W. Qingsheng. 1995. "Physiological effects of humic acid on drought resistance of wheat (in Chinese), " Yingyong Shengtai Xuebao 6, 363-367. http://agris.fao.org/agris - search/search.do?recordID=CN9611622.

Gamez MJ, J. Jimenez, C. Navarro, A. Zarzuelo. (1990): Study of the essential oil of *Lavandula dentata L*. Pharmazie 45: 69.

Garbaye, J. 1991. Biological interactions in the mycorrhizosphere. Experientia. 47(4): 370-375.

Ghasemi, S., A.H. Khoshgoftarmanesh, M. Afyuni, and H. Hadadzadeh. 2014. Iron(II)–amino acid chelates alleviate damages salt-stress induced oxidative on tomato grown in nutrient solution culture. Sci. Hortic. 165: 91-98.

Ghazanfar, S.A. 1994. Handbook of Arabian medicinal plants. CRC press.

Ghorbani. S., H.R. Khazaei, M. Kafi, and M. Banayan-Aval. 2010**.** The effect adding humic acid to irrigation water on yield and yield components of corn . J. Agric. Ecol. 2: 123-131.

Gianinazzi-Pearson, V., Gianinazzi, S.: The physiology of vesicular-arbuscular mycorrhizal roots. - Plant Soil 71: 197-209, 1983.

Gianinazzi-Peason, V., and Gianinazzi. 1983. The physiology of vesicular-arbuscular mycorrhizal roots. Plant Soil, 71:197-209.

Giannakoula, A.E., I.F. Ilias, J.J.D. Maksimović, V.M. Maksimović, and B.D. Živanović. 2012. The effects of plant growth regulators on growth, yield, and phenolic profile of lentil plants. J. Food Compos. Anal. 28(1): 46-53.

Gilani, A.H., N. Aziz, S.M. Ali, and M. Saeed. 2000. Pharmacological basis for the use of peach leaves in constipation. J. Ethnopharmacol. 73(1-2): 87-93.

Goel, N., H. Kim, and R.P. Lao. 2005. An olfactory stimulus modifies nighttime sleep-in young men. Chronobiol. Int. 22(5): 889-904.

Gomi, K., and M. Matsuoka. 2003. Gibberellin signalling pathway. Curr. Opin. Plant Biol. 6(5): 489-493.

Gong, Z, W. Qing., Z. Rong and W.R Jun. 2001. Effect of different VA mycorrhiza fungi on the growth of potato. Acto Agriculture Boreull Sinica .14:12-19.

Gosselin, P.A., and J.P. Gagné. 2011. Older adults expend more listening effort than young adults recognizing speech in noise. 50(11): 786-792.

Gouyon, P.H., P. Vernet, J. L. Guillerm, and G. Valdeyron. 1986. "Polymorphisms and environment: the adaptive value of the oil polymorphisms in Thymus vulgaris L." Heredity. 57(1): 59-66.

Raut, J. S., & Karuppayil, S. M. (2014). A status review on the medicinal properties of essential oils. Indust. Crops Products. 62, 250-26

Gryndler, M., J. Larsen, H. Hrselova, V. Rezacova, H. Gryndlerova, and J. Kubat. 2006. Organic and mineral fertilization, respectively, increase and decrease the development of external mycelium of arbuscular mycorrhizal fungi in a long-term field experiment. Mycorrhiza. 16(3): 159-166.

Guitton, Y., F. Nicolè, S. Moja, N. Valot, S. Legrand, F. Jullien, L. Legendre. 2010. Differential accumulation of volatile terpene and terpene synthase mRNAs during lavender (*Lavandula angustifolia* and *L. x intermedia*) inflorescence development. Physiol. Plant. 138(2): 150-163.

Gupta, G.R., D. Tripathi, and H.E. Mason. 2015. Spectroscopic observations of a coronal loop: basic physical plasma parameters along the full loop length. ApJ. 800(2): 140.

Habeeb, T.H., M. Abdel-Mawgoud, R.S. Yehia, A.M.A. Khalil, A.M. Saleh, and H.A. Elgawad. 2020. Interactive impact of arbuscular mycorrhizal fungi and elevated co2 on growth and functional food value of soil science and plant nutrition 7 *Thymus Vulgare*. *Thymus Vulgare*. J. Fungus. 6(3): 168.

Habibzadeh, Y., A. Pirzad, M.R. Zardashti, J. Jalilian, and O. Eini. 2013. Effects of Arbuscular Mycorrhizal Fungi on Seed and Protein Yield under Water-deficit Stress in Mung Bean. J. Agron. 105(1): 79-84.

Hagvall, L., M. Sköld, J. Bråred-Christensson, A. Börje, and A.T. Karlberg. 2008. Lavender oil lacks natural protection against autoxidation, forming strong contact allergens on air exposure. Contact Derm. 59(3):143-150.

Hajhashemi, V., A. Ghannadi, and B. Sharif. 2003. Anti-inflammatory and analgesic properties of the leaf extracts and essential oil of *Lavandula angustifolia* Mill. J. Ethnopharmacol. 89(1): 67-71.

Hajhashemi, V., A. Ghannadi, and B. Sharif. 2003. Anti-inflammatory and analgesic properties of the leaf extracts and essential oil of *Lavandula angustifolia* Mill. J. ethnopharmacol. 89(1), pp.67-71.

Hajisamadi Asl, B., M.B. Hassanpouraghdam, and A. Khalighi. 2011. Effects of Gibberellic Acid (GA3) Foliar Application on Growth Characteristics and Essential Oil of Lavender (*Lavandula officinalis* Chaix.). J. Agric. Sci. Sust prod. 21(2): 23-32.

Hallschmid, M., C. Benedict, J. Born, H.L. Fehm, and W. Kern. 2004. Manipulating central nervous mechanisms of food intake and bodyweight regulation by intranasal administration of neuropeptides in man. Physiol. Behav. 83(1): 55-64

Hamel, C., C. Neeser, U. Barrantes-Cartin, and D.L. Smith. 1991. Endomycorrhizal fungal species mediate ISN-transfer from soybean to corn in non-fumigated soil. Plant and soil. 138(1): 41-47.

Hanamanthagouda, M.S., S.B. Kakkalameli, P.M. Naik, P. Nagella, H.R. Seetharamareddy, and H.N. Murthy. 2010. Essential oils of *Lavandula bipinnata* and their antimicrobial activities. Food Chem. 118(3): 836-839.

Harborne, J.B.1973. Photochemical methods: A guide to Modern Technique of Plant Analysis. 1st ed. , Cox & Wyman , London.

Harborne, J.B.1998. Phytochemical Methods. 3rd ed., Chapman & Hall.

Hariharan, G., K. Kannan, and K.R. Sharma. 2009. Haar wavelet method for solving Fisher's equation. Appl. Math. Comput. 211(2): 284-292.

Harman, G.E., C.R. Howell, A. Viterbo, I. Chet, and M. Lorito. 2004. Trichoderma species—opportunistic, avirulent plant symbionts. Nat. Rev. Microbiol. 2(1): 43-56.

Harper, S.M., G.L. Kerven, D.G. Edwards, and Z. Ostatekboczyski. 2000. Characterization on fulvik and humic acids from leaves of (*Eucalyptus comaldulensis*) and from decomposed hay. Soil Biol. Biochem., 32(10): 1331-1336.

Hartwigson, I.A., and M.R.E. Vans. 2000. Humic acid seed and substrate treatments promote seedling root development. Hort. Science. 35(7):1231-1233.

Hassanpouraghdam, M.B., A. Hassani, L. Vojodi. 2011. Essential oil constituents of *Lavandula officinalis* Chaix from Northwest Iran. Chemija. 22(3): 167-171.

Hassiotis, C.N., D.M. Lazari, and K.E. Vlachonasios. 2010.The effects of habitat type and diurnal harvest on essential oil yield and composition of *Lavandula angustifolia* Mill. Environ. Bul. 19(8):1491-1498.

Hassiotis, C.N., F. Ntana, D.M. Lazari, S. Poulios, and K.E. Vlachonasios. 2014. Environmental and developmental factors affect essential oil production and quality of *Lavandula angustifolia* during flowering period. Ind. Crop. Prod. 62: 359-366.

Hay, R., 1998. Sense of place in developmental context. J. Environ. Psychol. 18(1): 5-29.

Hay, R.K.M., and P.G. Waterman. 1993.Volatile oil Crop and Longman Sci. and Tech. 181 p., New York.

Heller, R., R. Esnault, and C. Lance. 2006. Physiologie végétale. 6eme Ed de l'Abrégé. Éditions Dunod, Paris, France, p 294.

Herrera, C.M., I.M. García, and R. Pérez. 2008. Invisible floral larcenies: microbial communities degrade floral nectar of bumble bee-pollinated plants. Ecol. 89(9): 2369-2376.

Heuberger, M., M. Zäch, and N.D. Spencer. 2001. Density fluctuations under confinement: when is a fluid not a fluid? Sci. 292(5518): 905-908.

Hosseinzadeh, H. and S. Parvardeh. 2004. Anticonvulsant effects of thymoquinone, the major constituent of *Nigella sativa* seeds, in mice. Phytomed. 11(1): 56-64.

Hosseinzadeh, H., M. Ramezani, and N. Namjo. 2003. Muscle relaxant activity of *Elaeagnus angustifolia L.* fruit seeds in mice. J. Ethnopharmacol. 84(2-3): 275-278.

Houtsma, M.T. 1993. EJ Brill's First Encyclopaedia of Islam, Brill. 5: 1913-1936.

Huber, D.P.W., S. Ralph, and J. Bohlmann. 2004. Genomic hardwiring and phenotypic plasticity of terpenoid-based defense in conifers. J. Chem. Ecol. 30(12): 2399-2418.

Hudson, R., 1996. The value of lavender for rest and activity in the elderly patient. Complement. Ther. Med. 4(1): 52-57.

Hugoni, M., P. Luis, J. Guyonnet, and F.Z. Haichar. 2018. Plant host habitat and root exudates shape fungal diversity. Mycorrhiza. 28(5): 451-463.

Hui, L., L. He, L. Huan, L. XiaoLan, and Z. AiGuo. 2010. Chemical composition of lavender essential oil and its antioxidant activity and inhibition against rhinitis-related bacteria. Afr. J. Microbiol. Res. 4(4): 309-313.

Iijima, Y., R. Davidovich-Rikanati, E. Fridman, D.R. Gang, E. Bar, E. Lewinsohn, and E. Pichersky. 2004. The biochemical and molecular basis for the divergent patterns in the biosynthesis of terpenes and phenylpropenes in the peltate glands of three cultivars of basil. Plant physiol. 136(3): 3724-3736.

Imtiaz, M., A. Rashid, M.Y. Memon, and M. Aslam. 2010. The role of micronutrients in crop production and Human health. Pak. J. Bot. 42(4): 2565-2578.

Iqbal, M., and M. Ashraf. 2013. Gibberellic acid mediated induction of salt tolerance in wheat plants: Growth, ionic partitioning, photosynthesis, yield and hormonal homeostasis. Environ. Exp. Bot. 86: 76-85.

Iqbal, N., R. Nazar, M.I.R. Khan, A. Masood, and N.A. Khan. 2011. Role of gibberellins in regulation of source–sink relations under optimal and limiting environmental conditions. Curr. Sci. 100(7): 998-1007.

Ismaili, M.A., and P. Bernard. 1997. Asymptotic analysis and linearization of the randomly perturbed two-wells Duffing oscillator. Probabilistic Eng. Mech. 12(3): 171-178.

Jackson, M.B. 1993. Are plant hormones involved in root to shoot communication? Adv. Bot. Res. 19: 103-187.

Jager, W., G. Buchbauer, L. Jirovetz, and M. Fritzer. 1992. Percuta-neous absorption of lavender oil. 43(1): 49-54.

Jalayerinia, N., S.M.N. Kalat, and R.S. Abadi-Haghighi. 2017. The effect of vermicompost and spraying with humic acid and fertilizer on quantitative and qualitative characteristics of rosemary. Asian J. Biol. Life Sci. 6(1).

Joe, G.W., D.D. Simpson, and K.M. Broome. 1999. Retention and patient engagement models for different treatment modalities in DATOS. Drug Alcohol Depend. 57(2): 113-125.

Johnson, N.C., J.H. Graham, and F.A. Smith. 1997. Functioning of Mycorrhizal Associations along the Mutualism Parasitism Continuum. New Phytol. 135(4): 575-586.

Journet, E., Y. Balkanski, and S.P. Harrison. 2014. A new data set of soil mineralogy for dust cycle modeling. Atmos. Chem. Phys., 14(8): 3801-3816.

Jung, J., S. Sewenig, U. Hener, and A. Mosandl. 2005. Comprehensive authenticity assessment of lavender oils using multielement/multicomponent isotope ratio mass spectrometry analysis and enantioselective multidimensional gas chromatography–mass spectrometry. Eur. Food Res. Technol. 220(2): 232-237.

Kaka, G., K. Yaghoobi, S. Davoodi, S.R. Hosseini, S.H. Sadraie, and K. Mansouri. 2016. Assessment of the neuroprotective effects of *Lavandula angustifolia* extract on the contusive model of spinal cord injury in Wistar rats. Front. Neurosci. 10:25.

Kako, T., N. Kikugawa, and J. Ye. 2008. Photocatalytic activities of AgSbO3 under visible light irradiation. Catal. Today, 131(1-4): 197-202.

Kariali, E., and P.K. Mohapatra. 2007. Hormonal regulation of tiller dynamics in differentially–tillering rice cultivars. Plant Growth Regul. 53(3): 215 –223.

Kashani, M., M. Tavirani, S. Talaei and M. Salami. 2011. Aqueous extract of lavender (*Lavandula angustifolia*) improves the spatial performance of a rat model of Alzheimer's disease. Neurosci. Bull. 27(2): 99-106.

Kasper, S., M. Gastpar, W.E. Müller, H.P. Volz, H.J. Möller, A. Dienel, and S. Schläfke. 2010. Silexan, an orally administered *Lavandula* oil preparation, is effective in the treatment of 'subsyndromal' anxiety disorder: A randomized, double-blind, placebo-controlled trial. Int. Clin. Psychopharmacol. 25(5): 277-87.

Kaviani, B., N.A.R. Negahd, and M.V.F. Ghazinani. 2014. The effect of Iron nano-chelate and Cycocel on some morphological and physiological characteristics, proliferation and enhancing the quality of *Euphorbia pulcherrima.* Sci. Papers Ser. B Hortic. 58: 337-342.

Kenner, C., 1998. National coalition for health professional education in genetics. AACN clinical issues, 9(4): 582-587.

Kessler, R.C., M. Petukhova, N.A. Sampson, A.M. Zaslavsky, and H.U. Wittchen. 2012. Twelve-month and lifetime prevalence and lifetime morbid risk of anxiety and mood disorders in the United States. Int. J. Methods Psychiatric Res. 21(3): 169-184.

Keurentjes, J.J., G.C. Angenent, M. Dicke, V.A.M. Dos Santos, J. Molenaar, W.H. Putten, P.C. de Ruiter, P.C. Struik, and B.P. Thomma. 2011. Redefining plant systems biology: from cell to ecosystem. Trends Plant Sci. 16(4): 183-190.

Khan, N.A. and Samiullah. 2003. Comparative effects of modes of gibberellic acid application on photosynthetic biomass distribution and productivity of rapeseed-mustard. Physiol. Mol. Biol. Plants. 9: 141–145.

Khan, P., M.Y. Memon, M. Imatiaz, and M. Aslam. 2009. Response of wheat to foliar and soil application of urea at different growth stages. Pakistan J. Bot. 41(3): 1197-1204.

Khazaie, H.R., E.E. Rezaie, and M. Bannayan. 2011. Application times and concentration of humic acid impact on aboveground biomass and oil production of hyssop (*Hyssopus officinalis*). J. Med. Plants Res. 5(20): 5148-5154.

Khosravi, R. and J.J. Sendi. 2013. Effect of neem pesticide (Achook) on midgut enzymatic activities and selected biochemical compounds in the hemolymph of lesser mulberry pyralid, *Glyphodes pyloalis* Walker (Lepidoptera: Pyralidae). J. Plant Prot. Res. (3): 53.

Kiecolt-Glaser, J.K., J.E. Graham, W.B. Malarkey, K. Porter, S. Lemeshow, and R. Glaser. 2008. Olfactory influences on mood and autonomic, endocrine, and immune function. Psychoneuroendocrinology. 33(3): 328-339.

Kilstoff, K., and L. Chenoweth. 1998. New approaches to health and well-being for dementia day-care clients, family careers and day-care staff. Int. J. Nurs. Pract. 4(2): 70-83.

Kimbrough, K.A. and C.E. Swift. 2009. Growing lavender in Colorado (Doctoral dissertation, Colorado State University. Libraries).

King, A.J. and J.R. Dickinson. 2003. Biotransformation of hop aroma terpenoids by ale and lager yeasts. FEMS Yeast Research, 3(1): 53-62.

Kipngeno, P., T. Losenge, N. Maina, E. Kahangi, and P. Juma. 2015. Efficacy of Bacillus subtilis and Trichoderma asperellum against *Pythium aphanidermatum* in tomatoes. Biol. Control. 90: 92-95.

Kostadinov, K., and S. Kostadinova. 2014. Nitrogen efficiency in eggplants (*Solanum melongena* L.) depending on fertilizing. Bulg. J. Agric. Sci. 20(2): 287-292.

Kovacheva, P., K. Arishtirova, and S. Vassilev. 2001. MgO/NaX zeolite as basic catalyst for oxidative methylation of toluene with methane. Appl. Catal. A-Gen. 210(1-2): 391-395.

Kreis, P. and A. Mosandl. 1992. Chiral compounds of essential oils. Part XI. Simultaneous stereoanalysis of Lavandula oil constituents. Flavour Fragr. J. 7(4): 187-193.

Kreis, P., and A. Mosandl. 1992. Chiral compounds of essential oils. Part XI. Simultaneous stereoanalysis of lavandula oil constituents. Flavour Fragr. J. 7(4): 187-193.

Kwon, C.T., S.H. Kim, D. Kim, and N.C. Paek. 2015. The rice floral repressor Early flowering1 affects spikelet fertility by modulating gibberellin signaling. Rice. 8(1): 1-11.

Lahlou, M. 2004. Essential oils and fragrance compounds: bioactivity and mechanisms of action. Flavour Fragr. J. 19(2): 159-165.

Lai, P.K. 2004. Antimicrobial and chemopreventive properties of herbs and spices. Curr Med Chem., 11(11): 1451-60.

Lakshmana, M. 2000. Azotobacter Inoculation and Crop Productivity. In Azotobacter in sustainable Agriculture. Ch.11 (ed.) Neeru Narula. India.

Lakušic, B., D. Lakušic, M. Ristic, M. Marčetic, and V. Slavkovska. 2014. Seasonal variations in the composition of the essential oils of *Lavandula angustifolia* (Lamiacae). Nat. Prod. Commun. (6): 9.

Landmann, C., B. Fink, M. Festner, M. Dregus, K.H. Engel, and W. Schwab. 2007. Cloning and functional characterization of three terpene synthases from lavender (*Lavandula angustifolia*). Arch. Biochem. Biophys. 465(2): 417-429.

Lane, A., A. Boecklemann, G.N. Woronuk, L. Sarker, and S.S. Mahmoud. 2010. A genomics resource for investigating regulation of essential oil production in *Lavandula angustifolia*. Planta. 231(4): 835-845.

Laule, O., A.Fürholz, , H.S.Chang, , T.Zhu, , X.Wang, , P.B.Heifetz, , W.Gruissem, and M.Lange, , 2003. Crosstalk between cytosolic and plastidial pathways of isoprenoid biosynthesis in Arabidopsis thaliana. Proceedings of the National Academy of Sciences, 100(11), pp.6866-6871.

Langenheim, J.H. 1994. Higher plant terpenoids: a phytocentric overview of their ecological roles. J. Chem. Ecol., 20(6): 1223-1280.

Langeveld, W.T., E.J. Veldhuizen, and S.A. Burt. 2014. Synergy between essential oil components and antibiotics: a review. Crit. Rev. Microbiol. 40(1): 76-94.

Lawrence, B.M. 2004. Progress in essential oils. Perfume & Flavorist 29:70–91.

Leonardo, A.G. 2008. Humic Acid: 100% Natural, Many Uses. Golden Harrest Organic., LLCTM.

Lesage-Meessen, L., M. Bou, J.C. Sigoillot, C. Faulds and A. Lomascolo. 2015. Essential oils and distilled straws of lavender and lavandin: a review of current use and potential application in white biotechnology. Appl. Microbiol. Biotechnol. 99(8): 3375-3385.

Li, C., P. Wang, N.W. Menzies, E. Lombi, and P.M. Kopittke. 2017. Effects of changes in leaf properties mediated by methyl jasmonate (MeJA) on foliar absorption of Zn, Mn, and Fe. Ann. Bot. 120(3): 405-415.

Li, T., Y.J. Hu, Z.P. Hao, H. Li, Y.S. Wang, and B.D. Chen. 2013. First cloning and characterization of two functional aquaporin genes from an arbuscular mycorrhizal fungus *Glomus Intraradices*. New Phytol. 197(2): 617-630.

Lis Balchin, M., and S. Hart. 1999. Studies on the mode of action of the essential oil of lavender (*Lavandula angustifolia* P. Miller). Phytother .Res. 13(6): 540-542.

Lis, M.B., S. Hart, S.G. Deans and J.E. Eaglesham. 1995. Herbs Spices Med. Plants, 3 11.

Lis-Balchin, M. 2002. Lavender: The Genus Lavandula. Taylor and Francis, London, 283 pp.

Sienkiewicz, M., M. Lysakowska, J. Ciecwierz, P. Denys and E. Kowalczyk, 2011. Antibacterial activity of thyme and lavender essential oils. Med. Chem. 7(6): 674-689.

Lobartini, J.C., G.A. Orioli, and K.H. Tan. 1997. Characteristics of soil humic acid frations separated by ultrafiltration. Com. Soil Sci. Plant Anal. 28(9-10): 787-796.

Lu, X.H., and R.T. Koide. 1994. The effect of mycorrhizal infection on components of plant-growth and reproduction. New Phytol. 128(2): 211-218.

Lubbe, A., and R. Verpoorte. 2011. Cultivation of medicinal and aromatic plants for specialty industrial materials. Ind. Crop Prod. 34(1): 785-801.

Lucy, M., E. Reed, and B.R. Glick. 2004. Applications of free-living plant growth promoting rhizobacteria. Antonie van Leeuwenhoek. J. Microbiol. 86(1): 1-25.

Machado, M.P., A.L.L. Silva, and L.A. Biasi. 2011. Effect of plant growth regulators on *In vitro* regeneration of *Lavandula dentata* L. shoot tips. J. Biotec. Biodivers. 2(3): 28-31.

Mäder, P., F. Andreas, D. David, G. Lucie, F. Padruot, and U. Niggli1. 2002. Soil Fertility and Biodiversity in Organic Farming. Sci. 296(5573): 1694-1697.

Marotti, M., R. Piccaglia, and E. Giovanelli. 1996. Differences in essential oil composition of basil (*Ocimum basilicum* L.) Italian cultivars related to morphological characteristics. J. Agr. Food Chem. 44(12): 3926-3929.

Marth, P.C., W.V. Audia, and J.W. Mitchell. 1956. Effects of gibberellic acid on growth and development of plants of various genera and species. Botanical Gazette. 118(2): 106-111.

Mataroiev, I.A. 2002. Effect of humate on diseases plant resistance. Ch. Agri. J. 1: 15-16.

Mathur, N., and A. Vyas. 1995. I. Influence of VA mycorrhizae on net photosynthesis and transpiration of *Ziziphus mauritiana*. J. Plant Physiol. 147(3-4): 328-330.

Mbarga, J.B., G.M.T. Hoopen, J. Kuate, A. Adiobo, M.E.L. Ngonkeu, Z. Ambang, A. Akoa, P.R. Tondje, and B.A.D. Begoude. 2012. *Trichoderma asperellum*: a potential biocontrol agent for *Pythium myriotylum*, causal agent of cocoyam (*Xanthosoma sagittifolium*) root rot disease in Cameroon. Crop Protect. 36: 18-22.

McGimpsey, J.A. and N.G. Porter. 1999. Lavender: A Growers' Guide for Commercial Production. Publications Section, New Zealand Institute for Crop and Food Research Limited.

Millot J.L., and G. Brand. 2001. Effects of pleasant and unpleasant ambient odors on human voice pitch. Neurosci. Lett. 297(1): 61-63.

Miri, A., M. Sarani, M.R. Bazaz, and M. Darroudi. 2015. Plant-mediated biosynthesis of silver nanoparticles using *Prosopis farcta* extract and its antibacterial properties. Spectrochim. Acta A Mol. Biomol. Spectrosc. 141: 287-291.

Misra A.N., D. Latowski, and K. Strzalka. 2006. The xanthophylls cycle activity in kidney bean and cabbage leaves under salinity stress. Russian J. Plant Physiol. 53(1): 102-109.

Mohandas, S., R. Manjula, R.D. Rawal, H.C. Lakshmikantha, S. Chakraborty, and Y.L. Ramachandra. 2010. Evaluation of arbuscular mycorrhiza and other biocontrol agents in managing *Fusarium oxysporum* f. sp. cubense infection in banana cv. Neypoovan. Biocont. Sci. Technol. 20(2): 165-181.

Mullen, W., C.A. Edwards, and A. Crozier. 2006. Absorption, excretion, and metabolite profiling of methyl-, glucuronyl-, glucosyl-and sulpho-conjugates of quercetin in human plasma and urine after ingestion of onions. Br. J. Nutr. 96(1): 107-116.

Munoz-Bertomeu, J., B. Cascales-Minana, J.M. Mulet, E. Baroja-Fernández, J. Pozueta-Romero, J.M. Kuhn, J. Segura, and R. Ros. 2009. Plastidial glyceraldehyde-3-phosphate dehydrogenase deficiency leads to altered root development and affects the sugar and amino acid balance in Arabidopsis. Plant Physiol. 151(2), pp.541-558.

Murphy, B.R., L. Martin-Nieto, F. Doohan, and T. Hodkinson. 2015. Fungal endophytes enhance agronomically important traits in severely drought-stressed barley. J. Agron. Crop Sci. 201(6): 419–427.

Muthukumar, A., A. Eswaran, and K. Sanjeevkumas. 2011. Exploitation of Trichoderma species on the growth of Pythium aphanidermatum in chilli. Braz. J. Microbiol. 42(4): 1598-1607

Naðalin, V., Z. Lepojeviæ, M. Ristiæ, J. Vladiæ, B. Nikolovski, and D. Adamoviæ. 2014. Investigation of cultivated lavender (Lavandula officinalis L.) extraction and its extracts. Chem. Ind. Chem. Eng. Q., 20(1): 71-86.

Nair, M., Nancy, D., Krishnan, A.G., Anjusree, G.S., Vadukumpully, S. and Nair, S.V., 2015. Graphene oxide nanoflakes incorporated gelatin–hydroxyapatite scaffolds enhance osteogenic differentiation of human mesenchymal stem cells. Nanotechnol. 26(16): 161001.

Nair, R.P., P. Stuart, T. Henseler, S. Jenisch, N.V. Chia, E. Westphal, N.J. Schork, J. Kim, H.W. Lim, E. Christophers, J.J. Voorhees, and J.T. Elder. 2000. Localization of psoriasis-susceptibility locus PSORS1 to a 60-kb interval telomeric to HLA-C. Am. J. Hum. Genet. 66(6): 1833-1844.

Nascimento, G.G.F., J. Locatelli, P.C. Freitas, and G.L. Silva. 2000. Antibacterial activity of plant extracts and phytochemicals on antibiotic-resistant bacteria. Braz. J. Microbiol. 31(4): 247-256.

Naseby, D.C., J.A Pascual, and J.M Lynch.2000. Effect of biocontrol strains of Trichoderma on plant growth, Pythium ultimum population,soil microbial communities and soil enzyme. activities. J. Appl. Microbiol. 88(1): 161-169.

Nasiri, Y., S. Zehtabe-Salmasi, S. Nasrollahzade, N. Najafi, and K. Ghassemi- Golezani. 2010. Effect of foliar application of micronutrient (Fe and Zn) on flower yield and essential oil of chamomile (Matricaria chamomila L.). J. Med. Plants Res. 4(17): 1733-1737.

Neware, S., I. Yadav, and B. Meena. 2017. Effect of plant growth regulators and micronutrients on growth and yield of Sweet Orange (*Citrus sinensis* L. Osbeck) cv. Mosambi. Chem. Sci. 6(21): 213-218.

Ngumbi, E., and J. Kloepper. 2016. Bacterialmediated drought tolerance: Current and prospects. Appl. Soil Ecol. 105: 109-125.

Noroozisharaf, A., and M. Kaviani. 2018. Effect of soil application of humic acid on nutrients uptake, essential oil and chemical compositions of garden thyme (*Thymus vulgaris L.*) under greenhouse conditions. Physiol. Mol. Biol. Plants, 24(3): 423-431.

Nuvale, M.U., S.A. Aklade, J.R. Desai, and P.V. Nannvare. 2010. Influence of plant growth regulators on growth, flowering and yield of chrysanthemum cv.'IIHR-6. Int J. Pharma Bio Sci. 1: 2.

Oliveira, P. et al. 2016. Resposta de fungos e processos biológicos do solo a resíduos vegetais em sistema plantio direto. Pesqui. Agropecu. Trop. 46(1): 57-64.

Orcutt, D.M. and E.T. Nilsen. 2000. Physiology of plants under stress: Soil and biotic factors (Vol. 2). John Wiley & Sons.

Ouahmane, L., M. Hafidi, C. Plenchette, M. Kisa, A. Boumezzough, J. Thioulouse, and R. Duponnois. 2006. *Lavandula* species as accompanying plants in Cupressus replanting strategies: effect on plant growth, mycorrhizal soil infectivity and soil microbial catabolic diversity. Appl. Soil Ecol. 34(2-3): 190-199.

Peres-Montano, F. et al. 2014. Plant growth promotion in cereal and leguminous agricultural important plants: from microorganism capacities to crop production. Microbiol. Res. 169(5): 325-336.

Perry, N., and E. Perry. 2006. Aromatherapy in the management of psychiatric disorders clinical and neuropharmacological perspectives. CNS Drugs. 20(4): 257-280.

Phelpstek, L.D. 2002. Structure, properties, and soil Applicant. http://www.com/ clints/humic acid.html. 14: 30-39.

Phillips, P.J., H. Wechsler, J. Huang, and P.J. Rauss. 1998. The FERET database and evaluation procedure for face-recognition algorithms. Image Vis. Comput. 16(5): 295-306.

Piccaglia, R., M. Marotti, E. Giovanelli, S.G. Deans, and E. Eaglesham. 1993. Antibacterial and antioxidant properties of Mediterranean aromatic plants. Ind. Crop Prod. 2(1): 47-50.

Piccolo, A., G. Celano, and G. Pietramellara. 1993. Effects of fractions of coal-derived humic substances on seed germination and growth of seedlings (*Lactuga sativa* and *Lycopersicum esculentum*). Biol. Fertil. Soils. 16(1): 11-15.

Pimentel, D., H. Paul, H. James, D. David, and S. Rita. 2005. "Environmental, energetic, and economic comparisons of organic and conventional farming systems". BioScience. 55(7): 573-582.

Pinto, J.G., E.L. Fröhlich, G.C. Leckebusch, and U. Ulbrich. 2007. Changing European storm loss potentials under modified climate conditions according to ensemble simulations of the ECHAM5/MPI-OM1 GCM. Nat. Hazards Earth Syst. Sci. 7(1): 165-175.

Pirzad, A., and S. Mohammadzadeh. 2018. "Water Use Efficiency of Three Mycorrhizal Lamiaceae Species (*Lavandula Officinalis, Rosmarinus Officinalis* and *Thymus Vulgaris*)." Agric. Water Manag. 204: 1-10.

Pozo, M.J., J.A. López-Ráez, C. Azcón-Aguilar, and J.M. García-Garrido. 2015. Phytohormones as integrators of environmental signals in the regulation of mycorrhizal symbioses. New Phytol. 205(4): 1431-1436.

Prakash, B., A. Kedia, P.K. Mishra, N.K. Dubey. 2015. Plant essential oils as food preservatives to control moulds, mycotoxin contamination and oxidative deterioration of agri-food commodities Potentials and challenges. Food Cont. 47: 381-391.

Prusinowska, R., and K.B. Emigielski. 2014. Composition, biological properties, and therapeutic effects of lavender (*Lavandula angustifolia* L). A review. Herba Pol. 60(2): 56-66.

Qi, W.Z., and L. Zhao. 2013. Study of the siderophore-producing *Trichoderma asperellum* Q1 on cucumber growth promotion under salt stress. J. Basic Microbiol. 53(4): 355-364.

Qureshi, B.A., and S.M. Zubair. 2013. Mechanical sub-cooling vapor compression systems: Current status and future directions. Int. J. Refrig. 36(8): 2097-2110.

Raafat, N.Z., and E.R. Tharwat. 2011. Improving wheat grain yield and its quality under salinity conditions at a newly reclaimed soil by using different organic sources as soil or foliar applications. J. Appl. Sci. Res. 7(1): 42-55.

Reeds, P.J., D.G. Burrin, B. Stoll, and F. Jahoor. 2000. Intestinal glutamate metabolism. The J. Nutr. 130(4): 978-982.

Reetu, M.; S. Diwakar and M. K. Bhalala. 2009. Correlation a path analysis in musk melon. Indian J.Hort .66(3): 396-399.

Rieu, I., S. Eriksson, S.J. Powers, F. Gong, J. Griffiths, L. Woolley, R. Benlloch, O. Nilsson, S.G. Thomas, P. Hedden, and A.L. Phillips. 2008. Genetic analysis reveals that C19-GA 2-oxidation is a major gibberellin inactivation pathway in Arabidopsis. Plant Cell. 20(9): 2420-2436.

Roese, A. D. 2017. *Trichoderma* nativo cultivado em grãos de aveia controla o tombamento e aumenta a altura da soja. Pesqui. Agropecu. Trop. 47(1): 102-109.

Romanenko, L. 1973. Combining ability of lavender lines. Works of the All-Union Institute of Essential Oil-Bearing Crops, 6: 38-42.

Rood, S.B., R.I. Buzzell, D.I. Major and R.P. Pharis. 1990. Gibberellins and heterosis in maize. Quantitative relationships. Crop Sci. 30(2): 281-286.

Roosta, H.R., M. Jalali, and S.M.V. Vakili Shahrbabaki. 2015. Effect of nano Fe-chelate, Fe-EDDHA and $FeSO_4$ on vegetative growth, physiological parameters, and some nutrient elements concentrations of four varieties of Lettuce (*Lactuca sativa* L.) in NFT System. J. Plant Nutr. 38(14): 1-20.

Rosenvasser, S., S. Mayak, and H. Friedman. 2006. Increase in reactive oxygen species (ROS) and in senescence-associated gene transcript (SAG) levels during dark-induced senescence of *Pelargonium* cuttings, and the effect of gibberellic acid. Plant Sci. 170(4): 873-879.

Rout, S.P., K.A. Choudary, D.M. Kar, L. Das, and A. Jain. 2009. Plants in traditional medicinal system-future source of new drugs. Inter. J. Pharm. Sci., 1(1): 1-23.

Roy, A., A. Kucukural, and Y. Zhang. 2010. I-TASSER: a unified platform for automated protein structure and function prediction. Nat. Protoc. 5(4): 725-738.

Ruiz-Lozano, J.M., R. Aroca, A.M. Zamarreno, S. Molina, B. Andreo- Jiménez, R. Porcel. 2015. Arbuscular mycorrhizal symbiosis induces strigolactone biosynthesis under drought and improves drought tolerance in lettuce and tomato. Plant, Cell Environ. 39(2): 441-452.

Rumbangh, K.P., J.A. Griswold, B.H. Igiewski, and A.N. Hamood. 1999. Contribution of quorum sensing to the virulence of *Pseudomonas aeruginosa* in burn, wound infections. Infect. Immun. 67(11): 5854-5862.

Sadeghzade, A., M. Tajbakhsh, and A. Jalili. 2012. Effects of foliar application of stimurel, force 4–L and dulzee on yield and yield components of sorghum speed feed. Intern. Res. J. Biotechnol. 3(1): 18-21.

Saeed, B., H. Gul, A.Z. Khan, L. Parveen, N.L. Badshah, and A. Khan. 2012. Physiological and quality assessment of wheat (*Triticum aestivum* L.) cultivars in response to soil and foliar fertilization of nitrogen and sulfur. J. Agric. and Biol. Sci. 7(2): 121-129.

Salehi, B., D. Mnayer, B. Özçelik, G. Altin, K.N. Kasapoğlu, C. Daskaya-Dikmen, M. Sharifi-Rad, Z. Selamoglu, K. Acharya, S. Sen, and K.R. Matthews. 2018. Plants of the genus *Lavandula*: From farm to pharmacy. Nat. Prod. Commun. 13(10): 180-193.

Samuelsson, G. 1999. Drugs of Natural Origin: A Textbook of Pharmacognosy.4th revised ed. Swedish Pharmaceutical Press, Stockholm, Sweden. 484-487.

Sánchez-Sánchez, A., J. Sánchez-Andreu, M. Juárez, J. Jordá, and D. Bermúdez. 2006. Improvement of iron uptake in table grape by addition of humic substances. J. Plant Nutr. 29(2): 259-272.

Sangwan, N.S., A.H.A. Farooqi, F. Shabih, and R.S. Sangwan. 2001. Regulation of essential oil production in plants. Plant Growth Reg. 34(1): 3-21.

Saptari, R.T. and K. Dewi. 2013. Effect of borax and gibberellic acid on the growth and development of red chilli (*Capsicum annuum* L. "gelora"). In Third Basic Sci. Int. Conf. (B41). 1-3.

Sardoei, A.S., and M. Shahdadneghad. 2014. Effects of foliar application of gibberellic acid on chlorophyll and carotenoids of marigold (*Calendula officinalis* L.). Int. J. Adv. Biol. Biomed. Res. 2(6): 1887-1893.

Sarkar, S., S. Chakraborty, S. Banerjee, and P.K. Bose. 2002. Facies sequence and cryptic imprint of sag tectonics in the late Proterozoic Sirbu Shale, central India. Precambrian sedimentary environments: A modern approach to ancient depositional systems. Wiley. 369-381.

Sarker, L.S., Z.A. Demissie, and S.S. Mahmoud. 2013. Cloning of a sesquiterpene synthase from *Lavandula x intermedia* glandular trichomes. Planta. 238(5): 983-989.

Sarker, S.D., and L. Nahar. 2012. An introduction to natural products isolation. Nat. Prod. Isol. 1-25.

Saykhul, A., C. Chatzissawidis, I. Therios, K. Dimassi, and T. Chatzistathis. 2014. Growth and nutrient status of olive plants as influenced by foliar potassium applications. J. Soil Sci. Plant Nutr. 14(3): 602-615.

Schmiderer, C., P. Grassi, J. Novak, M. Weber, and C. Franz. 2008. Diversity of essential oil glands of clary sage (*Salvia sclarea* L., Lamiaceae). Plant Biol. 10(4); 433-440.

Schmidt, E. 2010. Production of essential oils. Handbook of essential oils: science, technology, and applications. 83-90.

Schwechheimer, C., and B.C. Willige. 2009. Shedding light on gibberellic acid signalling. Curr. Opin. Plant Biol. 12(1): 57-62.

Seen, T.L., and A.R. Kingman. 1998. A review of humus and humic acid research series no. 145, S.C. Agricultural experiment station, Clemson, SC.

Segura, J. and M.C. Calvo. 1991. *Lavandula* spp.(Lavender): in vitro culture, regeneration of plants, and the formation of essential oils and pigments. In Medicinal and Aromatic Plants III. Springer, Berlin, Heidelberg. 283-310.

Selim, S.M., F.M. Matter, M.A. Hassanain, and M. Samah-Youssef. 2017. Response of growth, flowering, concrete oil and its component of *Polianthes tuberosa* L. cv. Double to phosphorus fertilizer and Gibberellic acid. Int. J. Curr. Microbiol. Appl. Sci. 6(9): 1639-1652.

Shahbandeh, M. 2019. US Category Share of Avocado Sales by Type, 2019. Consumption in the United States from 1985 to 2018 (in Million Pounds).Available online: https://www.statista.com/statistics/591263/average-avocado-consumption-us-per-week/ (accessed on 17 October 2019)."

Shakarami, K., A. Doniavi, T. Azdast, and K.M. Aghdam. 2013. Microcellular foaming of PVC/NBR thermoplastic elastomer. Mater. Manuf. Process. 28(8): 872-878.

Shani, E., R. Weinstain, Y. Zhang, C. Castillejo, E. Kaiserli, J. Choryc, R.Y. Tsien, and M. Estelle. 2013. Gibberellins accumulate in elongating endodermal cells of *Arabidopsis* root. Plant Biol. 110(12): 4834-4839.

Shellie R, Mondello, L., P. Marriott, and G. Dugo. 2002. Characterization of lavender essential oils by using gas chromatography–mass spectrometry with correlation of linear retention indices and comparison with comprehensive two-dimensional gas chromatography. J. Chromatogr. A. 970(1-2): 225-234.

Sienkiewicz, E., F. Magkos, K.N. Aronis, M. Brinkoetter, J.P. Chamberland, S. Chou, K.M. Arampatzi, C. Gao, A. Koniaris, and C.S. Mantzoros. 2011. Long-term metreleptin treatment increases bone mineral density and content at the lumbar spine of lean hypoleptinemic women. Metabolism. 60(9): 1211-1221.

Silva, A.L.L., C. Rodrigues, J.D.L. Costa, M.P. Machado, R.D.O. Penha, L.A. Biasi, L.D.S. Vandenberghe, and C.R. Soccol. 2013. Gibberellic acid fermented extract obtained by solid-state fermentation using citric pulp by *Fusarium moniliforme*: Influence on *Lavandula angustifolia* Mill. cultivated in vitro. Pak. J. Bot. 45(6): 2057-2064.

Singal, P.K., N. Kapur, K.S. Dhillon, R.E. Beamish, and N.S. Dhalla. 1981. Role of free radicals in spectrometry. J. Chromato. A. 1141: 279-286.

Singh, A, S. Singh, and S.M. Prasad. 2016. Scope of nanotechnology in crop science: Profit or Loss. Research and Reviews: J. Bot. Sci. 5(1): 1-4.

Singh, A., J. Singh, and S. Sharma. 1989. Multivariate Analysis in Relation to Genetic Improvement in Lavender, *Lavandula officinalis Chaix*. Plant Breed. 102(4): 302-305.

Singh, P., D. Singh, D.K. Jaiswal, D.K. Singh, and V. Singh. 2017. Impact of Naphthalene Acetic Acid and Gibberellic Acid on Growth and Yield of Capsicum, *Capsicum annum* (L.) cv. Indra under Shade Net Conditions. Int. J. Curr. Microbiol. Appl. Sci. 6(6): 2457-2462.

Śmigielski, K., A. Raj, K. Krosowiak, and R. Gruska. 2009. Chemical composition of the essentials oil of *Lavandula angustifolia* cultivated in Poland. J. Essent. Oil Bearing Plants. 12(3): 338-347.

Sofy, M.R., A.M. Sharaf, and H.M. Fouda. 2016. Stimulatory effect of hormones, vitamin C on growth, yield and some metabolic activities of *Chenopodium quinoa* plants in Egypt. Plant Biochem. Physiol. 4(1): 1-10.

Soltani, R., S. Soheilipour, V. Hajhashemi, G. Asghari, M. Bagheri, and M. Molavi, 2013. Evaluation of the effect of aromatherapy with lavender essential oil on post-tonsillectomy pain in pediatric patients: a randomized controlled trial. Int. J. Pediatr. Otorhinolaryngol. 77(9): 1579-1581.

Sonmez, C. and E. Bayram. 2017. The influence of different water and nitrogen applications on some yield parameters and antioxidant activity in sage (*Salvia officinalis* L.). Turkish J. Field Crops. *22*(1): 96-103.

Stashenko, E.E., and J.R. Martínez. 2008. Sampling flower scent for chromatographic analysis. J. Sep. Sci. 31(11): 2022-2031.

Stevenson, F.J., 1994. Humus chemistry: genesis, composition, reactions. John Wiley and Sons.

Stojanova, B., S. Maurice, and P.O. Cheptou. 2016. Is plasticity across seasons adaptive in the annual cleistogamous plant Lamium amplexicaule? Ann. Bot. 117(4): 681-691.

Sudriá, C., M.T. Pinol, J. Palazón, R.M. Cusidó, R. Vila, C. Morales, M. Bonfill, and S. Cañigueral. 1999. Influence of plant growth regulators on the growth and essential oil content of cultured *Lavandula dentata* plantlets. Plant Cell Tissue Organ Cult. 58(3): 177-184.

Svenningsen, N.B., S.J. Watts-Williams, E.J. Joner, F. Battini, A. Efthymiou, C. Cruz-Paredes, O. Nybroe, and I. Jakobsen. 2018. Suppression of the activity of arbuscular mycorrhizal fungi by the soil microbiota. ISME J. 12(5): 1296-1307.

Swanepoel, K.M., and W. Alberts. 2009. Good agriculture practices for production of essential oil crops. S. Afr. J. Bot. 75(2): 421-422.

Szejtli, J., L. Szente, G. Kulcsar, L.Z. Kernoczy. 1986. Beta-cyclodextrin complexes in talc powder compositions. Cosmet. Toilet. 101(10): 74-79.

Taiz, L. and Zeiger, E., 2010. Responses and adaptations to abiotic stress. Plant Physiology, Fifth Edition. Sunderland, MA: Sinauer Associates, Inc. 755-778.

Taiz, L., and E. Zeiger. 2004. *Fisiologia Vegetal*. Porto Alegre, Artmed. 613-641.

Tamalla, N.S., F. Seraj, H. Pirdashti, and Y. Yaghoubian. 2014. The effect of seed biopriming by *Piriformospora indica* and *Trichoderma virens* on the growth, morphological and physiological parameters of mung bean (*Vigna radiate* L.) seedlings. Iran. J. Seed Sci. Res. (2): 1.

Tan, K.H., 2003. Humic matter in soil and the environment: principles and controversies. CRC press.

Tanimoto, E. 2012. Tall or short? Slender or thick? A plant strategy for regulating elongation growth of roots by low concentrations of gibberellins. Ann. Bot. 110(2): 337-381.

Tanker, M., and N. Tanker. 1990. Pharmacognesi (vol 2) Ankara University Faculty of Pharmacology editions No 65 Ankara.

Tapsell, L.C., 2006. Health benefits of herbs and spices: the past, the present, the future. Med. J. Aust. August 1.

Tapsell, L.C., I. Hemphill, I. Cobiac, D.R. Sullivan, M. Fenech, C.S. Patch, S. Roodenrys, J.B. Keogh, P.M. Clifton, P.G. Williams, and V.A. Fazio. 2006. Health benefits of herbs and spices: the past, the present, the future. J. Soil Sci. Plant Nutr. 14(3): 602-615.

Tayade, A.B., Dhar, P., Sharma, M., Chauhan, R.S., Chaurasia, O.P. and Srivastava, R.B., 2013. Antioxidant capacities, phenolic contents, and GC/MS analysis of *Rhodiola imbricata* Edgew. Root extracts from Trans-Himalaya. J. Food Sci. 78(3), C402–10.

Tesserand, R. 1988. Lavender beats benzodiazepines. Int J Aromather 1: 1-2. the essential oils and aromatic water from *Teucrium persicum Boiss*. Iran. J. Pharm. Ther. Med. 4(1): 52-57.

Tholl, D. 2006. Terpene synthases and the regulation, diversity, and biological roles of terpene metabolism. Curr. Opin. Plant Biol. 9: 297-304.

Tisserand, R., and T. Balacs. 1999. Essential oil safety. A Guide for Health care professional Harcourt. Glasgow. 1:1-9

Tripathi, L., and J.N. Tripathi. 2003. Role of biotechnology in medicinal plants. Trop. J. Pharm. Res. 2(2): 243-53.

Tuna, A.L., C. Kaya, M. Dikilitas, and D. Higgs. 2008. The combined effects of gibberellic acid and salinity on some antioxidant enzyme activities, plant growth parameters and nutritional status in maize plants. Environ. Exp. Bot. 62(1): 1-9.

Turkman, O., A. Dursun, M. Turan, and C. Erdin. 2004. Calcium and humic acid effect seed germination, growth, and nutrient content of tomato (*Lycopersicon esculentum* Mill.) seedling under salin soil conditions. Acta Agric. Scandinavica-Section B-Soil Plant Sci. 54(3): 168-174.

Tyler, E.V., L.R. Brady, and J.E. Robbes. 1988. Pharmacognosy. 9th ed. Lea and Febiger, Philadelphia, 82-85.

Uhlenhuth, E.H., M.B. Balter, T.A. Ban, K. Yang. 1999. Depress. Anxiety 1999, 9, 107-116. Ashton, H.C. Drugs 1994, 48: 25-40.

Umezu, T., K. Nagano, H. Ito, K. Kosakai, M. Sakaniwa, and M. Morita. 2006. Anticonflict effects of lavender oil and identification of its active constituents. Pharmacol. Biochem. Behav., 85(4):713-721.

Upson, T. 2002. The taxonomy of the genus *Lavandula* L. In: Lis-Balchin M, editor. Lavender. Medicinal and aromatic plants: industrial profiles. London, UK: CRC Press; p.234.

Upson, T., and S. Andrews. 2004. The genus *Lavandula*. Royal Botanic Gardens Kew.

Vafayi-Rostami, S., R. Abbasi, H. Pirdashti, and M. Ghajar-Sepanlou. 2019. Effect of Piriformospora indica and Trichoderma harzianum on Morphological Characteristics, Yield and Essential Oil of Peppermint (*Mentha piperita*) at Different Amount of Phosphorus and Irrigation. J. agr. Sci. sust. Prod. *29*(4): 37-50.

Vakili, S., M.M. Sharifat, A. Akhavan, and R. Bandegi. 2014. Effect of lavender oil *(Lavandula).*

Varanini, Z., and R. Pinton. 1995. Humic substances and plant nutrition, progress in botany. Springer, Berlin. 97-117

Verma, R.S., L.U. Rahman, C.S. Chanotiya, R.K. Verma, A. Chauhan, A. Yadav, A. Singh, and A.K. Yadav. 2010. Essential oil composition of *Lavandula angustifolia* Mill. cultivated in the mid hills of Uttarakhand, India. J. Serbian Chemical Soc. 75(3): 343-348.

Vinayarani, G., and H. Prakash. 2018. Growth promoting rhizospheric and endophytic bacteria from *Curcuma longa* L. as biocontrol agents against rhizome rot and leaf blight diseases. Plant Pathol. J. 34(3): 218.

Vokou, Despina, Stella Kokkini, and Jean-Marie Bessiere. 1993. Geographic variation of Greek oregano (*Origanum vulgare ssp. hirtum*) essential oils. Biochemical Systematics and Ecology 21, no. 2: 287-295.

Vranová, R.D., W. Coman, and W. Gruissem. 2013. Network analysis of the MVA and MEP pathways for isoprenoid synthesis Annu. Rev. Plant Biol., 64: 665-700.

Wahyuni, N.L., W.B. Sunarharum. D.R.A. Muhammad, and A.D. Saputro. 2021. Formation and development of flavour of cocoa (*Theobroma cacao* L.) cultivar Criollo and Forastero: a review. In IOP Conference Series: Earth and Environmental Science (Vol. 733, No. 1, p. 012078).

Walsh, E., and C. Wilson. 1999. Complementary therapies in long-stay neurology in-patient settings. Nurs. Stand. 13(32): 32.

Wandruszka, R.V., M. Schimpf, M. Hill, and R. Engebretson .1999. Characterization of acid size fraction by SES and MALS. Org. Geochem. (30) 4: 229- 235.

Webb, N.R., R.T. Clarke, and J.T. Nicholas. 1984. Invertebrate diversity on fragmented Calluna-heathland: effects of surrounding vegetation. J. biogeogr.41-46.

Welsh, C. 1995. Three essential oils for the medicine cabinet. Alternative Health Practitioner; 3: 11-15.

Werker, E. 1993. Function of essential oil-secreting glandular hairs in aromatic plans of Lamiacea—a review. Flavour Frag. J. *8*(5): 249-255.

Wiley, J., Sons, Ltd; Chichester. 2011. Thormar H. Front matter. In: Thormar H, Kako H.

Wilkinson, R.G., 2002. Unhealthy societies: the afflictions of inequality. Routledge.

Williams, D.H., M.J. Stone, P.R. Hauck, and S.K. Rahman. 1989. Why are secondary metabolites (natural products) biosynthesized? J. Nat. Prod, 52: 1189-1208.

Wink, M. 1999. Introduction: biochemistry, role, and biotechnology of secondary products. *In* M Wink, ed, Biochemistry of Secondary Product Metabolism. CRC Press, Boca Raton, FL. 1–16.

Woelk, H., and S. Schlafke. 2010. A multi-center, double-blind, randomised study of the Lavender oil preparation Silexan in comparison to Lorazepam for generalized anxiety disorder. Phytomedicine 17: 94-99.

Wojcik, P. 2004. Uptake of mineral nutrients from foliar fertilization (review). J. Fruit Ornam. Plant Res. 12: 201-218.

World Health Organization, and Research for International Tobacco Control. WHO report on the global tobacco epidemic, 2008: the MPOWER package. World Health Organization, 2008.

Woronuk, G., Z. Demissie, M. Rheault, and S. Mahmoud. 2011. Biosynthesis and therapeutic properties of *Lavandula* essential oil constituents. Planta medica, *77*(01):7-15.

Wright, S.F., and A. Upadhyaya.1996. Extraction of an abundant and unusual protein from soil and comparison with hyphal protein of arbuscular mycorrhizal fungi. Soil Sci., 161: 575-586.

Yacoub, A., J. Gerbore, N. Magnin, R. Haidar, S. Company, and P. Rey. 2017. Transcriptional analysis of the interaction between the oomycete biocontrol agent, *Pythium oligandrum*, and the roots of Vitis vinifera L. Biol. Control. 120: 26-35.

Yaghoobi, K., G. Kaka, K. Mansouri, and S.R. Hosseini. 2016. *Lavandula angustifolia* extract improves the result of human umbilical mesenchymal Wharton's jelly stem cell transplantation after contusive spinal cord injury in Wistar rats. Stem Cells International 2016:1–12.

Yamada, K., Y. Mimaki, and Y. Sashida. 1994. Anticonvulsive effects of inhaling lavender oil vapour. Biol. Pharm. Bull. 17: 359-360.

Yang, S.A., S.K. Jeon, E.J. Lee, C.H. Shim, and I.S. Lee. 2010. Comparative study of the chemical composition and antioxidant activity of six essential oils and their components. Nat. Prod. Res., 24(2): 140-151.

Yanga, L., F. Zhaoa, Q. Changa, T. Li, and F. Li. 2015. Effects of vermicomposts on tomato yield and quality and soil fertility in greenhouse under different soil water regimes. Agric. Water Manage., 160: 98-105.

Yasseen, A.A. 2002. Total cellular ribose nucleic acid analysis of multi-drug resistant cancer cells. Saudi med. J., *23*(10): 1206-1209.

Zakaria, M. 1991. Isolation and characterization of active compounds from medicinal plants. Asia Pac. J. Pharmacol., 6, 15-20.

Zhang, B.B., W.Z. Liu, S.X. Chang, and A.O. Anyia. 2013. Phosphorus fertilization and fungal inoculations affected the physiology, phosphorus uptake and growth of spring wheat under rainfed conditions on the Canadian prairies. J. Agron. Crop Sci. 199(2): 85-93.

Zhu, J.K. 2001. Plant salt tolerance. Trends Plant Sci., *6*(2): 6-71.

CHAPTER III

IMPACT OF IRON CHELATE AND GA_3 ON GROWTH AND PRODUCTION OF ESSENTIAL OILS IN *LAVANDULA*

Abstract

The diversity of medicinal and aromatic plants requires special attention through improved production of some species, such as *Lavandula sp.* L. A greenhouse experiment was performed to assess the effects of GA_3 and iron chelate on the growth characteristics and essential oil content and yield of three lavender species: *Lavandula* x *intermedia, L. angustifolia,* and *L. stoechas*. The following treatments were applied in a factorial using a RCBD with five replications: GA_3 foliar spray at 0, 200, or 400 mg/L, and iron chelate foliar spray at 0, 1, 2, or 3 g/L. The experiment was repeated two times. The essential oil was extracted from the air-dried flowers, leaves, and stems using 70% ethanol in a Soxhlet distillation apparatus for 9 h per sample. Growth parameters measured included plant material dry weight, plant height, number of branches, number of flowers. Essential oil production was evaluated based on the plant material dry weight. Both quantity and quality of oil were examined. GC/MS analysis was used to quantify essential oil content and identify essential oil components of the dried flowers, leaves, and stems. Essential oil ratio and yield were greatest in all species with applications of 400 mg/L GA_3, and 3 g/L iron chelate. Only 400 mg/L GA_3 with 3 g/L of iron chelate affected growth parameters. *Lavandula stoechas* flower number was not affected by GA_3 or iron chelate**.** Iron chelate at 3 g/L increased the production of essential oil. The preferred treatment to enhance

growth and essential oil production of *Lavandula* was determined to be 400 mg/L GA_3 with 3 g/L iron chelate.

Keywords: Lavender, Iron chelate, Gibberellic acid, essential oil, GC-MS

Introduction

The genus *Lavandula* belongs to the *Lamiaceae* family and is native to the Mediterranean. It is cultivated worldwide for its therapeutic and economic value as well as for ornamental and decorative purposes (González-Coloma et al., 2011; Touati et al., 2011). The genus consists of about 30 species and 100 varieties grown mainly for essential oils. In addition to the essential oil economic value, lavender contains phenolic compounds known as antioxidants and has pharmacological properties. Lavender is used as a spasmolytic, carminative, gastric, or diuretic and is nowadays used as a mild sedative and medicine in various phytopharmaceuticals **(**Basch et al., 2004; Cavanagah and Wilkinson, 2002). There are three important lavender species with high commercial value in the world: Lavender (*Lavandula angustifolia.* Syn. *L. officinalis, L. vera*), Lavandin (*Lavandula* × *intermedia* [*L. angustifolia* × *L. latifolia*] Syn. *L. hybrida*), and Spike lavender (*Lavandula spica* Syn. *L. latifolia*) (Tucker, 1985). Lavender flowers contain an essential oil of primarily linalyl acetate (40%), linalool (30%), limonene, beta-osmine, 1,8-cineole, camphor, beta-terpineol, and borneol, but also, phenolic acids (rosmarinic acid), ursolic acid, coumarin (umbelliferone, herniarin), flavonoids and sterols (Nartowska J., 2012). Lavender essential oil contains high levels of linalool and linalyl acetate and the mild essential oils lavandolyl acetate, terpenine-4 ole, and lavandulol. Lavender may contain different levels of 1,8 cineole and camphor (McGimpsey 1999). The calming effect of lavender appears to be due to the presence of the compound linalool, a mono terpene, one of the main components of lavender oil

(Linck et al., 2009). Research indicates lavender oil acts on the gamma-aminobutyric acid (GABA) pathways thus acting as a general inhibitor of neurotransmission)Huang et al., 2008).

Essential oils are widely used in cosmetics, and perfumes. Due to the aroma and properties of lavender essential oil, it can be used as an insect repellant, a relaxant, antioxidant, and anti-microbial agent (Sabara and Kunicka-Styczynska, 2009). Many internal and external factors influence growth, development, and secondary development metabolite biosynthesis and accumulation in medicinal and aromatic plants (Hassanpouraghdam et al., 2008). Gibberellins are among the plant hormones that encourage growth and are found naturally in all plants of the plant kingdom in the developing stems, young leaves, fruits, and early seed germination (Taiz and Zeiger, 2010). Gibberellins are plant growth regulators (PGRs) that stimulate accelerated cell division and enlargement causing unique effects on the flowering behavior of plants (Abdul et al., 2009; Akter et al., 2007). Treatment with gibberellins contributes to the transferring of nutrients to a greater degree towards the site of growth. (Iqbal et al., 2011). The use of PGRs is one of the techniques that helps in increasing the growth and the active substances of many medicinal plants. Iron, although it may be required more than other trace minerals, is considered a trace mineral essential for plants (Taiz and Zeiger, 2010). It is the primary co-factor for 140 enzymes that include biochemical reactions (Brittenham, 1994). Jensen (2004) showed that seaweed extracts contain auxins, gibberellins, and cytokinins and spraying on plants increases roots absorption of nutrients, stem thickness and growth.

Micronutrients improve the vegetative growth and increase plant yield. They are necessary components for building organic compounds and improving the course of vital activities inside the plant, which is reflected in the growth and active substances in the plant (Haynes and Goh, 2013). In this specific context about lavender, more clarification is needed

regarding the advantages and disadvantages of using minerals to improve the production volume and quality of essential oils (Shekofteh et al., 2013).

The aim of this study was to evaluate the effect of different rates of iron chelate and GA_3 on greenhouse production of lavender (*Lavandula* sp.). This work was carried out to study the performance of three varieties of lavender in response to foliar applications of chelated iron and GA_3 under greenhouse conditions. In this investigation we studied the effects of chelated iron and GA_3 on essential oil yield and quality and morphological characteristics of three species of lavender aiming to the future cultural management of this high- value aromatic plant.

Materials and methods

Plant material and experimental design

The experiments were conducted in two seasons in the greenhouse from February to August 2019-2020. Lavender cuttings were obtained for three cultivars (Fig 3.1) *Lavandula* × *intermedia* 'Provence', *L. angustifolia* 'Hidcote' (Fig 3.1), and *L. stoechas* 'Otto Quast' (Fig 3.3) (Emerald Coast Growers, Inc., Pensacola, FL). They were planted in 15 cm (1 L) containers in a peat-based substrate (Pro Mix BX, Premier Tech Horticulture, Quakertown, PA) when their height was 15 cm. A total of 180 plants were potted in the greenhouse between 2019-2020. No pesticides were used throughout the trial period. The experiment was carried out in a Randomized Complete Block Design (RCBD) with five replications. The following treatments were applied as a foliar spray: GA_3 (Fine Americas, Inc., Walnut Creek, CA) at 0, 200, or 400 mg/L, and iron chelate (Miller Chemical & Fertilizer Corp., Hannover, PA) at 0, 1.5, 2.5, or 3.5 g/L.

The growing substrate was tested in the Mississippi State University soil testing laboratory (Mississippi State, MS) (Table 3.1). Plants were grown in Mississippi State

University's Department of Plant and Soil Sciences greenhouses (latitude 27-33 ° N; longitude 88-47 ° W) (Fig 3.2) with the temperature set points at 24/20° C Day/Night.

Table 3.1 Chemical and Physical properties of the potting soil of experimental/ Mississippi State University (Soil Testing Laboratory)

Characteristics	Units	Values
Soil Ph	-----	6.3
Phosphorus	lbs./acre	905
Potassium	lbs./acre	3956
Magnesium	lbs./acre	2924
Zinc	lbs./acre	36.0
Calcium	lbs./acre	8000
Total soluble salts		0.4 (Low)
Soil texture	-----	Peatmoss

pH of soil is slightly acid.
No lime recommended for this crop.

Plants were grown for 8 months (2019-2020) and the experiment repeated twice. When 50% of the plants were flowering, the plants and flowers were harvested at the soil line, placed in 24.5 x 17 x 4 cm paper trays (#500 unbleached paper food trays; Specialty Quality Packaging, Scotia, NY) and air dried at 20°C for one month with the plant samples being turned daily (Fig 3.4). After one month the herbs were totally dried without any molding problems, and stored in paper bags (Bettina and Helge, 2015). Dry samples were weighed to determine the dry weight (Dw-g) of total plants (buds, flowers, leaves, and stems) (Table 3.2).

L. intermedia ***L. stoechas*** ***L. angustifolia***

Figure 3.1 The three species of lavender used in the study.

Figure 3.2 The experiment in the greenhouse.

Figure 3.3 *Lavandula stoechas* "Otto Quast"

Measurements of vegetative growth:

Vegetative growth was recorded at the end of the growing season for all plants, with the following assessments made: Plant height (cm) was measured from the soil to the growing top, number of branches was counted as the number of main branches of each plant, number of inflorescences on each plant at harvest, and shoot dry weight (DW-g).

Figure 3.4 Method of drying plants in the lab. Dry lavender plants in the lab in kraft paper food trays.

Table 3.2 Lavender genotypes profiled in this study, Their geographic distribution, main characteristics, and dates of harvest in the study.

Species	Distribution notes	harvest date
L. angustifolia	the Mediterranean region (Demas, et al, 2018) Augustin 2019-and the second study also in August in 2020 for all species.	
L. intermedia	mainly distributed in the Mediterranean (Shan, et al, 2005)	
L. stoechas	France and north Europe (Cavanagh and Wilkinson, 2002)	

Soxhlet extraction of essential oils and GC-MS analysis

Essential oils were extracted from the air-dried samples from the three cultivars of lavender and treatments using a Soxhlet extraction apparatus (Reverchon and Della, 1995) (Figs 3.5, 3.6). Samples were prepared by placing 5 gm of dried, finely ground (Cuisinart Spice and Nut Grinder MODEL SG-10, East Windsor, NJ) leaves and flowers in a 33 x 80 mm paper thimble filter. 150 mL ethyl alcohol was placed in the boiling chamber and the samples were extracted over 9 hours. After the extraction, the samples were filtered from the plant tissues deposited at the bottom of the flask so that the oil-based plant extract is 100% pure of impurities that affect the analysis stages using GC-MS. Then the samples were transferred to the oven at 68°C to evaporate the ethanol from the extracted oil leaving a small yield of the extracted oil (about 2 to 3 mL) in the glass flask. After that, the total weight of the oil was taken. Oils were kept in glass bottles at room temperature (approx. 20ºC) until GC-MS analysis (Fig.3.7).

Figure 3.5 The Soxhlet used in the study (Photo credit Dr. Richard L. Harkess).

Figure 3.6 Diagram of Soxhlet extraction equipment. Credit Keefah AL-Garallaa.

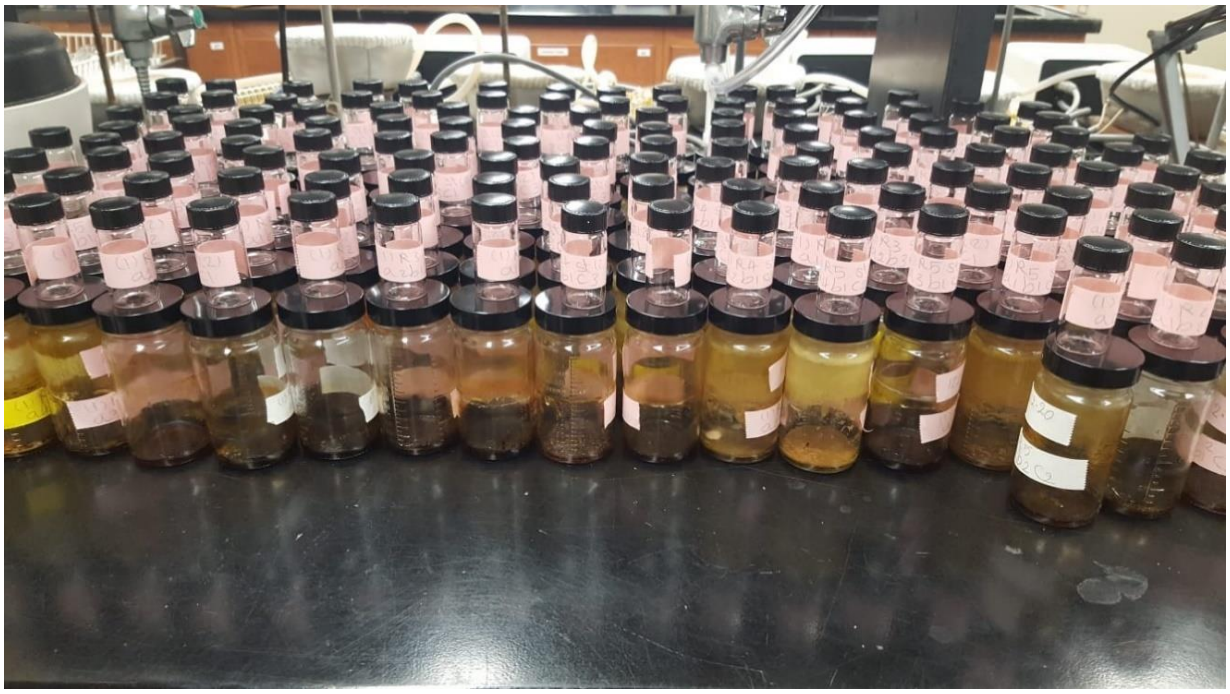

Figure 3.7 The sample after extraction (photo credit Keefah AL-Garallaa)

GC-MS Chemical identification and quantification

Linalyl and linalool (98% pure) standards (batch: LD80S110 and LP40O145 respectively) were supplied by J&K Scientific (Beijing, China). For the biological assays, the essential oil and its two pure components were dissolved in Ethanol at 5 mg/ml and serially diluted in RPMI 1640 broth (Frontier- Scientific) (concentrations expressed as %vol./vol.). The standards were used as Gas chromatography reference standards.

The chemical analysis of lavender oil and its extract was performed by gas chromatography (GC) and gas chromatography - mass spectrometry (GC-MS) techniques (Adams, 1995; Betts, 1992). Samples were compared to the standards. Quantitative data were based on normalization of the peak area without using factor correction**.** The essential oil content of the flowers and leaves was measured using GC-MS where 2 microliters were injected into the

GC-MS (Fig.3.8) which includes the automatic compound determination unit depending on the mass spectra according to the following conditions: 1. The separation column consists of 100% dimethyl multichannel and dimension (30nm × 0.25nm × 1μm). 2. Carrier helium gas at a flow rate of 1 ml. 3. The temperature of the injector is 250°C and the temperature of the ion source is 200°C. The oven temperature was automatically programmed to obtain a thermal gradient starting from 40°C (equal temperature for 3 minutes) and increasing 15°C every 1 minute to 180°C and then increasing 10°C every 3 minutes up to 300°C, and then the temperature stabilizes at 300°C.

Figure 3.8 AOC 201 Gas Chromatograph Mass Spectrometer GC/MS System(photo credit Keefah AL-Garallaa).

Pyroprobe -GC/MS

About 2 mg of each extracted dry oil was individually held in a CDS quartz tube and packed with quartz wool. Each prepared sample was pyrolyzed individually and the pyrolysis products were identified. All pyrolysis experiments were conducted using a 5200 model pyrolyzer (CDS Analytical Inc.) at 300°C. Each pyrolysis experiment was repeated twice to determine the consistency of the experimental results. The pyrolysis heating rate was 1000°C/sec at 10 second pyrolysis time for all experiments. The interface temperature was set at 300°C with the ramp at 100°C/min. The pyrolysis vapors were carried directly by ultra-high purity helium carrier gas stream (99.99%) through the transfer line to a Perkin Elmer Clarus 500 Gas Chromatograph /Mass Spectrometer (GC/MS) system. The transfer line temperature was set at 300°C and the carrier gas rate was 2 mL/min. The gas chromatograph was equipped with a DB-5MS capillary column of 30m x 0.32mm ID x 1 mm film thickness. Samples were injected in the split less mode and the injector temperature was 280°C. The initial oven temperature of the GC was 40°C for 4 min. and then programmed at a rate of 10°C/min to 280°C and maintained for 2 minutes with a total run time of 30 min. The mass spectrometer detector was an electron impact ionization device operating at 70 eV with a source temperature of 210°C and interface temperature of 225°C. The chemical component data obtained from GC/MS was analyzed using a chemical integration program together with NIST mass spectral search library. The peak area percentage was calculated for each identified compound. However, compounds that were associated with some peaks were not identified.

Recorded Data

210 days after transplanting, 3 replicates each with 18 plants of 3 lavender cultivars were used to determine the following criteria: Plant height (cm), number of branches/plant, number of flowers /plant, dry weight of /plant (g), and oil weight.

Following extraction, essential oils were collected using Pasteur pipettes and their mass was used to determine essential oil content, calculated by the formula:

$$\text{Essential oil content (\%)} = \left(\frac{\text{mass of the extracted essential oil}}{\text{plant mass used}}\right) \times 100$$

Statistical analysis

A Randomized Complete Block Design (RCBD) was used in a factorial experiment including three factors. Four concentrates of iron chelate, three concentrations of gibberellic acid, and three species of lavender. SAS statistical software (SAS® ver 9.4, SAS Institute, Cary, NC) was used for statistical analysis. The significant differences between means of experimental units among each treatment were compared using the least significant difference (LSD) at $P\alpha = 0.05$. Data were subjected to analysis of variance (ANOVA) separation of means by the LSD test at $P\alpha \leq 0.05$; the significance of iron chelate, GA_3, cultivars, and their interaction was calculated.

The interaction between species and fertilization, was evaluated by calculating the least-squares means (LS means) selecting $P \leq 0.0001$, $P \leq 0.01$, and $P \leq 0.05$ for significance of comparisons. Normalized (average = 0, variance = 1) data were submitted to principal component analysis (PCA) with the aim of discriminating species based on the studied variable

association. All statistics were performed with SAS 8.2 for Windows (SAS Institute, Cary, NC) (Maher, 3019).

Results and Discussion

Plant height (cm)

The results of the analysis of variance in table (3.3) show there is a significant effect of the experiment factors and their interaction on the plant height for the two seasons of experiment. A significant superiority of *angustifolia* was observed over the other two cultivars in this characteristic and it achieved the greatest height of 44.83 and 38.17 cm for the two seasons respectively, followed by the *intermedia* and *stoechas* with heights of 40.97 and 35.33 cm for the first season and 36.17 and 35.82 for the second season, respectively. It was also found that spraying chelate iron with a concentration of 3 g iron/L led to a significant increase in plant height, which reached 42.26 cm, compared to 1 g iron/L, which amounted to 38.72 cm for the first season.

The spraying of GA_3 at a concentration of 400 mg /L resulted in a significant superiority of plant height at 42.2 and 37.53 cm, while the shortest of the plants was the control sprayed with distilled water(Table 3.3). However, the results of the two-way interaction between chelated iron and the species indicate a significant superiority for the interaction treatment between 2 g iron / L and *intermedia*. The height was 47.03 cm for the first season, and 2 g iron /L and *angustifolia* the height reached 41.64 cm for the second season, respectively, while the minimum plant height with the interaction treatment was 3 g iron / L and *angustifolia* which gave 32. 97 cm for the first season and 3 g iron/ L and *stoechas* which gave 34.27 cm for the second season, respectively.

The treatment of the two-way interaction between spraying iron chelate at 3 g / L and 400 mg GA_3/ L was significantly superior. The lowest average resulted in the control from spraying only distilled water without iron chelated or GA_3. Significant superiority was observed for the treatment of the two-way interaction between *angustifolia* and 400 mg GA_3 / L with the highest mean recorded at 47.8 cm and 39.08 cm for the two seasons, respectively. It is also noted the significant superiority of the three-way interaction treatment 3 g iron chelate / L by 400 mg GA_3 / L by *angustifolia* which produced the greatest heights of 52.20 and 43.60 cm for the two experiment seasons, respectively. The shortest height resulted from treatment with 2 g iron chelated by *stoechas* by 0 mg GA_3 /L. The reason may be attributed to the increased plant height due to GA_3 treatment in that gibberellin has a role in cell division and increase cell absorption of water, which is reflected in the increase in their protoplasmic volume and the increase in the surface area of the plant, its tissues, and its size. Another reason may be that gibberellin has a role in increasing the leaf area and increasing the effectiveness of the photosynthesis process in the manufacture of nutrients in the leaves and their transfer to the plant, thus encouraging plant growth (Byers et al., 1990). Machado et al. (2011) indicated gibberellins are involved in the growth of buds and meristems; moreover, they also promote the breaking of bud dormancy. Considering all the results 300 mg GA_3/L was the most appropriate treatment for foliar spray of lavender plants regarding increasing plant height.

Hassanpouraghdam et al. (2011) also, indicated that considering the promoting effects of 300 mgl/L GA3 foliar application upon most of the studied traits it seems that this and likely more than 300 mg GA_3 /L would be the preferable foliar application level for promoting the plant height and essential oil production of this value-added aromatic plant. These results are also consistent with what Sajjad et al, (2014), found where the plant morphological

characteristics like plant height, spike length, spike diameter and number of florets per spike were significantly affected by foliar application of GA_3 as shown that GA_3 at 1mM concentration increased the plant height (122.14cm) of white Prosperity' Gladiolus. The foliar application of GA_3 increased the growth characters of mung bean (*Vigna radiata* L.). The growth responses were higher in all GA_3 treated plants compared to control plants. The results indicated the growth character was maximum with 50 mg/L GA_3, (Baliah et al, 2018). In this study, Baliah et al. (2018) reported that the foliar application of GA_3 increased the growth characters such as shoot length of mung bean. Fahmy and Nosir (2021) reported the spraying of iron chelate recorded the highest values of plant height with differences in iron chelate and the control in lavender plant. Similar results were demonstrated by Zehtab-Salmasi et al. (2008) on peppermint. Medicinal plants (2012) reported the maximum plant height was obtained by use of iron chelate fertilizer at a concentration of 0.5 g / L with methanol at a concentration of 30%. It was also indicated iron chelate fertilizers at a concentration of 0.5, 1, and 1.5 g /L had similar effects on plant height of sweet basil *(Ocimum basilum L.)*

Table 3.3 Effect of Gibberellic acid and iron chelate treatments on plant height cm in three species of lavender.

2019-2020						2020-2021			
Iron Chelate ($g.L^{-1}$)	Lavender Species	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species
		0	200	400		0	200	400	
0	Intermediate	35.00	42.00	42.20	39.73	34.20	35.12	35.83	35.05
	Angustifolia	40.20	46.10	46.70	39.53	35.87	36.40	36.70	35.88
	Stoechas	34.50	37.20	35.00	41.97	34.27	35.13	35.87	36.62
1	Intermedia	40.00	35.40	43.20	42.63	35.20	35.63	36.80	37.13
	Angustifolia	41.80	39.50	44.20	44.33	35.50	36.30	36.87	36.32
	Stoechas	35.00	36.70	32.70	41.83	35.53	36.07	36.33	36.22
2	Intermedia	41.60	43.00	41.30	47.03	36.04	36.57	37.27	38.48
	Angustifolia	42.40	44.10	44.60	46.13	37.80	38.50	39.13	41.64
	Stoechas	31.60	32.70	34.60	35.57	35.70	36.77	37.40	35.09
3	Intermedia	37.60	43.30	47.00	34.8	36.50	37.13	37.77	35.98
	Angustifolia	39.80	46.40	52.20	32.97	39.73	41.60	43.60	36.62
	Stoechas	34.20	40.60	39.20	38.00	34.27	35.77	36.73	35.59
LSD (0.05)			4.12		2.38		0.38		0.22
					Species Means				Pecies Means
Species× Gibberellic	Intermedia	38.55	40.92	43.43	40.97	35.49	36.11	36.92	36.17
	Angustifolia	42.55	44.15	47.8	44.83	37.23	38.20	39.08	38.17
	Stoachas	33.83	36.8	35.38	35.33	34.94	35.93	36.58	35.82
LSD (0.05)			2.06		1.19		0.19		0.11
					Iron Chelate Means				Iron Chelate Means
Iron × Gibberellic	0	36.57	41.77	41.3	39.88	34.78	35.55	36.13	35.49
	1	38.93	37.2	40.03	38.72	35.41	36.00	36.67	36.03
	2	40.53	40.1	41.33	40.66	36.51	37.28	37.93	37.24
	3	37.2	43.43	46.13	42.26	36.83	38.17	39.37	38.12

Table 3.3 (continued)

LSD (0.05)	**2.38**			**1.37**		**0.22**			**0.12**
Gibberellic acid Means	38.31	40.62	42.2			35.88	36.75	37.53	
LSD (0.05)	1.19					0.11			

Number of branches/plants

The results of the analysis of variance in (Table 3.4) show a significant effect of the experiment factors and the interaction between them in the of the number of branches for the two seasons of the experiment. The species *angustifolia* averaged more branches than the other two species (Table 3.4). Spraying iron chelate at 3 g iron/L led to a significant increase in the number of branches compared to spraying with distilled water. The number of branches was significantly greater than 3 g iron chelate / L in the second season. The spraying of GA_3 at a concentration of 400 mg GA_3 / L resulted in the greatest average branch number for the two experiment seasons, as the highest average number of branches was 28.25and 27.84 branch respectively, while the number of branches without GA_3 and sprayed with distilled water only reached 22.17and 21.97 branches for two seasons, respectively.

The results of the two-way interaction between iron chelate and the cultivars resulted in more branches for the interaction between 2 g iron / L and *angustifolia*, which, the branch number reached 38.1 for the first season and 2 g/L and *intermedia* where branch number reached 37.7 branch for the second season. The lowest mean of branch number was in the interaction of 0 g iron / L and *intermedia* which gave 19.6 and 19.2 branches for the first and second season, respectively.

The two-way interaction between spraying iron chelated and gibberellic acid was significantly superior at 3 g iron / L and 400 mg GA_3 / L compared to the lowest average branch number achieved by spraying distilled water and 0 mg GA_3 /L. Significance was observed for the two-way interaction between *angustifolia* and 400 mg GA3 /L where the highest average number of branches was observed compared to the lowest average with *stoechas* and 0 g GA_3 /L.

There was a significant three-way interaction treatment of 3 g iron / L x *angustifolia* x 400 mg GA_3 / L which produced the highest number of plant branches compared to the lowest average obtained with 0 g iron / L x *stoechas* x 0 mg GA_3 / L. It is possible that GA_3 has the potential to accelerate nutrient diversion towards cells and active growth locations and concomitant increased nutrient absorption by increasing root potential, and finally, the minerals condense with the accumulation of associated biomolecules. As with many of the studied traits, the treatment 400 mg GA_3 /L resulted in greatest branch number in lavender. Fahmy and Nosir. (2021) indicated the concentration from iron spraying recorded the highest number of branches per plan, with significant differences due to concentrations in lavender. Similar results were demonstrated by Ziedan and Eisa (2016) on dill and Mehrab (2017) on lemon balm plants. In this study, Nemati et al (2018) reported the results showed iron significantly increased chlorophyll content and stem number of peppermint (*Mentha piperita L.).* In a study of Medicinal Plants (2012) using different concentrations of methanol an interaction between methanol and iron chelate fertilizer with the largest number of branches in sweet basil (*Ocimum basilum L.*) obtained by applying methanol at 40% with iron chelate at 0.5 and 1 g/ L.

Table 3.4 Effect of GA_3 and iron chelate on number of branches per plant in three species of lavender.

2019-2020						2020-2021			
Iron Chelate ($g.L^{-1}$)	Lavender Spicies	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species
		0	200	400		0	200	400	
0	Intermedia	16.60	20.00	22.20	19.60	16.20	19.70	21.77	19.22
	Angustifolia	20.60	22.40	24.20	28.20	20.20	21.03	23.80	27.68
	Stoechas	18.20	21.40	24.40	25.20	17.80	21.00	23.70	24.37
1	Intermedia	26.60	28.40	29.60	24.80	26.07	27.70	29.27	21.68
	Angustifolia	26.40	30.40	31.60	22.40	25.90	29.97	31.30	29.06
	Stoechas	19.40	20.00	22.20	29.47	18.77	19.50	21.23	29.20
2	Intermedia	21.20	26.00	28.40	30.67	20.67	25.17	27.27	37.66
	Angustifolia	19.80	35.80	36.40	38.07	19.30	33.27	35.03	20.83
	Stoechas	20.40	33.40	23.80	21.33	21.30	32.13	26.42	19.83
3	Intermedia	22.20	24.60	27.60	20.53	22.50	24.40	27.50	26.62
	Angustifolia	32.00	40.40	41.80	25.87	32.20	40.47	40.30	24.56
	Stoechas	22.60	24.60	26.80	24.67	22.73	24.43	26.50	19.22
LSD (0.05)			2.74		1.58		1.36		0.78
					Species Means				Species Means
Species× Gibberellic Acid	Intermedia	21.65	24.75	26.95	24.45	21.36	24.24	26.45	24.02
	Angustifolia	24.70	32.25	33.50	30.15	24.40	31.18	32.61	29.40
	Stoechas	20.15	24.85	24.30	23.1	20.15	24.27	24.46	22.96
LSD (0.05)			1.37		0.79		0.68		0.39
					Iron Chelate Means				Iron Chelate Means
Iron × Gibberellic	0	18.47	21.27	23.60	21.11	18.07	20.58	23.09	20.58
	1	24.13	26.27	27.80	26.07	23.58	25.72	27.27	25.52
	2	20.47	31.73	29.53	27.24	20.42	30.19	29.57	26.73
	3	25.60	29.87	32.07	29.18	25.81	29.77	31.43	29.00

LSD (0.05)	1.58		0.91		0.78		0.45	

Table 3.4 (continued)

Gibberellic acid Means	**22.17**	**27.28**	**28.25**		**21.97**	**26.56**	**27.84**	
LSD (0.05)	0.79				0.39			

Number of flowers/plants

The results of the analysis of variance in (Table 3.5) indicate a significant effect of some experiment factors and their interaction on the of the number of flowers per plant for season one experiment. The species *angustifolia* had more flowers than *intermedia* followed by *stoechas.* Spraying iron chelate at 3 g/L increased the number of flowers per plant. The lowest average flower number was from spraying plants with distilled water only. In the second season, 3g iron chelate/ L produced an average of 62.5 flowers / plant, compared with 1g/L which had the lowest average. Plants sprayed with 400 mg GA_3/L also produced the highest average flower number in season two.

The results of the two-way interaction indicate there was no significant effect of the treatments between iron chelate and the species. To the presence of a significant interaction of 2g iron chelate/L× *intermedia* which reached 113.9 and 124.9 flowers/plant in season two experiment, respectively. The species *stoechas* receiving 0 or 400 mg GA_3/ L never bloomed. Two-way interactions between 1mg GA_3/L × 400m g iron chelate /L produced the greatest number of flowers per plant for season two. The lowest average flower numbers occurred when treated with 3mg GA_3/L in season two. A species by GA_3 resulted in the highest average flower numbers at 0 mg GA_3/ L. A significant three-way interaction between species, iron chelate and GA_3 was observed. In this interaction 1g /L iron chelate× *angustifolia* × 400 mg GA3/ L produced the highest average number of flowers.

The effects of GA_3 on flower number may be due to the role it has in seed germination, endosperm nutrient mobilization, stem elongation, leaf expansion, reducing maturation time, and increasing flower and fruit set (Roy and Nasiruddin 2011). According Hassanpouraghdam et al. (2011), GA_3 with its stimulant effects has been reported to boost the morphological, physiological, and biochemical aspects of plant growth and have additional effects on all-around growth and plant development. These effects lead plants toward the flowering growth phase in lavender. EL-Naggar et al. (2009) reported GA_3 foliar application had stimulating effects on the floral induction of *Dianthus caryophyllus* L. and thus resulted in increased flowering biomass and essential oil production. Fahmy and Nosir (2021) reported the lavender plant growth and yield due to iron and zinc application also had a direct link in improving growth and development and increased volatile oil production of dill (Mershkary and Siamese, 2014). Similar results were observed by Amini et al. (2018) on hyssop plants. Nemati et al (2018) concluded that foliar application of Nano iron (0.5 g/l) in the flowering stage increased dry matter yield and essential oil content and composition of peppermint (*Mentha piperita L.*). Samadiyan et al (2013) found applying chelated iron to the flowers of the Damascene rose plant increased the number of flowers, flower yield and oil. These results were consistent with other studies on Damascene rose (Daneshkhah et al., 2007; Rezaie et al., 2003), *Amaranthus retroflexus* (Amir-Rezaie, 2011) and *Coriandrum sativum*. Rahimi et al. (2008) reported iron promoted good vegetative growth, which led to an increase in the number of flowers. This led to an increase in the weight of the flowers due to an increase in photosynthesis and more carbohydrates stored in the flowers.

Table 3.5 Effect of GA_3 and iron chelate treatments on number of flowers per plant in three species of lavender.

2019-2020						2020-2021			
Iron Chelate ($g.L^{-1}$)	Lavender Species	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species
		0	200	400		0	200	400	
0	Intermedia	19.2	4.6	60.0	27.9	23.3	59.7	69.3	50.8
	Angustifolia	156.4	51.2	38.8	38.5	166.0	64.3	42.7	44.3
	Stoechas	0.0	30.8	0.0	23.6	0.0	27.7	0.0	32.1
1	Intermedia	73.8	13.2	28.4	46.6	84.7	16.2	32.0	56.0
	Angustifolia	98.4	59.4	88.0	82.1	102.7	62.7	93.0	91.0
	Stoechas	0.0	0.0	0.0	81.9	0.0	0.0	0.0	86.1
2	Intermedia	25.8	34.0	11.0	113.9	35.7	46.3	14.3	124.9
	Angustifolia	205.4	102.0	34.4	104.9	219.0	108.0	47.7	114.8
	Stoechas	0.0	0.0	0.0	10.3	0.0	0.0	0.0	9.2
3	Intermedia	28.8	50.0	61.0	0.0	32.0	60.7	75.3	0.0
	Angustifolia	141.6	139.0	34.0	0.0	157.0	145.7	41.7	0.0
	Stoechas	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
LSD (0.05)			82.5		47.6		25.1		14.5
					Species Means				Species Means
Species× Gibberellic	Intermediate	36.9	25.5	40.1	34.2	43.9	45.7	47.8	45.8
	Angustifolia	150.4	87.9	48.8	95.7	161.2	95.2	56.2	104.2
	Stoechas	0.0	7.7	0.0	2.6	0.0	6.9	0.0	2.3
LSD (0.05)			41.2		23.8		12.5		7.2
					Iron Chelate Means				Iron Chelate Means
Iron × Gibberellic	0	58.5	28.9	32.9	40.1	63.1	50.6	37.3	50.3
	1	57.4	24.2	38.8	40.1	62.4	26.3	41.7	43.5
	2	77.1	45.3	15.1	45.8	84.9	51.4	20.7	52.3
	3	56.8	63	31.7	50.5	63.0	68.8	39.0	65.9

LSD (0.05)	47.6			27.5		14.5			8.4

Table 3.5 (continued)

Gibberellic acid Means	**62.5**	**40.4**	**29.6**			**68.4**	**49.3**	**34.7**	
LSD (0.05)	23.8					7.2			

Dry weight /plant (g)

The results of the analysis of variance in (Table 3.6) show a significant effect of the experiment factors and the interaction between them in the dry weight for the two seasons of the experiment. The species *angustifolia* had greater dry weight than the other two cultivars. Iron chelate at 3 g iron / L increased dry weight over the other treatments. Treatment with 400 mg GA_3/ L resulted in greater plant dry weight for both experiment seasons.

The results of the two-way interaction between iron chelated and the species indicate 2 g iron / L × *angustifolia* resulted in the greatest plant dry weight. Higher rates of iron at 3g iron / L on *intermedia* resulted in the lowest plant dry weights. The two-way treatment between iron chelate and GA_3 resulted in greatest plant dry weights when 3 g iron chelate / L and 400 mg GA_3/ L was applied. The least plant dry weights were observed when plants were treated with 1 g iron chelate/L and 0 mg/l GA_3. A two-way interaction between species and GA_3 was observed where 400 mg GA_3/L applied to *angustifolia* resulted in the greatest plant dry weight. The least average dry weight was observed in *intermedia* when 0 mg GA_3/L was applied.

There was a three-way interaction of iron chelate, GA_3 and species. The treatment combination of 3 g iron chelate / L x *angustifolia* x 400 mg GA_3/ L produced the greatest dry weight. The lowest average dry weight was with treatment 0 g iron chelate / L x *intermedia* x 0 mg GA_3/ L. Sure et al. (2012) indicated GA_3 is an important PGR that affects plant growth and

development by inducing metabolic activities and regulating nitrogen use. GA_3 can delay aging and improve the growth and development of chloroplasts, thereby increasing photosynthetic efficiency leading to higher yields (Yuan & Xu, 2001). Hassanpouraghdam et al. (2011) found the dry weight of lavender was increased by GA_3 applications. GA_3 affects cell division and amplification and thus growth parameters such as accumulation of dry weight and biomass. These results are consistent with the results from Akter et al. (2007) in mustard. Likewise, it has been found that the application of GA_3 raised fresh and dry biomass of lavender (Hassanpouraghdam et al, 2011). Baliah et al. (2018) reported the foliar application of GA_3 increased fresh weight and dry weight of mung bean. Fahmy and Nosir. (2021) also indicated the maximum average values of total plant yield/plant and the total dry weight/acre on lavender were recorded using 200 mg iron /L. These results are in accordance with those found by Salamatbakhsh et al. (2012) on castor bean and by Yadegari. (2015) on Borage, thyme, and marigold and Abd-Elkader. (2016) on garlic. Nemati et al. (2018) concluded foliar application of Nano iron (0.5 g/l) during the flowering stage increased dry matter yield of peppermint (*Mentha piperita L.*).

Table 3.6 Effect of GA_3 and iron chelate treatment on plant dry weight (g) in three species of lavender.

2019-2020						2020-2021			
Iron Chelate ($g.L^{-1}$)	Lavender Species	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species
		0	200	400		0	200	400	
0	Intermedia	10.42	10.79	11.58	10.93	9.83	9.97	10.54	10.11
	Angustifolia	10.96	11.49	11.49	11.32	9.45	11.02	11.95	11.13
	Stoechas	11.91	10.60	10.58	11.44	9.65	10.12	10.33	11.26
1	Intermedia	11.11	11.32	11.52	11.56	10.96	11.11	11.33	11.35
	Angustifolia	11.28	11.67	12.11	11.31	11.07	11.43	11.93	10.81
	Stoechas	10.65	10.67	10.92	11.69	10.42	10.54	10.75	11.48
2	Intermedia	11.06	11.47	11.77	11.65	11.03	11.22	11.52	11.46
	Angustifolia	11.24	11.73	11.99	12.20	11.09	11.54	11.76	12.08
	Stoechas	10.72	11.21	11.57	11.03	10.56	11.06	11.38	10.03
3	Intermedia	11.19	11.60	11.89	10.75	11.00	11.41	11.62	10.57
	Angustifolia	11.38	12.51	12.71	11.17	11.12	12.34	12.79	11.00
	Stoechas	10.78	11.95	11.80	11.51	10.57	11.73	11.65	11.32
LSD (0.05)		0.47			0.27	0.14			0.08
					Species Means				Species Means
Species× Gibberellic Acid	Intermediate	10.95	11.30	11.69	11.31	10.70	10.93	11.25	10.96
	Angustifolia	11.21	11.85	12.08	11.71	10.68	11.58	12.10	11.46
	Stoechas	11.02	11.11	11.22	11.11	10.30	10.86	11.03	10.73
LSD (0.05)		0.23			0.13	0.07			0.04
					Iron Chelate Means				Iron Chelate Means

Table 3.6 (continued)

Iron × Gibberellic Acid	**0**	**11.10**	**10.96**	**11.22**	**00.09**		**9.64**	**10.37**	**10.94**	**10.32**
	1	11.02	11.22	11.52	11.25		10.82	11.03	11.34	11.06
	2	11.01	11.47	11.78	11.42		10.89	11.27	11.55	11.24
	3	11.12	12.02	12.14	11.76		10.90	11.83	12.02	11.58
LSD (0.05)			0.27		0.15			0.08		0.05
Gibberellic acid Means		11.06	11.42	11.66			10.56	11.13	11.46	
LSD (0.05)			0.13					0.04		

Oil yield /plant

The results of the analysis of variance in (Table 3.7) show a significant effect of the experiment factors and the interaction between them in the of the percentage of oil for the two seasons of the experiment. The species *angustifolia* produced more oil than *intermedia* which produced more oil than *stoechas*. Iron chelate applied at 3 g/L significantly increased oil yield over the control. Oil yield was greatest at 1 g iron chelate/L in the second season. GA_3 at 400 mg /L resulted in the highest average oil yield while the treatment without GA_3 had the least oil yield.

There was a two-way interaction between iron chelate and variety. Oil yield was improved with application of 2 g iron/L to *angustifolia* which provided the greatest oil yield of the treatments. The second highest oil yield was when iron chelate was applied at 1g/L to *stoechas*. The lowest mean oil yield occurred in *intermedia* when treated with 3 or 1g iron/L. The two-way treatment between iron chelate and GA_3 resulted in the highest oil yields with 3 g iron/L and 400 mg GA_3/ L. The lowest average oil yield occurred with 0 g iron/L and 0 mg GA_3/ L.

There was also an interaction effect of GA_3 and species. The species *angustifolia* when treated with 400 mg GA_3/L produced the greatest average oil yield in season one while *stoechas* at 400 mg GA_3/L produced the most oil in season two. The lowest oil yields occurred when 0 mg GA_3 was applied to any of the species.

In the three-way interaction of iron chelate, GA_3, and variety, 3 g iron/L x *angustifolia* x 400 mg GA_3/ L produced the highest oil yield for the first season and 3 g iron/L x *stoechas* x 400 mg GA_3/ L produced the highest oil weight in season two. In general, when 0 g iron and 0 mg GA_3 /L were applied, oil weight was greatly reduced.

Several factors influence the quantity and quality of secondary metabolites in medicinal and aromatic plants. Hassanpouraghdam et al. (2008) noted PGRS critically affect the primary and secondary metabolism of plants. The production of essential oils and the accumulation of plants carrying volatile oils is affected in a positive manner. Among PGRs, there is strong evidence GA_3 has consistent effects on plant growth and development, and hence on active ingredient content and productivity. Applying GA_3 to lavender plants appeared to increase the light efficiency and assimilation potential of the plants resulting in intensified production of secondary metabolites and increased biosynthesis and storage of essential oils. Moreover, it is possible the number of branches and the dry weight of the plants under the application of 400 mg L of GA_3 resulted in suitable primary and secondary metabolism reactions favoring the essential oil production (Hassanpouraghdam et al., 2008; Marshner, 1995). The highest amounts of essential oil content and yield after GA_3 application fully agrees with the growth traits of lavender in this study and consistent with the results of Hassanpouraghdam et al. (2011). It was found that the foliar application of 300 mg GA_3/L had a significant effect on most of the studied

traits. It appears this concentration of GA_3 may be the preferred foliar application level to enhance the growth properties and production of the essential oils of aromatic lavender.

Several authors confirm the effect of iron on essential oil production (e.g., Amuamuha et al., 2012; Tavallali, 2018; Yousefzadeh and Sabaghnia, 2016). These scientists reported similar results on the essential oil yield in other essential oil-bearing crops. The type of iron compound seems to be a crucial factor and have an efficacy in enhancing the content of essential oils in lavender. Nemati et al. (2018) concluded that foliar spraying of nano-iron (0.5 g / L) at the flowering stage increased the essential oil content and mint composition. In other studies, by Zehtab-Salmasi et al. (2008), the positive effect of foliar application of iron chelate is reported on the essential oil yield of peppermint, cress (Salarpour et al., 2013) and chamomile (Nasiri et al., 2010). Due to the role iron plays to influence plant growth, one of the reasons for increased photosynthetic activity is the role of this element in the structure of chloroplasts, which leads to the production of more essential oil glands in the leaves (Evans, 2009).

The findings of an experiment conducted by Ziaeyan and Malakouti)1998) indicated micronutrients increase the essential oil yields through their effect on dry matter. This increase in oil content is because of the important role in controlling the biosynthesis pathways which led to the production of various metabolic compounds including the oil (Cseke et al., 2006). Tavallali et al. (2019) reported the basil essential oil yields under the influence of different iron sources. Foliar application of iron contributed to a considerable increase in the essential oil yield. Other research (Tavallali, 2018; Amuamuha et al., 2012; Yousefzadeh and Sabaghnia, 2016) indicated a beneficial effect of iron on the essential oils production in some essential oil-bearing crops.

Table 3.7 Effect of GA_3 and iron chelate treatments on oil yield/plant (g) in three species of lavender.

2019-2020						2020-2021			
Iron Chelate ($g.L^{-1}$)	Lavender Species	Gibberellic acid ($Mg.L^{-1}$)			Iron Chelate × Species	Biofertilizer ($Mg.L^{-1}$)			Iron Chelate × Species
		0	200	400		0	200	400	
0	Intermedia	23.32	24.17	25.46	24.32	19.37	20.84	21.37	20.53
	Angustifolia	24.66	25.49	25.96	25.69	22.23	22.5	22.67	24.93
	Stoechas	21.98	22.4	23.02	25.95	22.43	22.87	23.4	23.79
1	Intermedia	24.4	25.98	26.7	26.28	23.73	25.17	25.9	23.36
	Angustifolia	25.24	26.52	27.14	25.37	23.53	26.3	27.9	22.47
	Stoechas	20.76	21.8	22.3	26.30	24.3	25.5	27.47	25.91
2	Intermedia	24.6	26.28	26.96	26.54	23.3	23.53	24.53	24.33
	Angustifolia	25.51	26.66	27.44	27.54	22.8	24.4	25.8	25.28
	Stoechas	21.19	22.05	24.76	22.47	24.3	25.3	25.9	22.90
3	Intermedia	24.74	26.52	27.57	21.62	21.3	23.4	25.37	25.76
	Angustifolia	25.8	28.04	28.78	22.66	23.6	25.37	26.87	25.17
	Stoechas	21.52	23.5	24.92	23.31	23.4	25.67	27.77	25.61
LSD (0.05)		0.58			0.33	0.43			0.25
					Species Means				Species Means
Species× Gibberellic Acid	Intermedia	24.27	25.74	26.67	25.56	21.92	23.24	24.29	23.15
	Angustifolia	25.3	26.68	27.33	26.44	23.04	24.64	25.81	24.86
	Stoechas	21.36	22.44	23.75	22.52	23.61	24.83	26.13	24.50
LSD (0.05)		0.29			0.16	0.21			0.12
					Iron Chelate Means				Iron Chelate Means

Table 3.7 (continued)

Iron × Gibberellic Acid	**0**	**23.32**	**24.02**	**24.81**	**24.05**		**21.34**	**22.07**	**22.48**	**21.96**
	1	23.47	24.77	25.38	24.54		23.86	25.66	27.09	25.53
	2	23.76	24.99	26.39	25.05		23.47	24.41	25.41	24.43
	3	24.02	26.02	27.09	25.71		22.77	24.81	26.67	24.75
LSD (0.05)			0.33		0.19			0.25		0.14
Gibberellic acid Means		23.64	24.95	25.92			22.86	24.24	25.41	
LSD (0.05)			0.16					0.12		

Results and discussion of GC-MS (Essential oil composition)

Essential oil composition

Chromatogram of lavender oil from GC-MS analysis of *Lavandula* oil were obtained from essential oil extracts of plants treated with different concentrations of iron chelate and GA_3, from the first and second seasons, 2019/2020 (Figs 3.9, 3.23). The main ingredients of the lavender essential oil are linalool, linalyl acetate, camphor, Cis linalool, lavandulol,camphene, α pinene, and myrcene (Tables 3.8, 3.9).

GC-MS Chromatographs of lavender oil

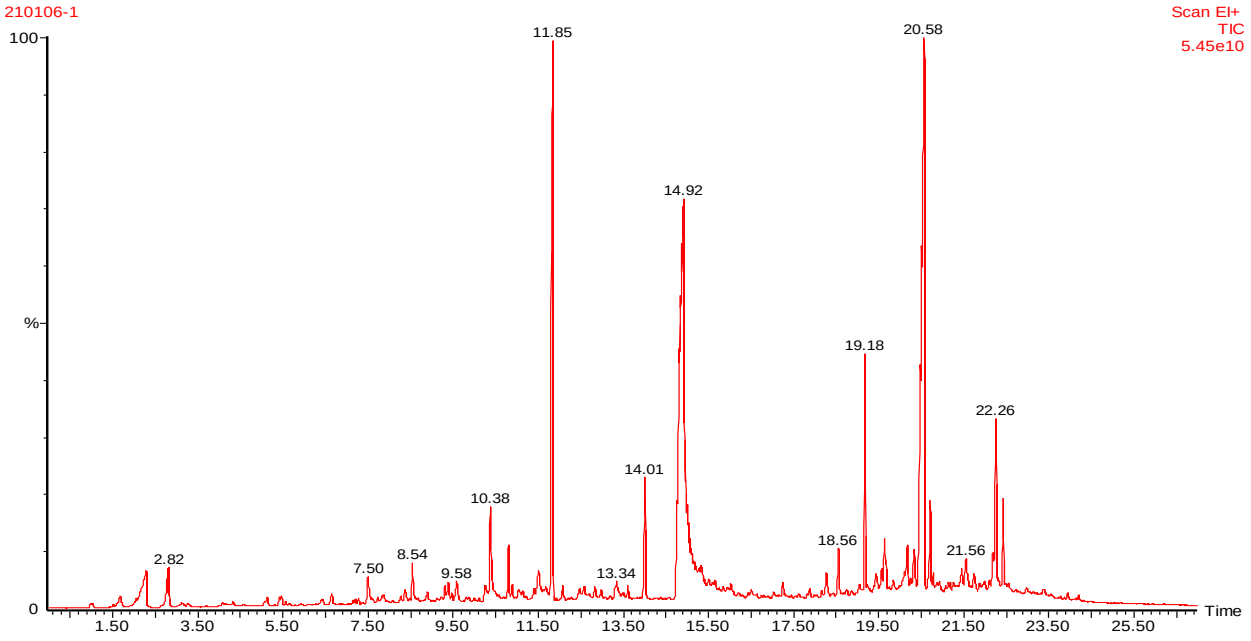

Figure 3.9 Chromatogram of essential oil extracted from 2g/l iron chelate, 400 mg/l GA_3, with *L. angustifolia*

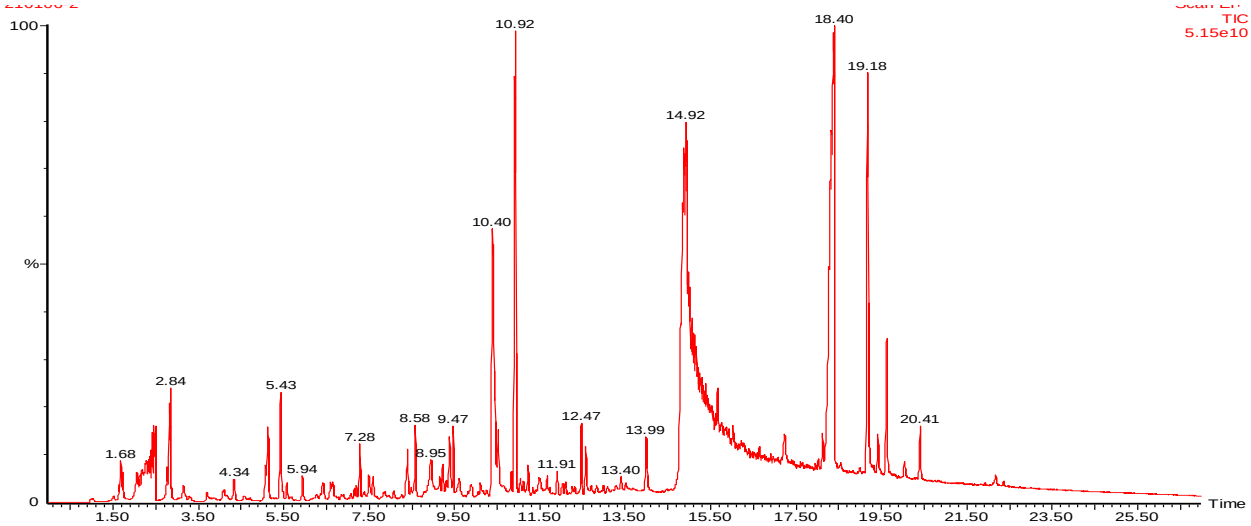

Figure 3.10 Chromatogram of essential oil extracted from 1g/l iron chelate, 400 mg/l GA3, with *L. intermedia*.

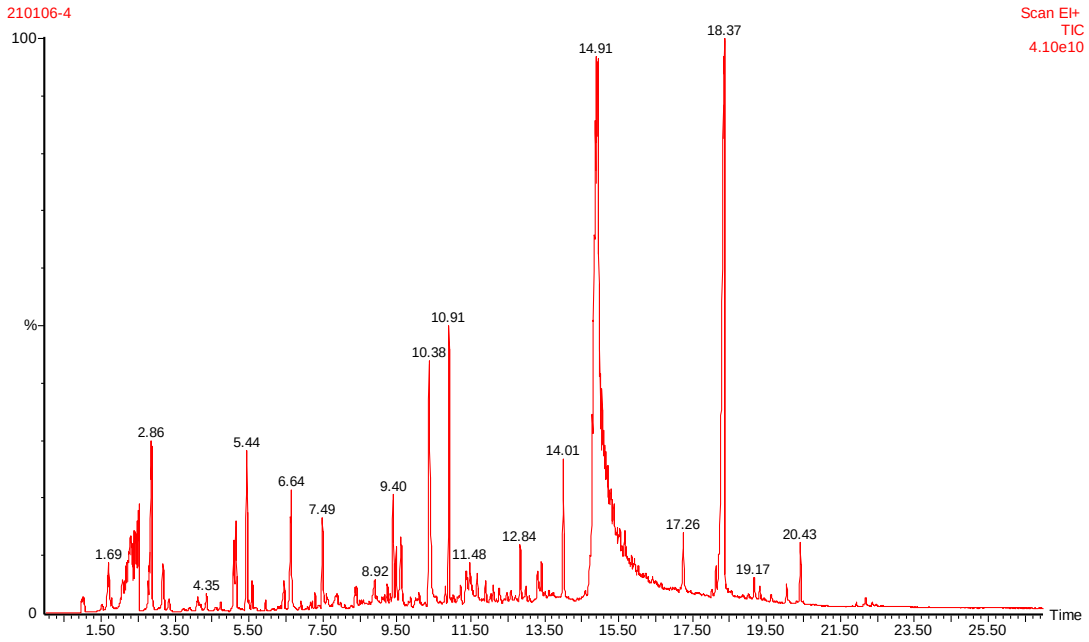

Figure 3.11 Chromatogram of essential oil extracted from 2g/l iron chelate, 400 mg/l GA3, with *L. intermedia*

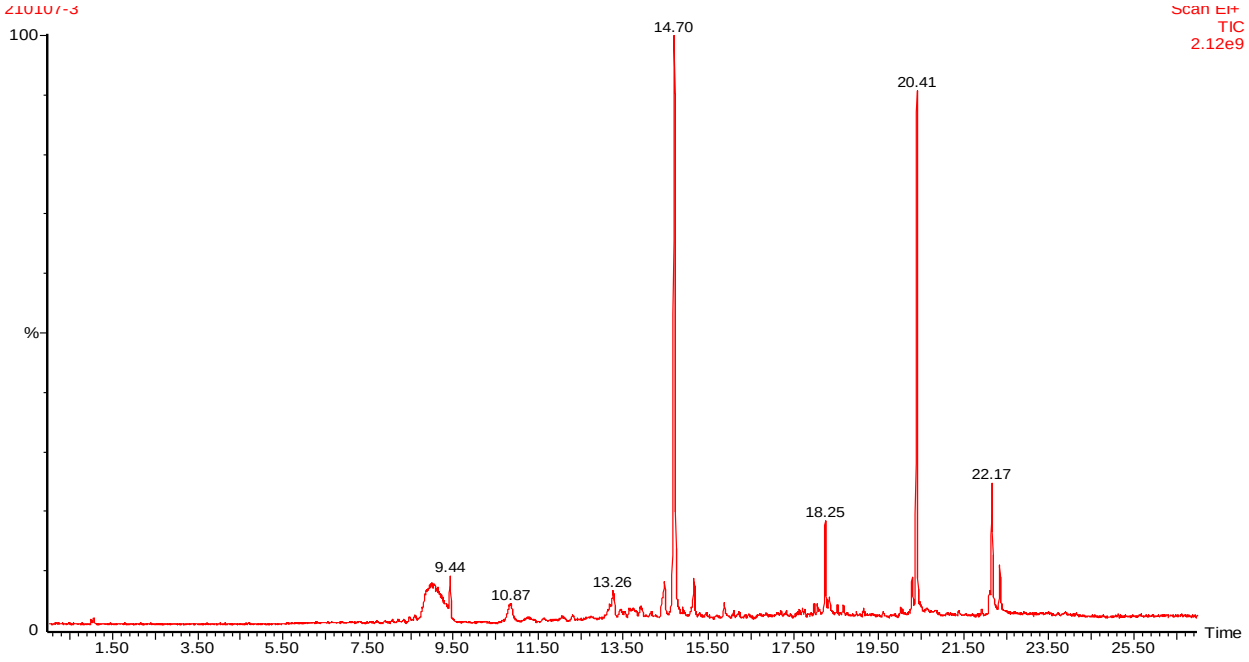

Figure 3.12 Chromatogram of essential oil extracted from 0g/l iron chelate, 0 mg/l GA3 with *L. stoechas*.

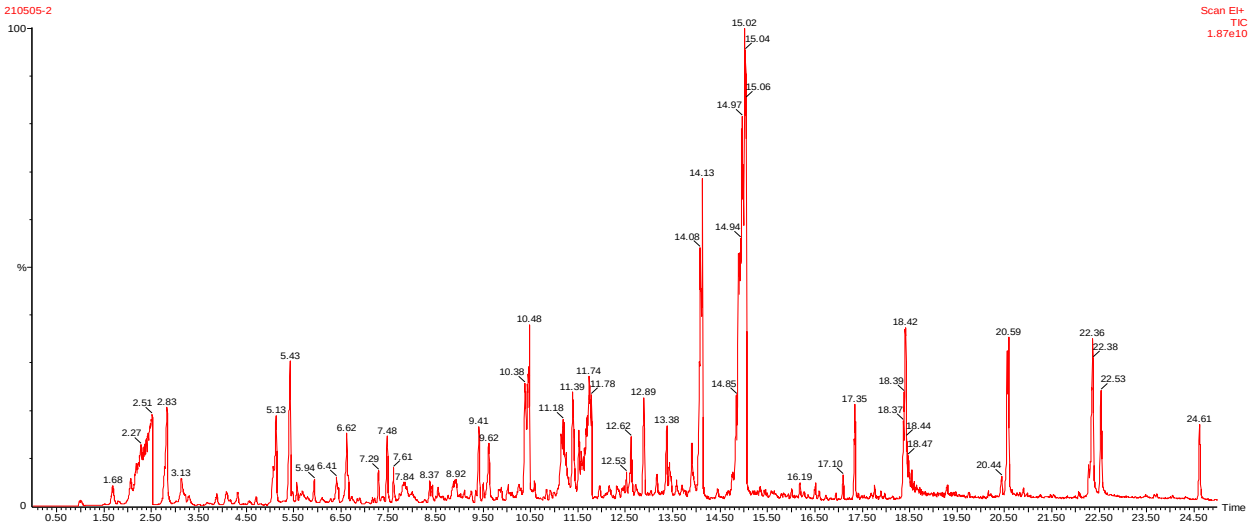

Figure 3.13 Chromatogram of essential oil extracted from 1g/l iron chelate, 200 mg/l GA3, with *L. stoechas*

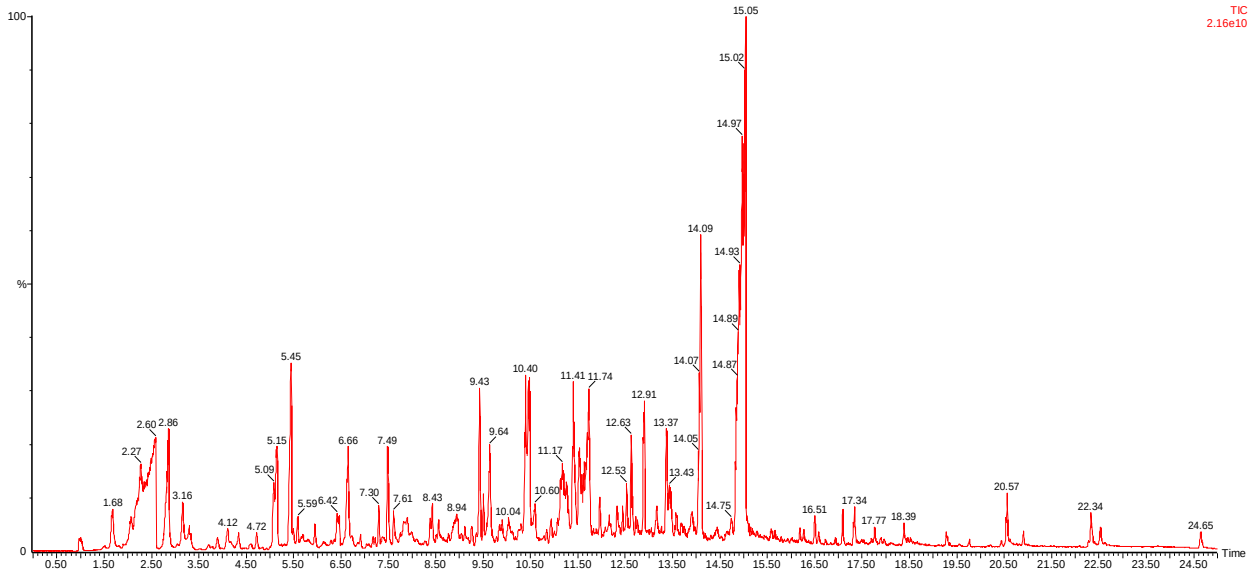

Figure 3.14 Figure: 3.14 Chromatogram of essential oil extracted from 1g/l iron chelate, 400 mg/l GA3, with *L. stoechas*

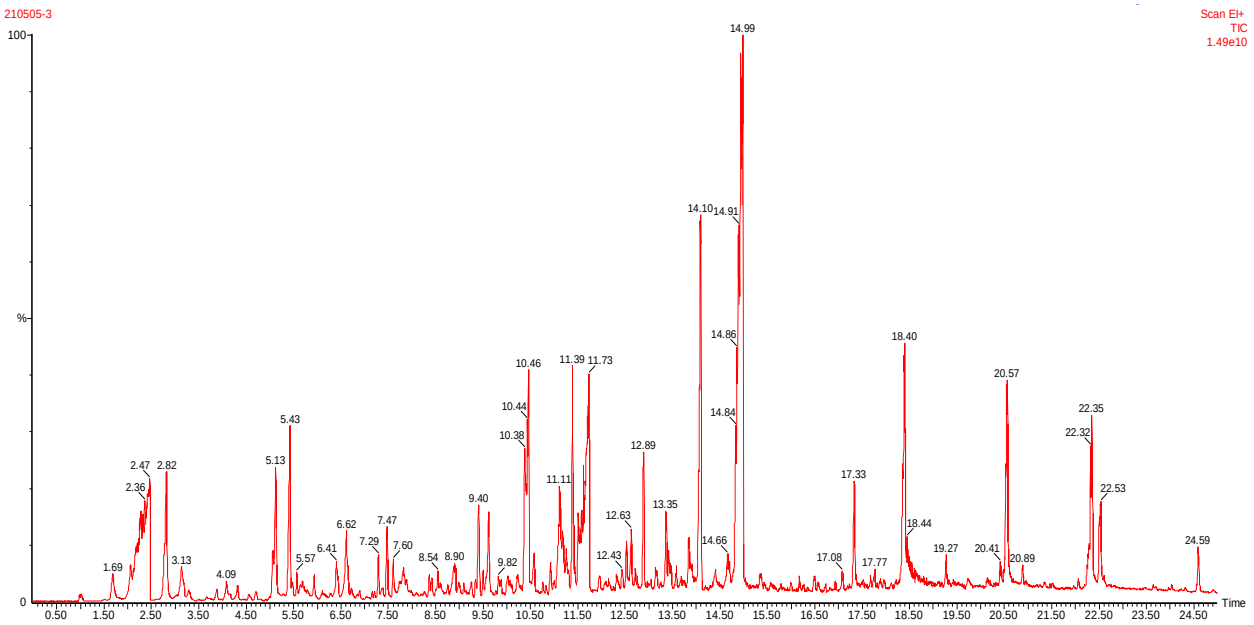

Figure 3.15 Chromatogram of essential oil extracted from 2g/l iron chelate, 200 mg/l GA3, with *L. stoechas*

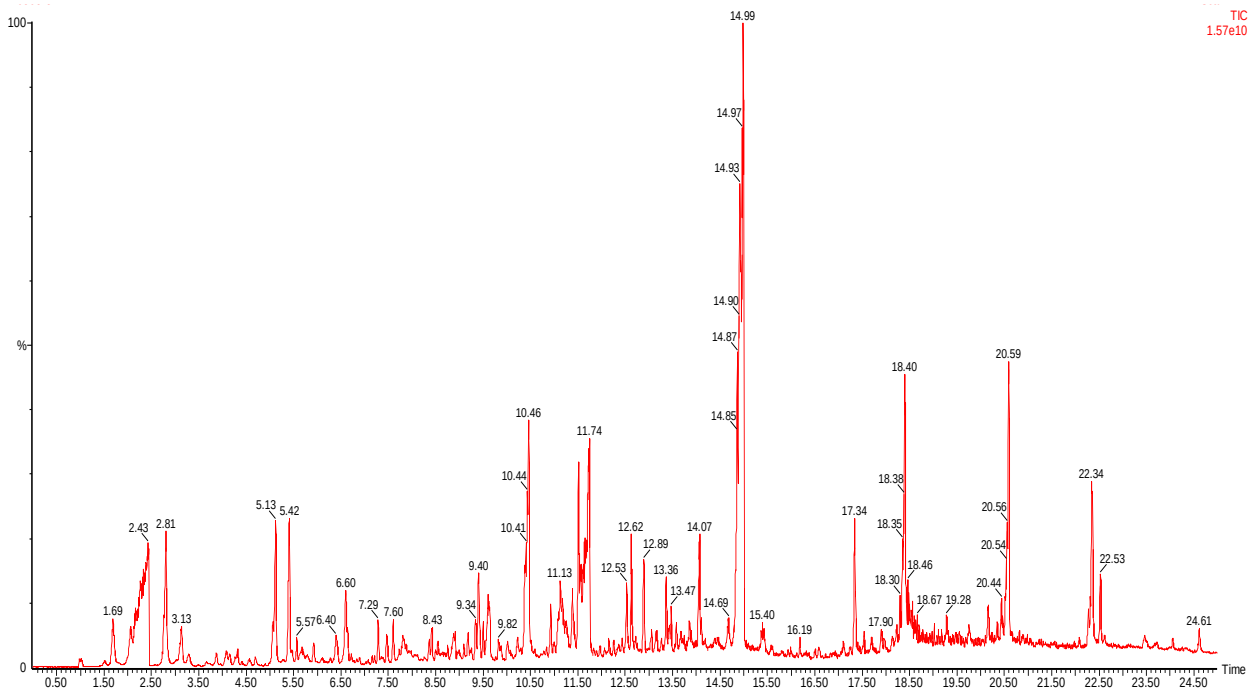

Figure 3.16 Chromatogram of essential oil extracted from 3g/l iron chelate, 400 mg/l GA3, with *L. angustifolia* .

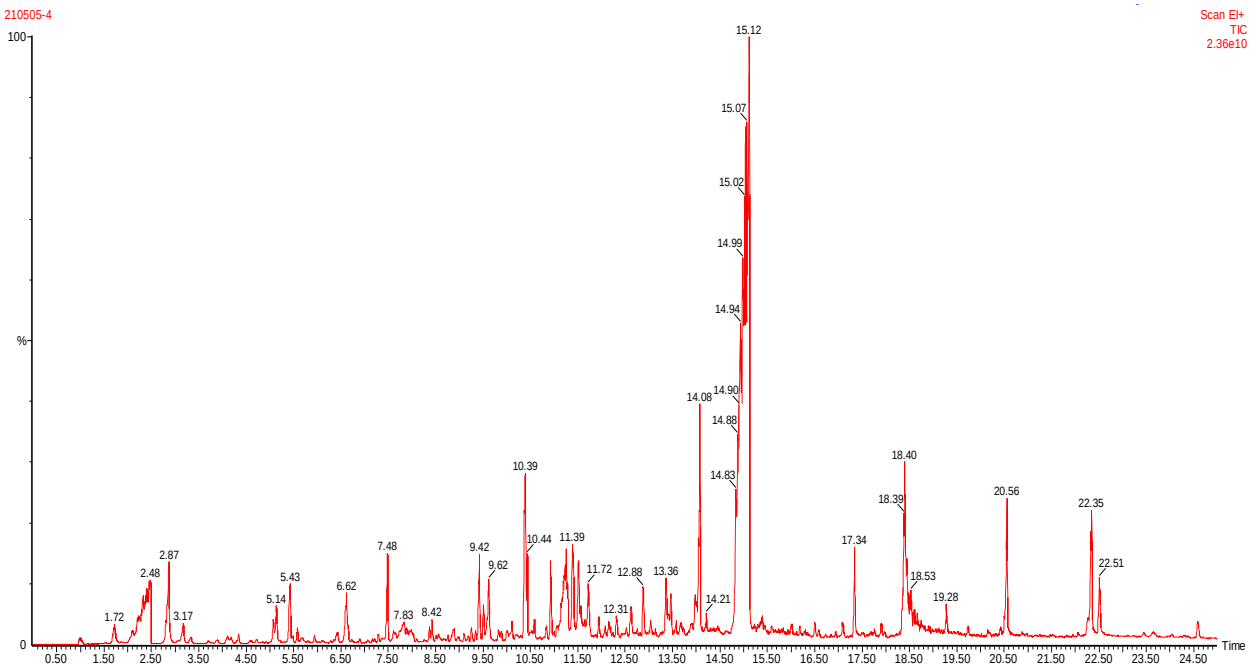

Figure 3.17 Chromatogram of essential oil extracted from 0g/l iron chelate, 400 mg/l GA3, with *L. intermedia*

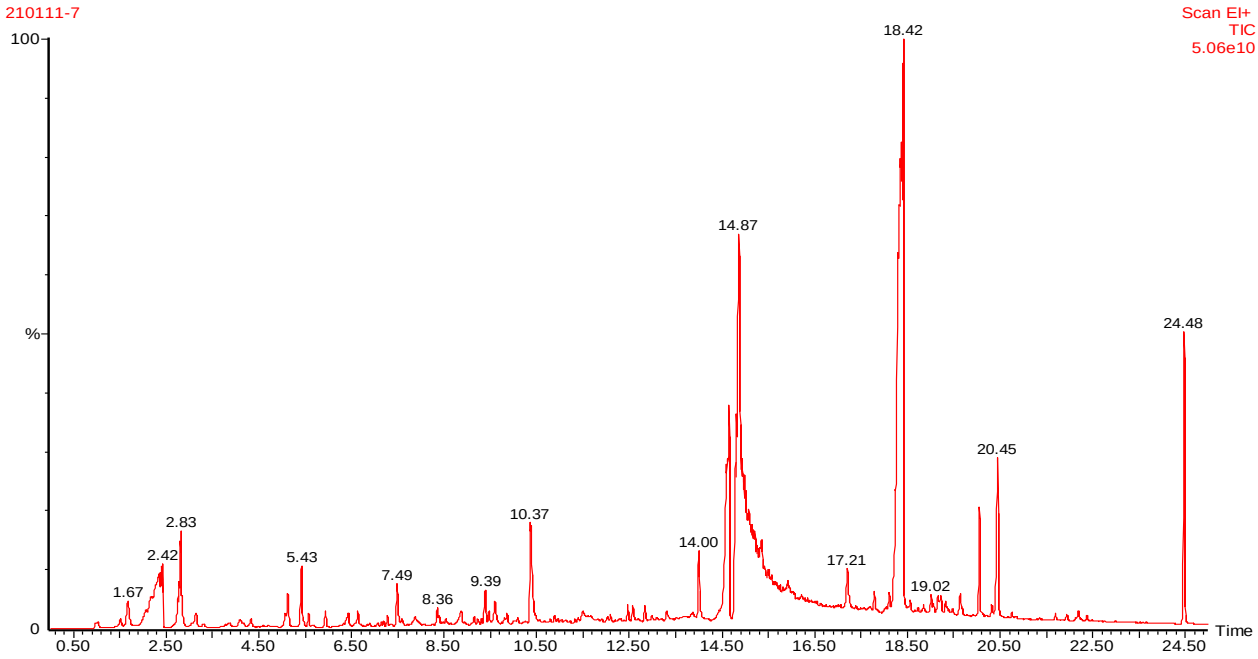

Figure 3.18 Chromatogram of essential oil extracted from 0 g/l iron chelate,0 mg/l GA3 with *L. angustifolia* .

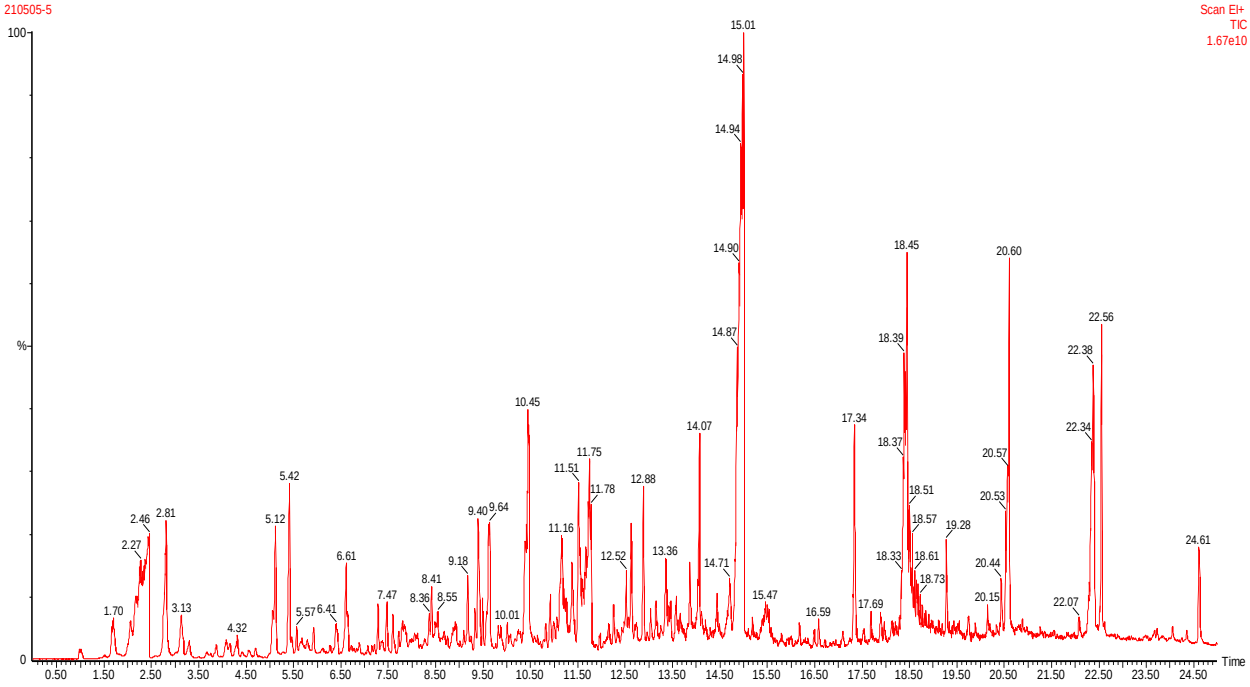

Figure 3.19 Chromatogram of essential oil extracted from 3G/L iron chelate, 200 mg/l GA3, with *L. angustifolia* .

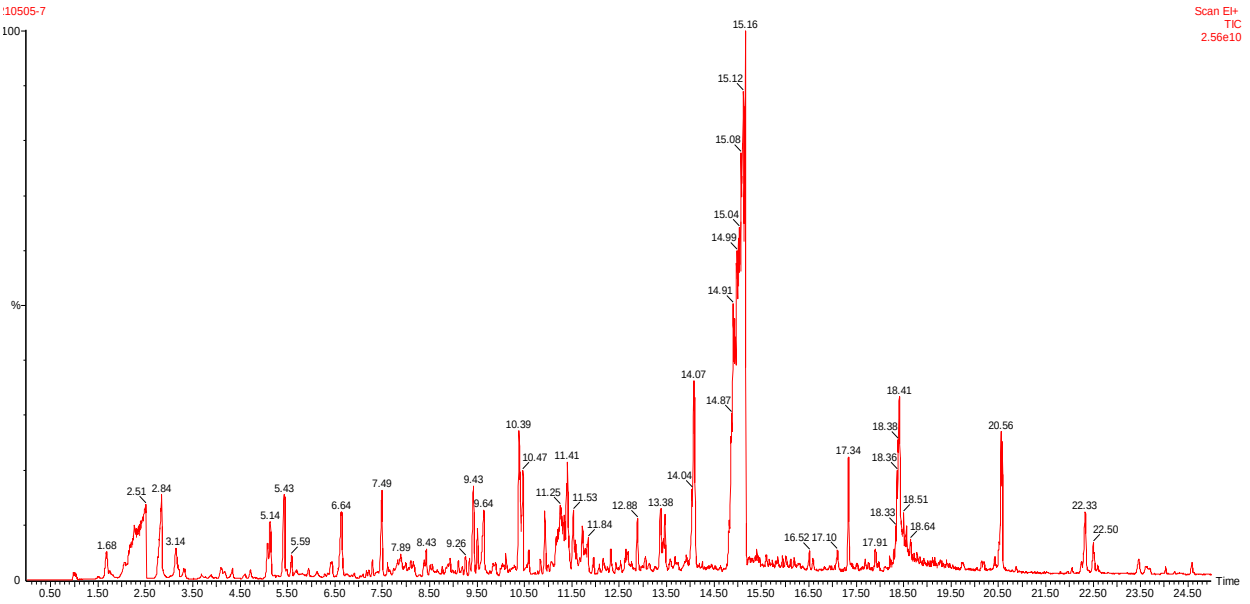

Figure 3.20 Chromatogram of essential oil extracted from2g/l iron chelate, 200 mg/l GA3, with *L. intermedia.*

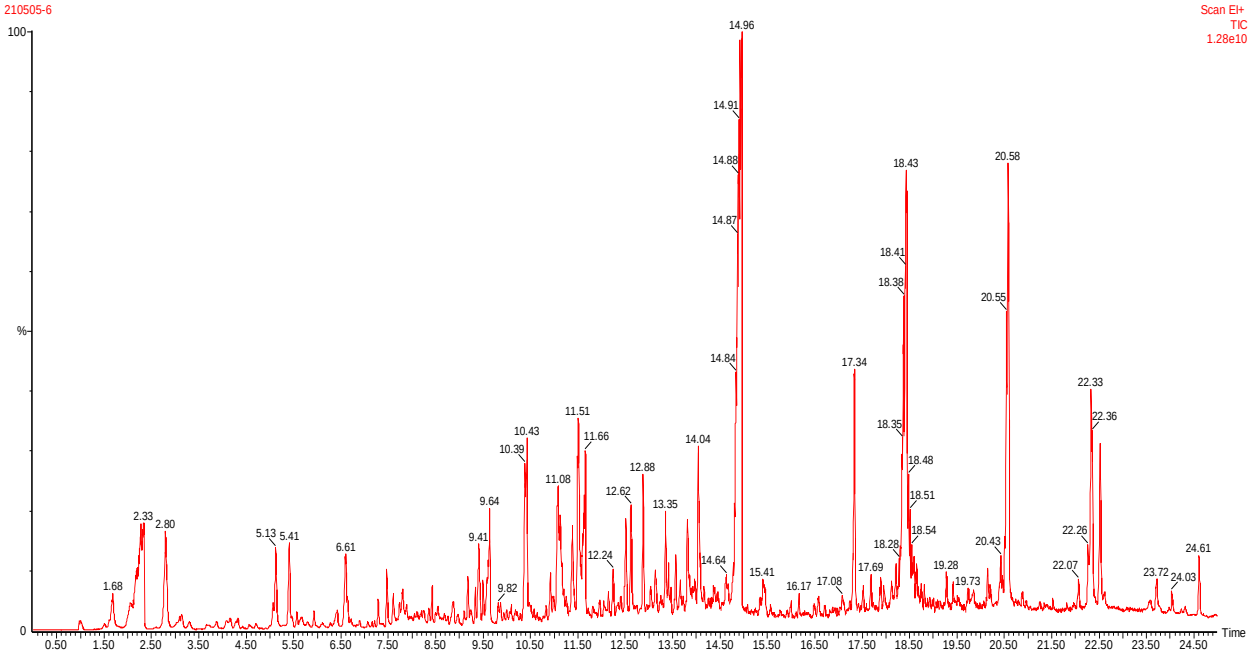

Figure 3.21 Chromatogram of essential oil extracted from 3g/l iron chelate, 200 mg/l GA3, with *L. angustifolia* .

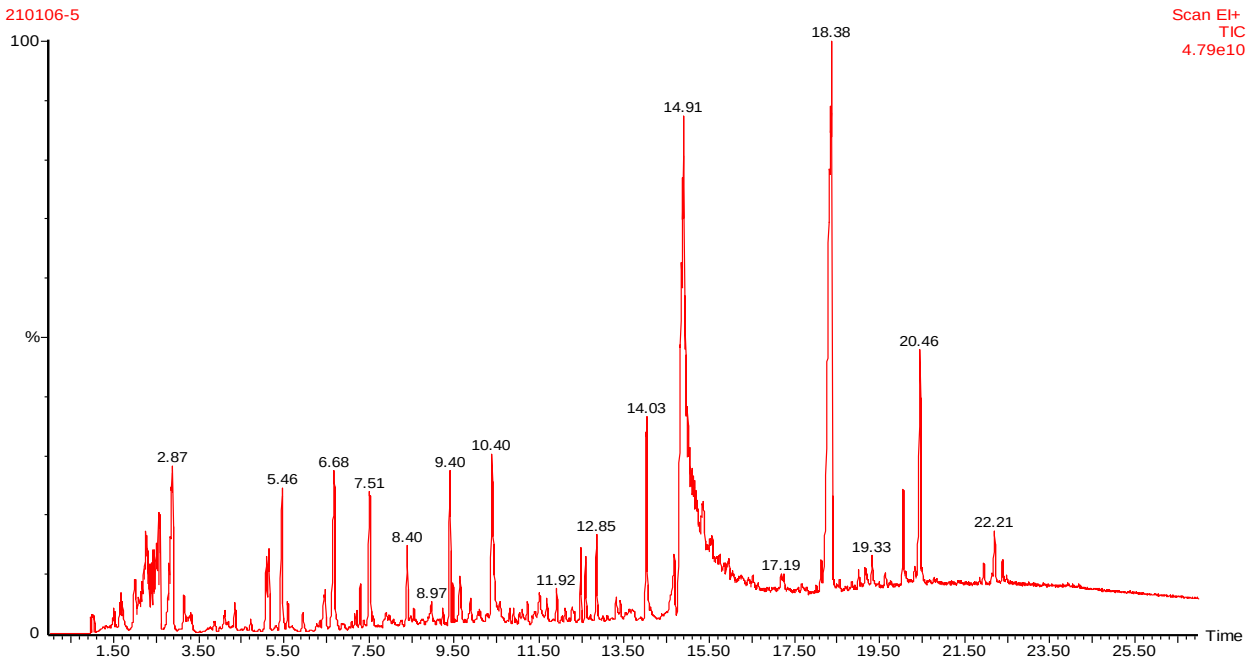

Figure 3.22 Chromatogram of essential oil extracted from 3g/l iron chelate, 0 mg/LGA3, with *L. stoechas*

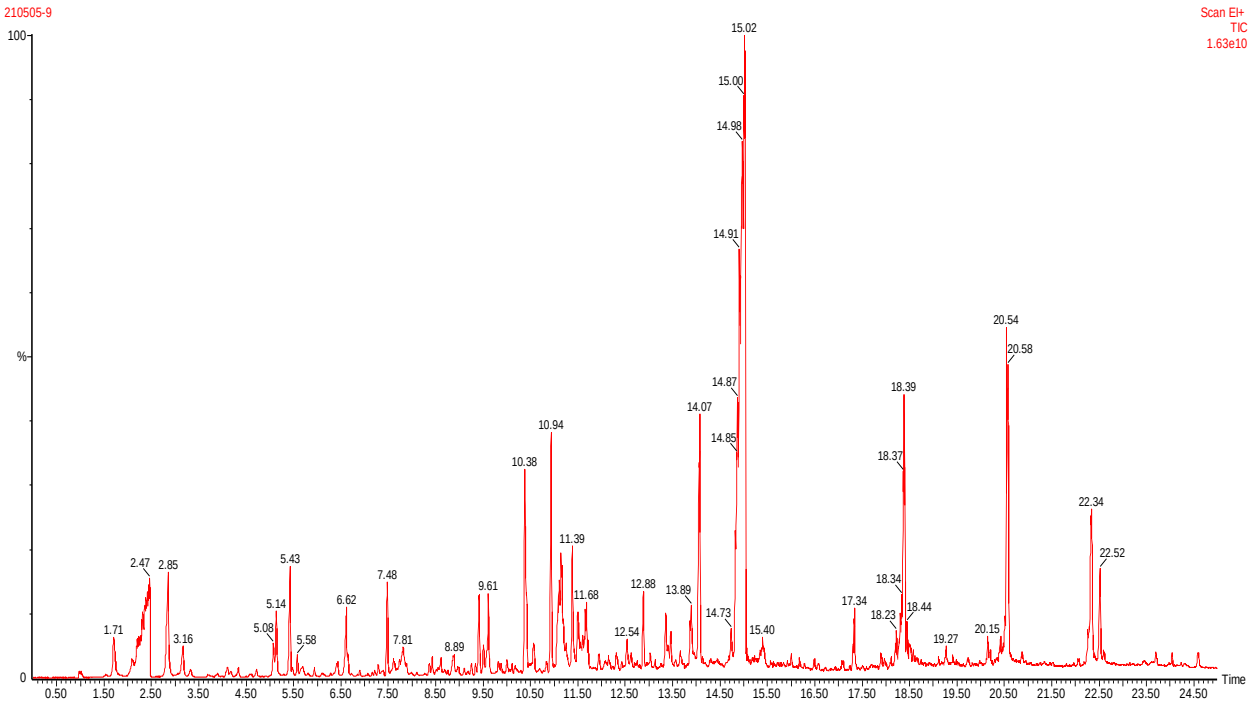

Figure 3.23 Chromatogram of essential oil extracted from 3g/l iron chelate, 200 mg/l GA3, with *L. intermedia*

Table 3.8 GC-MS analysis identification and relative percentage concentration for lavender essential oil with species (*L. angustifolia, L. intermedia,* and *L. stoechas*).

No.	RT(min)	Essential Oil Compound	Formula	percentage %	Molecular class	Identified Methods
1	1.008	Carny acetate	$C_{12}H_{18}O2$	0.192549812		Pyro-GC-MS
2	1.679	Trans- β-Ocimene	$C_{10}H_{16}$	0.439794	Monoterpene	Pyro-GC-MS
3	2.254	Hexanol	$C_6H_{14}O$	1.332310156	Aliphatic alcohol	Pyro-GC-MS
4	2.491	1-Penten -3-ol	C_6H_{12}	0.628748541	Monoterpene	Pyro-GC-MS
5	2.839	α-Pinene	$C_{10}H_{16}$	1.35064767	Monoterpene	Pyro-GC-MS
6	3.233	β-pinene	$C_{10}H_{16}$	0.669156872	Monoterpene	Pyro-GC-MS
7	4.201	3-Carene	$C_{10}H_{16}$	0.393119149	Monoterpene	Pyro-GC-MS
8	4.685	Cis-β -Ocimene	$C_9H_{16}O_2$	0.258659199	Monoterp.alcohol	Pyro-GC-MS
9	5.099	Myrcene	$C_{10}H_{16}$	1.409498356	Monoterpenoids	Pyro-GC-MS
10	5.447	Cymene	$C_{10}H_{16}$	2.287461117	Monoterpene	Pyro-GC-MS
11	5.951	- terpinene-4-ol	$C_{10}H_{16}O$	0.291386732	Monoterpene	Pyro-GC-MS
12	6.319	unknown	/	0.409573426	/	Pyro-GC-MS
13	6.455	Sabinene hydrate	$C_{10}H_{18}O$	0.457893432	Monoterpene	Pyro-GC-MS
14	6.662	Limonene	$C_{10}H_{16}$	1.45379587	Monoterpene	Pyro-GC-MS
15	7.217	Lavandulol	$C_{10}H_{18}O$	0.492380375	Monoterp.alcohol	Pyro-GC-MS
16	7.292	γ-Terpinene	$C_{10}H_{16}$	0.616937659	Monoterpene	Pyro-GC-MS
17	7.358	2-methyl-1-butanal	$C_5H_{12}O$	0.346796732	Monoterp. ether	Pyro-GC-MS
18	7.514	Bicyclo (4.1-0)hept-3-ene	C_7H_{8O}	0.557129136	Monoisotopic	Pyro-GC-MS
19	7.6	Terpinolene	$C_{10}H_{16}$	0.781592822	Monoterpene	Pyro-GC-MS

Table 3.8 (continued)

20	7.781	-7-Trans- Linalool oxide	$C_{10}H_{18}O_2$	0.255958234	Monoterp.alcohol	Pyro-GC-MS
21	7.953	Hexyl butyrate	$C_{10}H_{20}O_2$	0.344285033	Aliphatic ester	Pyro-GC-MS
22	8.402	trans-3-Caren-2-ol	$C_{10}H_{16}O$	1.000915334	Monoisotopic	Pyro-GC-MS
23	8.487	Toluene	C_7H_8	0.667027161	Methylbenzene	Pyro-GC-MS
24	8.966	3,4-hexanedione	$C_6H_{10}O_2$	0.588212315	Monoisotopic	Pyro-GC-MS
25	9.103	Linalool oxide	$C_{10}H_{18}O_2$	0.518074081	Monoterp.alcohol	Pyro-GC-MS
26	9.168	(Z)-Cinnamaldehyde	C_9H_8O	1.176005655	Monoisotopic	Pyro-GC-MS
27	9.319	α- Thujone	$C_{10}H_{16}O$	1.022646462	Monoisotopic	Pyro-GC-MS
28	9.405	Limonene oxide	$C_{10}H_{16}O$	2.141247091	Monoterpene	Pyro-GC-MS
29	9.481	Camphene	$C_{10}H_{16}$	1.251525003	Monoterpene	Pyro-GC-MS
30	9.627	Linalool	$C_{10}H_{16}O$	2.233760957	Monoterp.alcohol	Pyro-GC-MS
31	9.788	Sabinene	$C_{10}H_{16}$	0.422233368	Monoterpene	Pyro-GC-MS
32	10.01	Butanoic acid	$C_4H_8O_2$	0.422149502	Monoisotopic	Pyro-GC-MS
33	10.449	Linalyl acetate	$C_{12}H_{20}O_2$	1.861968756	Monoterp. ester	Pyro-GC-MS
34	10.822	α- Terpinene	$C_{10}H_{16}$	0.653615571	Monoterpene	Pyro-GC-MS
35	10.898	Par-Cymenene	$C_{10}H_{14}$	0.928306709	Monoterpene	Pyro-GC-MS
36	10.973	Camphor	$C_{10}H_{16}O$	0.710154277	Monoterp. ketone	Pyro-GC-MS
37	11.508	3-Caren-10-al	$C_{10}H_{14}O$	0.723946095	Monoterpene	Pyro-GC-MS
38	12.396	Cumin aldehyde	$C_{10}H_{12}O$	0.4123527	Monoterpene	Pyro-GC-MS
39	12.481	D-Carvone	$C_{10}H_{14}O$	1.05792457	Monoterpene	Pyro-GC-MS
40	12.597	Geraniol	$C_{10}H_{18}O$	1.385554837	Monoterp. ether	Pyro-GC-MS

Table 3.8 (continued)

41	12.85	-1,8-Cireole	$C_{10}H_{18}O$	1.328351869	Monoterp.alcohol	Pyro-GC-MS
42	13.485	Cis-Linalool oxide	$C_{10}H_{18}O_2$	1.876236291	Monoterp.alcohol	Pyro-GC-MS
43	14.009	Nerol	$C_{10}H_{18}O$	.792425201	Monoterp.alcohol	Pyro-GC-MS
44	14.115	3- Carene	$C_{10}H_{16}$	0.800928203	Monoterpene	Pyro-GC-MS
45	14.846	α-Terpineol	$C_{10}H_{18}O$	4.592931332	Monoterpene	Pyro-GC-MS
46	15.558	Borneol	$C_{10}H_{18}O$	3.293906627	Monoterp.alcohol	Pyro-GC-MS
47	15.986	Caryophyllene oxide	$C_{15}H_{24}O$	0.910607198	Sesquiterpene	Pyro-GC-MS
48	16.576	-Cadinol	15H	0.549516129	Sesquiterpenoid	Pyro-GC-MS
49	17.257	(E,E)-Farnesol	$C_{15}H_{26}O$	0.895903525	Monoisotopic	Pyro-GC-MS
50	17.625	Thuja-2,4(10)-diene	$C_{10}H_{14}$	0.350204307	Monoterpene	Pyro-GC-MS
51	17.918	Cis-α-Bergamotene	$C_{15}H_{24}$	0.290768582	Sesquiterpene	Pyro-GC-MS
52	18.291	Geranyl acetate	$C_{12}H_{20}O_2$	9.767224029	Monoterp. ester	Pyro-GC-MS
53	18.573	Trans-α-Bergamotene	$C_{15}H_{24}$	0.317684787	Sesquiterpene	Pyro-GC-MS
54	18.71	Di isobutyl phthalate	$C_{16}H_{22}O_4$	0.324690403	Monoterp. ester	Pyro-GC-MS
55	19.32	Myrlenyl acetate	$C_{12}H_{18}O_2$	0.961492764	Monoisotopic	Pyro-GC-MS
56	19.693	Neryl acetate	C12H20O2	0.492731271	Monoisotopic	Pyro-GC-MS
57	19.965	unknown	/	0.308380609	/	Pyro-GC-MS
58	20.071	Trans-Linalool oxide	$C_{10}H_{18}O_2$	3.948007842	Monoterp.alcohol	Pyro-GC-MS
59	20.132	α-Caryophyllene	$C_{15}H_{24}$	0.82842743	Sesquiterpene	Pyro-GC-MS
60	20.374	α- Long pinene	$C_{10}H_{16}$	3.634527334	Monoterpene	Pyro-GC-MS
61	20.576	E-Caryophyllene	$C_{15}H_{24}$	10.08404173	Sesquiterpene	Pyro-GC-MS

Table 3.8 (continued)

62	20.818	β-Caryophyllene	$C_{15}H_{24}$	0.265155555	Sesquiterpene	Pyro-GC-MS
63	20.949	1-Tricosene	$C_{23}H_{46}$	0.1780754	Monoisotopic	Pyro-GC-MS
64	21.877	Germacrene D	$C_{15}H_{24}$	0.306505569	Sesquiterpenes	Pyro-GC-MS
65	21.962	Neryl acetate	$C_{12}H_{20}O_2$	0.880270706	Monoterp. ester	Pyro-GC-MS
66	22.063	Benzaldehyde	C_6H_5CHO	0.212411536	Monoisotopic	Pyro-GC-MS
67	22.245	α-Cedrene	$C_{15}H_{24}$	4.166936997	Monoterpenoids	Pyro-GC-MS
68	22.426	Lavandulyl acetate	$C_{12}H_{20}O_2$	2.330746741	Monoterp.alcohol	Pyro-GC-MS
69	22.497	α-Bergamotene	$C_{15}H_{24}$	0.638138505	Sesquiterpene	Pyro-GC-MS
70	22.779	Methoxy cinnamaldehyde	$C_{10}H_{10}O_2$	0.191081033	Aliphatic alcohol	Pyro-GC-MS
71	23.082	α-Bulnesene	$C_{15}H_{24}$	0.19898576	Sesquiterpenoids	Pyro-GC-MS
72	24.086	Eugenol	$C_{10}H_{12}O_2$	0.252126818	Phenylpropene	Pyro-GC-MS
73	24.202	Limonene diol	$C_{10}H_{18}O_2$	0.271911769	Monoterpene	Pyro-GC-MS
74	24.514	Cadala-1(10)-3-8-triene	$C_{15}H_{22}$	7.885120051	Monoisotopic	Pyro-GC-MS
75	24.6	Trans-Cadinene ether	$C_{15}H_{24}$	0.577726498	Monoisotopic	Pyro-GC-MS
76	24.716	α-Element	$C_{15}H_{24}$	0.148524836	Actinium	Pyro-GC-MS

Total Molecular class identified of essential oils in this study are:

Monoterpene hydrocarbons
Sesquiterpene hydrocarbons
Oxygenated monoterpenes
Oxygenated sesquiterpenes
Esters
Others

Table 3.9 GC-MS Data for essential oil components identified in lavender. Total chromatogram (GC-MS) of lavender (leaves, flowers, and stems) essential oil, name, and retention time (min) of compounds: linalool (10.73), linalyl acetate (7.64), Camphor (7.31) , Cis-linalool (2.29), Lavandulol (5.94), Camphene (3.28), α -pinene (7.67), myrcene (4.94).

	RT	Compound	Treatment														
			1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
1	1.008	Caray acetate		-				0.17		0.17							
2	1.679	Trans-β-Ocimene	0.27	0.90	1.02		0.38	0.85		0.85	0.42	0.83	0.67	0.54	0.68	0.72	0.72
3	2.254	Hexanol	1.22	1.51	2.40		2.07	3.54	3.36	2.05	0.87	2.49	1.81	2.53	3.08	2.51	1.06
4	2.491	1-penten-3-ol		1.60	2.59		4.68		3.07	3.19			3.26	3.78	1.18	3.08	2.18
5	2.839	α-Pinene	0.95	2.93	4.87		3.64	7.67	2.94	2.29	1.93	3.98	1.50	6.14	5.49	5.00	1.69
6	3.233	β-pinene		0.30	1.21		0.71	0.58	0.18	0.20	0.12	0.43	1.55	1.19			0.31
7	4.201	3-Carene					0.20		0.19	0.19	0.11		0.22	0.14		0.51	0.16
8	4.685	Cis- β-Ocimene		-			0.11		0.13								
9	5.099	Myrcene		1.83	1.63		1.79	0.87	0.35	1.26	4.94	1.01	0.96	0.45	1.80	1.77	0.44
10	5.447	Cymene		2.65	4.24		1.45	2.61	1.34	1.77	0.71	1.89	1.47	1.25	3.07	4.14	0.65
11	5.951	-terpinen-4-ol					2.03	0.24	0.93	0.22	0.11		0.50				
12	6.319	Unknow															
13	6.455	Sabinene hydrate	-				1.21	0.23	0.60		0.23		0.42	033	0.31	0.97	0.65
14	6.662	Limonene			2.87		0.11	1.75	0.38	1.24	1.06		1.11	1.34	1.13	4.55	2.26
15	7.217	Lavandulol		1.03					0.35		1.30						
16	7.292	γ-Terpinene		1.03			0.32	0.37	1.38	0.39			0.30	0.15	0.21		
17	7.358	2-methyl-1-butanol						0.13	0.12				0.35	1.12			
18	7.514	Bicyclo (4.1.0)hept-3	0.61		2.29		0.81	2.31	3.98	0.32	0.16	1.30	1.98	2.37		3.68	5.33
19	7.6	Terpinolene					0.34		0.57	0.42			0.31		0.27		0.64
20	7.781	-7trans-Linalool oxide			0.23	1.20	0.81	0.58	0.63		1.32	0.26	0.26	0.26	0.37	0.38	0.64
21	7.953	Hexyl butyrate						0.29	0.17	0.18	0.41			0.27			0.11
22	8.402	trans-3-Caren-2-ol		1.19	0.33		0.180 .23	0.34	0.48	0.34	0.18	0.42	0.45	0.29	0.23	1.53	1.90

Table 3.9 (continued)

23	8.487	Toluene	0,86	1.68				0.30	0.37	0.20	1.20		0.25		0.24		0.30
24	8.966	3,4-hexanedione	-	0.95			0.46	0.83	0.26	0.16	0.28	0.44	0.16	0.17	1.05		
25	9.103	Linalool oxide					0.14	0.15					0.50		0.44		0.90
26	9.168	(Z)-cinnamaldehyde					0.10		0.46	0.25			1.86	0.24			4.16
27	9.319	α-Thujone		1.24			0.12	0.23	0.17	0.39	0.18		0.28	0.14	0.18		0.14
28	9.405	Limonene oxide			2.59	2.48		1.62	4.66	1.07	1.10		3.32	1.20	0.89	3.59	8.83
29	9.481	Camphene		1.17	1.34		1.00	0.32	2.09	0.30	0.33		1.80	0.34	0.26		3.28
30	9.627	Linalool			1.71		1.00	1.56	1.38	0.56	1.00		1.71	1.09	10.73	0.84	2.53
31	9.788	Sabinene					0.20		0.38		0.16		0.18	0.29	0.16		1.22
32	10.01	Butanoic acid					0.21	0.44	0.25	0.24	0.18		0.19	0.14			0.72
33	10.449	Linalyl acetate	3.01	8.11	7.46		2.79	24.26	1.10	7.64	0.60		4.18	1.18	2.83	4.60	0.44
34	10.822	α-Terpinene	1.43	0.68			0.21	0.13			0.18		0.16	0.14			2.48
35	10.898	Paracymene	0.51						0.29	0.47					3.18		0.56
36	10.973	Camphor		14.30	7.31		0.13	0.26	0.60	0.42	1.00		0.39	0.71	0.35		0.52
37	11.508	3-Caren-10-al	0.99		0.79		0.95	1.15	1.15	1.26	0.90	0.35	1.89	0.24	0.90	0.40	1.08
38	12.39	Cumin aldehyde					0.20	0.45	0.23	0.16	0.33			0.27			0.25
39	12.48	D-carvone		1.47	1.16		0.19	0.20	0.40	0.69			0.32	0.14	0.92	1.41	0.31
40	12.59	Geraniol	0.36	0.97			0.76	0.38	0.41	0.80	0.50		0.70	0.17	1.03	1.31	0.55
41	12.85	-1,8-Cireole	0.29				10.87	1.66	4.01	0.90	7.56	4.31	0.73	22.22	0.91		21.53
42	13.485	Cis-Linalool oxide	0.49		0.87	0.40	0.50	1.00	0.87	0.32	0.54		0.88	0.54	0.19	0.44	2.29
43	14.00	Nerol	0.35		3.46		3.46	4.95		1.32	2.78	2.66	1.42	1.21	1.70	5.51	0.28
44	14.115	Z-caryphyllene					2.92		5.04	0.25	0.24		0.18				3.10

Table 3.9 (continued)

45	14.846	α-Terpineol	16.11		19.03	38.87	89.43	5.07	0.35	31.76	16.7	17.95	12.29	7.01	49.34	1.72	0.96
46	15.558	Borneol		2.74	1.98		0.15	0.13		0.25	0.11		0.32		0.67		0.22
47	15.986	Caryophyllene oxide												0.15			0.32
48	16.576	-Cadinol					0.18	0.12			0.18		0.18	0.19	0.21		0.12
49	17.257	(E,E)-Farnesol		1.03	1.53			0.80	1.25	1.43	0.84	1.71	1.73	1.04	1.60	0.45	0.30
50	17.625	Limonene oxide					0.14	0.20	1.98	0.19		0.54	0.19		0.27		1.19
51	17.918	Cis-α-Bergamotene				0.53				0.24	0.22		0.25	0.30	0.24		0.12
52	18.291	Geranyl acetate			18.74	5.00		0.80	3.97	0.88		3.35	33.87	7.47	0.26		0.59
53	18.573	Trans-α-Bergamotene	1.33				0.32		0.31	0.21	0.50	25.88	0.48	0.87	0.18		0.14
54	18.71	Diisobutylphthalate					0.20		0.15		0.15		0.20	0.28	0.22		
55	19.32	Myrlenyl acetate	0.39				0.18	0.18	0.36	0.42	0.27		0.58	0.16	0.19		0.25
56	19.693	Neryl acetate	1.36	3.80					0.24	0.36	0.87	0.58	0.23	0.17	0.27		0.15
57	19.965																
58	20.071	Trans-Linalool oxide		0.31						0.55			1.88			2.71	
59	20.132	α-caryophyllene	1.30			2.08				0.50	0.15		0.19		0.33		0.29
60	20.374	α-Long pinene	1.40	1.04	1.36	26.56	3.01	5.86	0.12	0.90	1.70	6.42		0.21	0.54		0.52
61	20.576	E-Caryophyllene	20.42				1.41	0.59	3.08	3.56	1.84		3.02	1.54	4.56		3.85
62	20.818	β-caryophyllene	2.70						0.19	0.16							0.22
63	20.949	1-Tricosene						0.19									
64	21.877	Germacrene D															
65	21.962	Unknow															
66	22.063	Benzaldehyde													0.29		
67	22.245	Lavendulol	5.94	0.16		7.82	0.33	0.14	1.08	0.36	2.17		4.44	0.98	0.43	0.93	0.35

Table 3.9 (continued)

68	22.426	Lavandulyl acetate	2.48			2.78	2.86		0.40	0.72			1.01		1.51		
69	22.497	α-Bergamottin					1.49	0.25	1.34	0.26			1.83	0.41	0.19		1.06
70	22.779	Methoxy cinnamaldehyde															
71	23.082	α -Bulnesene						45.37	0.54								
72	24.086	-Eugenol													0.21		0.13
73	24.202	Limonene diol															
74	24.514	Geraniol		1.54	2.59			0.24	0.76		0.32	11.81	17.37	4.65			
75	24.6	Trans-Cadinene ether								0.34		0.87			0.55		0.26
76	24.716	α-Element															

1=2g/l iron chelate, 400 mg/l GA3, with *L. angustifolia*
2=1g/l iron chelate, 400 mg/l GA3, with *L. intermedia*
3=2g/l iron chelate, 400g/l GA3, with *L. intermedia*
4=0g/l iron chelate, 0mg/l GA3 with *L. stoechas.*
5=1g/l iron chelate, 200mg/l GA3, with *L. stoechas*
6=1g/l iron chelate, 400mg/l GA3, with *L. stoechas*
7=2g/l iron chelate, 200mg/l GA3, with *L. stoechas*
8=3g/l iron chelate, 400mg/l GA3, with *L. angustifolia* .
9=0g/l iron chelate, 400mg/l GA3, with *L. intermedia*
10=0 g/l iron chelate,0mg/l GA3 with *L. angustifolia* .
11=3G/L iron chelate, 200mg/l GA3, with *L. angustifolia* .
12=2g/l iron chelate, 200mg/l GA3, with *L. intermedia.*
13=3g/l iron chelate, 200mg/l GA3, with *L. angustifolia* .
14=3g/l iron chelate, 0g/LGA3, with *L. stoechas*
15=3g/l iron chelate, 200mg/l GA3, with *L. intermedia , RT= Retention Time, unit measure=%*

Statistical analysis for GC-MS

The study which included 3 rates of GA_3, 4 rates of iron chelated, and 3 species of lavender resulted in 36 treatments which were replicated 5 times and the study repeated twice. This resulted in 360 samples for analysis. This would be costly and require a long time to complete the analysis of essential oils using GC-MS technology. To reduce analysis, duplicates of each treatment were combined, and one homogeneous sample was analyzed to represent that treatment in the statistical analysis (Table 3.10) (Fig. 3.24). For estimating the significance of the statistical differences, the mean of the control parameters was used as a constant for the purpose of the assay using the T test of one sample at a probability level of 0.05 of SPSS (Statistical Package for the Social Sciences version 26, IBM Inc., Armonk, NY) (Daniel, 1974).

Table 3.10 Results of analysis of effect of iron chelate and gibberellic acid in diagnosing essential oils of *Lavandula* using Gas Chromatography Mass Spectroscopy (GC-MS).

Essential oils compounds								
Samples[z]	Linaloo l	Linalyl acetate	Camphor	Cis-Linalool	Lavandulol	Camphene	α-Pinene	Myrcene
I^2 × Angustifolia × GA^{400}	0.02	3.01	0.04	0.49	5.94	0.03	0.95	0.02
I^1× Intermedia × GA^{400}	0.05	8.11	14.3	0.03	0.16	0.35	2.93	1.83
I^2 × Intermedia × GA^{400}	1.71	7.46	7.31	0.87	0.3	1.34	4.87	1.63
Stoechas	0.04	0.02	0.03	0.4	7.82	0.02	0.05	0.03
I^1 × Stoechas × GA^{200}	1.0	2.79	0.13	0.5	0.33	1.0	3.64	1.79
I^1 × Stoechas × GA^{400}	1.56	24.26	0.26	1.0	0.14	0.32	7.67	0.87

Table 3.10 (continued)

I^2 × Stoechas	1.38	1.1	0.6	0.87	1.08	2.09	2.94	0.35
I^4 × Angustifolia × GA^{400}	0.56	7.64	0.42	0.32	0.36	0.3	2.29	1.26
Intermedia × GA^{400}	1.0	0.6	1.0	0.54	2.17	0.33	1.93	4.94
Angustifolia	0.05	0.03	0.02	0.04	0.02	0.04	3.98	1.01
I^4 × Angustifolia × GA^{200}	1.71	4.18	0.39	0.88	4.44	1.8	1.5	0.96
I^2 × Intermedia × GA^{200}	1.09	1.18	0.71	0.54	0.98	0.34	6.14	0.45
I^2 × Angustifolia × GA^{200}	10.73	2.83	0.35	0.19	0.43	0.26	5.49	1.8
I^4 × Stoechas	0.84	4.6	0.03	0.44	0.93	0.04	5	1.77
I^4 × Intermedia × GA^{200}	2.53	0.44	0.52	2.29	0.35	3.28	1.69	0.44
Constant value	0.03	0.05	0.04	0.2	3.91	0.02	1.99	0.505
(*p* value=0.05)	0.033	0.012	0.109 ns	0.011	0.00	0.00	0.022	0.027
T test	2.363	2.876	1.71	2.951	-3.569	3.047	2.574	2.461

[z]I=iron chelate at 1, 2 or 4 g/L: GA_3= Gibberellic Acid at 200 or 400 mg/l

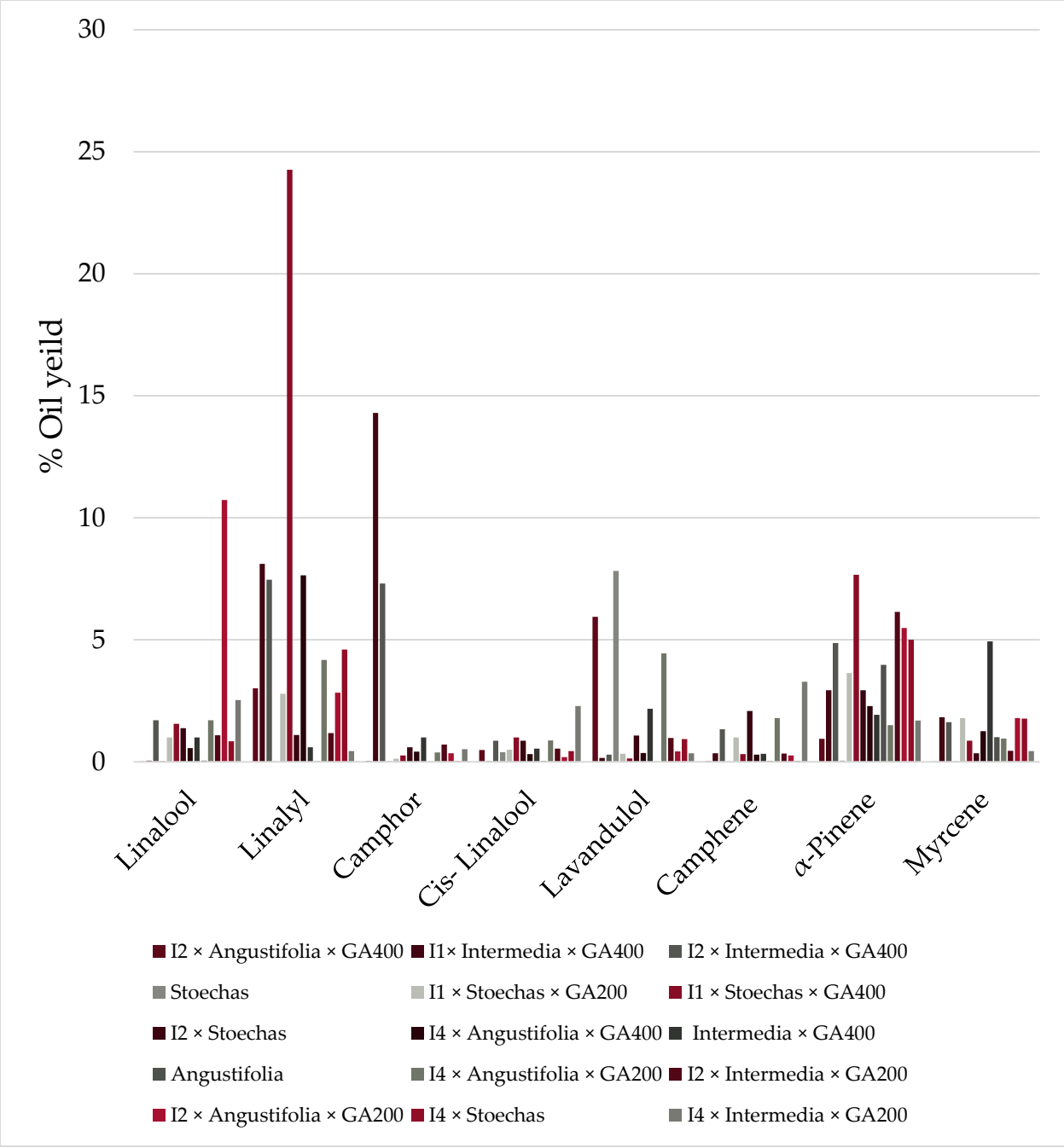

Figure 3.24 Figure: 3.24 Lavender essential oil components identified by GC-MS from treatments with iron chelate and GA_3.

Essential oil components and their rates obtained with GC-MS in lavender species are shown in (Table 3.10). Linalool, linalyl acetate, camphor, Cis linalool, lavandulol, , Camphene, α-pinene, myrcenes, was determined to be the main essential oil constituents in the dried foliar and floral tissues of lavender. The highest linalool content was determined to occur in plants treated with iron 2g/L × *L. angustifolia* × 200 mg/L GA_3. The lowest linalool content was obtaine from treatment with iron 2g/l × *L. angustifolia* × 400 mg/l GA_3. The highest linalyl acetate content was found in plants treated with iron 1g/l × *L. stoechas* × 400 mg/l GA_3. Iron chelate increased the proportion of linalool in the oil compared to other treatments. Camphor highest content was in plants treated with iron 1g/l× *L. intermedia* ×400 mg/l GA_3 and iron 2g/l ×*L. intermedia* ×400 mg/l GA_3. The highest Cis- Linalool content was from the treatment iron 4g/l × Intermedia × 200 mg/l GA_3 and iron 4g/l × *L. angustifolia* × 200 mg/l GA_3. The lowest Cis-linalool content was obtained from treatment iron1g/l× *L. intermedia* ×400 mg/l GA_3. The highest Lavandulol content was obtained when treated with iron 2g/l× *L. angustifolia* ×400mg/l GA_3 and *L. stoechas*. The lowest Lavandulol content was obtained from *angustifolia*. The highest Camphene content was determined from treatments with iron 4g/l × *L. intermedia* × 200 mg/l GA_3 and iron 2g/l× *L. stoechas*. The lowest Camphene content was obtained from *L. stoechas*. The highest α-Pinene content was determined from treatment iron 1g/l × *L. stoechas* ×400 mg/l GA_3 and iron 2g/l × *L. intermedia* × 400 mg/l GA_3. The lowest α-Pinene content was obtained from *L. stoechas*. The highest Myrcene content was determined from *L. intermedia* × 400 mg/l GA_3 and iron 1g/l × *L. intermedia* × 400 mg/l GA_3.

There are significant differences between the species and linalool, linalyl acetate, Cis-Linalool, Lavandulol, Camphene, α-Pinene, and Myrcene compounds in essential oils of lavender. There were no differences between the species, iron chelate levels, or GA_3, for

camphor essential oils content. Depending on the different species of lavender, differences in chemical composition have been found. Generally, *L. angustifolia* was higher linalool or linalyl acetate content in all the lavender cultivars. These results are agreement with what Smigielski et al. (2009) reached from analyzes of the essential oil from *L. angustifolia.* Seventy-eight compounds have been identified making up 99.4% of the total essential oil. The main components were Linalool, Linalyl Acetate, Cis linalool, lavandulol, camphene, α -pinene, myrcene. Baydar and Kineci (2009) reported the main compounds in lavender were Linalyl acetate, linalol, alpha bisabolol, borneol, camphor, Farnesyl acetone, caryophylline oxide, and 1.8-cineole. In lavender, volatile solvents and extraction procedures can affect the yield and composition of the essential oil. Guo and Wang (2020) reported that a total of thirty-three volatile compounds were determined by GC-MS (extraction with pure water) of lavender volatile substances belonging to different chemical groups, including alkenes, alcohols, ketones, esters, and aldehydes. Linalool was the most abundant alcohol in the samples and was considered the key aroma-active compound imparting a floral note for lavender (Wang et al., 2000; Djenane et al., 2012).

Fu et al. (2017) reported that the GC-MS analysis, was able to identify 39 compounds from three varieties of lavender. Jalali-Heravi et al. (2015) reported a total of 143 components identified by the GC-MS for Iranian lavender essential oil from which 47 had concentrations greater than 0.1%. These components accounted for 89.90% of the total relative content of the lavender essential oil. This research also demonstrated that the Iranian lavender essential oil is rich in **α**-pinene and could be a good source for this compound. On the other hand, Zagorcheva et al. (2013) indicated the GC / MS analysis identified a total of 32 individual volatiles including all 11 volatiles listed in the ISO 3515 lavender standard. In addition, the relationship of the GC-

MS data on volatiles to the composition of distilled lavender essential oils was assessed by analyzing the volatile recovery rates of the analyzed species. Dong et al. (2020) reported the chemical composition of lavender essential oil obtained from aerial parts by pGC and GC-MS equipped with three capillary columns of different polarity was studied for the first time and 15 newly identified compounds were identified. Stierlin et al. (2020) indicated thirty compounds were separated and detected in all lavender above-ground samples in only 3 min of analysis by GC-MS. Tavallali et al. (2019) indicated that in total, 72 compounds were identified accounting for over 99% of the total oils. A few compounds belonging to different chemical classes have been reported, among which phenylpropanoids have been shown to play a significant role in the establishment of chemotypes in *O. basilicum*: linalool, methyl chavicol, linalool-methyl chavicol, linalool-eugenol, and methyl chavicol-methyl eugenol chemotypes are common ones. Nurzyńska-Wierdak (2013) explained that the essential oil biosynthesis in plants has a genetic determination and environmental conditions play an important role. The growing conditions (such as the method of planting, fertilization, and irrigation) largely determine the productivity of raw materials, as well as the quality of the raw materials. Chemically speaking, essential oils are very diverse compounds, as they are biosynthesized in plants along different metabolic pathways, through the presence and availability of nutrients in the surrounding environment.

Conclusion

This study dealt with application of iron chelate and GA_3 to three species of lavender to enhance growth, flowering, and essential oil yield. English lavender *(L. angustifulia)* responded most favorably with increased growth and essential oil yield followed by *L.* x *intermedia* to the application of GA_3. The increasing trend of growth and oil yield with the highest tested rates of GA_3 makes it possible that higher levels of GA_3 may need to be explored in future research on

this high-value crop. Foliar applications of iron chelate also enhanced growth and essential oil yield. The combination of 400 mg GA_3/L with 3 g iron chelate/L resulted in the greatest plant response. *L. angustifulia* responded most favorably with increased growth and essential oil yield followed by *L.* x *intermedia* and then *L. stoechas*.

Literature Cited

Abd-Elkader, D.Y. 2016. Effect of foliar spraying with micronutrients and salicylic acid on growth, yield, and quality of garlic plants. J. Agric. Sci. 61(6): 649-658.

Abdul C. Jaleel, G. Wang, P. Ahmad, and U.H. Ikram-. 2009. Changes in the photosynthetic characteristics of (*Catharanthus roseus L.*) because of exogenous growth regulators. Plant Omics. J. 2(4): 169-174.

Adams R. P. 1995. Identification of essential oil by GC–MS. Allured Publ. Corp. Carol Stream IL.

Akter, E., A. Ali, M.M.Z. Islam, R. Karim, A.H.M. Razzaque. 2007. Effect of GA_3 on growth and yield of mustard. Int. J. Sustain. Crop Prod. 2(2): 16-20.

Amini, M. S. Yousefzadeh, and K. Sadat- Asilan. 2018. A study on variations of essential oil yield and composition of *Hyssopus officinalis* L. affected by foliar application of zinc, iron, and manganese. Iranian J. Medicinal Aromat. Plants. 34(1):131-143.

Amir-Rezaie E. 2011. Investigation yield, nutrient uptake amount and antioxidant enzymes of 2 imported varieties of *Amaranthus retroflexus*. Master's thesis. Islamic Azad University of Jiroft, Iraq.

Amuamuha, L., A.R. Pirzad, H. Hadi. 2012. Effect of varying concentrations and time of nanoiron foliar application on the yield and essential oil of pot marigold. Int. Res. J. Appl. Basic Sci. 3(10): 2085-2090.

Baliah, N.T., P.C. Sheeba, and S. Mallika. 2018. Encouraging effect of gibberellic acid on the growth and biochemical characters of green gram (*Vigna radiata L.*). J. Global Biosci. 7(8): 5522-5529.

Baliah, N.T., P.C. Sheeba, and S. Mallika. 2018. Encouraging effect of gibberellic acid on the growth and biochemical characters of green gram (*Vigna radiata L.*). J. Global Biosci. 7(8): 5522-5529.

Basch E., I. Foppa, R. Liebowitz. 2004. Lavender (*Lavandula angustifolia* Miller). J. Herb. Pharmacother. 4(2): 63-68.

Baydar, H. and S. Kineci. 2009. Scent composition of essential oil, concrete, absolute and hydrosol from lavandin (*Lavandula* x *intermedia* Emeric ex Loisel.). J. Essent. Oil-Bear. Plants. 12(2): 131-136.

Bettina M. and S. Helge. 2015. The essential oil maker's handbook, IV TP958.M27513, 661: 806.

Betts T.J. 1992. Possible value for the gas chromatographic analysis of essential oils of some unusual phase commercial capillaries, J. Chromatogr. 626(2): 294-300.

Brittenham. G.M. 1994. New advances in iron metabolism, iron deficiency and iron overload Curr. Opin. in Hematol. 1(2): 549-556

Byers , R. E., H . D . Carbough and C.N. Presleyu. 1990. "Styman" Fruit cracking as effected by surfactants plant growth regulators and other chemicals J. Amer . Soc. Hort. Sci. 115(3): 405-411.

Cavanagah H. M., J.M. Wilkinson. 2002. Biological activities of lavender essential oil, Phytother Res. 16(4): 301-308.

Cseke, L. J., P.B. Kaufman, A. Kirakosyan, S.L. Warber, J.A. Duke, and H.L. Brielmann. 2006. Regulation of metabolite synthesis in plants. Natural Products from Plants. (Ed. 2): 101-141.

Daneshkhah M, M. Kafi, A. Nikbakht, M.H. Mirjalili. 2007. Effect of different levels of nitrogen and potassium on yield indicators and oil of *Rosa damascena*. J. Hortic. Sci. Technol. 8(2): 83-90.

Demas S., M. Caser, M. Lonati, P.L. Cioni, L. Pistelli, B. Najar, V. Scariot. 2018**.** Latitude and altitude influence secondary metabolite production in peripheral alpine populations of the mediterranean species Lavandula angustifolia Mill. Front. Plant Sci. 9,983.

Djenane, D., M. Aïder, J. Yangüela, L. Idir, D. Gómez, P. Roncalés. 2012. Antioxidant and antibacterial effects of *Lavandula* and *Mentha* essential oils in minced beef inoculated with *E. coli* O157:H7 and *S. aureus* during storage at abuse refrigeration temperature. Meat Sci. 92(4): 667-674.

Dong, G., X. Bai, A. Aimila, H.A. Aisa, and M. Maiwulanjiang. 2020. Study on lavender essential oil chemical compositions by GC-MS and improved pGC. Molecules. *25*(14): 3166.

Daniel, W.W., 1974. Biostatistics; a foundation for analysis in the health sciences (No. 574.0182 D3).

El-Naggar, A. H.A.A.M. El-Naggar, N.M. Ismaiel. 2009. Effect of phosphorus application andgibberellic acid on the growth and flower quality of (*Dianthus caryophyllus* L.). American-Eurasian J. Agric. and Environ. Sci. 6(4): 400-410.

Fahmy, A.A. and W.S. Nosir. 2021. Influence of chitosan and micronutrients (fe+ zn) concentrations on growth, yield components and volatile oil of lavender plant. SJFOP. 8(1): 87-100

Fu, J., J. Zhao, Y. Zhu, and J. Tang. 2017. Rapid analysis of the essential oil components in dried lavender by magnetic microsphere-assisted microwave distillation coupled with HS-SPME followed by GC-MS. Food Anal. Methods. 10(7): 2373-2382.

González-Coloma, A., C. López-Balboa, O. Santana, M. Reina, and B.M. Fraga. 2011. Triterpene-based plant defenses. Phytochem. Rev. 10(2); 245-260.

Guo, X. and P. Wang. 2020. Aroma characteristics of lavender extract and essential oil from *Lavandula angustifolia* Mill. Molecules, 25(23): 5541.

Hassanpouraghdam, M.B., B.H. Asl, and A. Khalighi. 2011. Gibberellic acid foliar application influences growth, volatile oil, and some physiological characteristics of lavender (*Lavandula officinalis* Chaix.). Rom. Biotechnol. Lett. *16*(4): 6322-6327.

Hassanpouraghdam, M.B., S.J. Tabatabaie, H. Nazemiyeh. 2008. Aflatuni.N and K nutrition levels affect growth and essential oil content of costmary (*Tanacetum balsamita* L.). *J.* Food, Agric. Environ. 6(2): 145-149.

Haynes, R.J., and K.M. Goh. 2013. Evaluation of potting media for commercial nursery production of container grown plant. New Zealand J. Agric. Res. 20(3): 371-381.

Huang, L.S., M.J. Abuhamdah, C.L. Howes, M.S. Dixon, C. Elliot, C. Ballard. 2008. Pharmacological profile of essential oils derived from *Lavandula angustifolia* and *Melissa officinalis* with anti-agitation properties: focus on ligand-gated channels. Journal of Pharm. Pharmacol. 60(11): 1515-1522.

Iqbal, N., R. Nazar, M.I.R. Khan, A. Masood, and N.A. Khan. 2011. Role of gibberellins in regulation of source–sink relations under optimal and limiting environmental conditions. Curr. Sci. 100(7): 998-1007.

Jalali-Heravi, M., R.S. Moazeni-Pourasil, and H. Sereshti. 2015. Elimination of chromatographic and mass spectrometric problems in GC–MS analysis of lavender essential oil by multivariate curve resolution techniques: Improving the peak purity assessment by variable size moving window-evolving factor analysis. J. Chromatogr. 983: 83-89.

Jensen, E. 2004. Fact or fancy. From the organic broad caster. Midwest organic and Sustainable. Edu. *12*(3): 164-170.

Linck, V.M., A.L. da Silva, M. Figueiró, A.L. Piato, A.P. Herrmann, F. Dupont Birck. 2009. Inhaled linalool-induced sedation in mice. Phytomed. 16(4): 303-307.

Machado, M.P., A.L.L. Silva and L.A. Biasi. 2011. Effect of plant growth regulators on *In vitro* regeneration of *Lavandula dentata* L. shoot tips. J. Biotec. Biodivers. 8(2): 28-31.

Marshner, H. 1995. Mineral nutrition of higher plants. Academic Press. London, UK.

McGimpsey, J. A., and N.G. Porter. 1999. Lavender. A grower's guide for commercial production. New Zealand Institute for Crop and Food Research Limited, Christchurch, New Zealand.

Medicinal Plants, A.C.E.C.R. 2012. Morphological traits of sweet basil (*Ocimum basilum L.*) as influenced by foliar application of methanol and nano-iron chelate fertilizers. Ann. Biol. Res. 3(12): 5511-5514.

Mehrab, Y. 2017. Effects of Zn, Fe, Mn and Cu foliar application on essential oils and morpho-physiological traits of lemon balm (*Melissa officinalis* L.). J. Essent. Oil-Bear. Plants. 20(2): 485-495.

Mirshekari, B. and R. Siyami. 2014. Positive response of essential oil percentage and its yield to seed pretreatment with some of major microelements. Case study: *Anethum graveolens* as a medicinal plant. Indian J. Fundam. App. Life Sci. 4(2): 453-461.

Nasiri, Y., S. Zehtab-Salmasi, S. Nasrullahzadeh, N. Najafi, K. Ghassemi-Golezani. 2010. Effects of foliar application of micronutrients (Fe and Zn) on flower yield and essential oil of chamomile (*Matricaria chamomilla L.*). J. Med. Plants Res. 4(17): 1733-1737.

Maher, H.S. 2019.GenStat for Analyzing Agricultural Experiments, AL-Qassim Green University- Agriculture College.

Nemati Lafmejani, Z., A.A. Jafari, P. Moradi, and A. Ladan Moghadam. 2018. Impact of foliar application of iron-chelate and iron nano particles on some morpho-physiological traits and essential oil composition of peppermint (*Mentha piperita L.*). J. Essent. Oil-Bear. Plants. 21(5): 1374-1384.

Nurzyńska-Wierdak, R. 2013. Does mineral fertilization modify essential oil content andchemical composition in medicinal plants. Acta Sci. Pol. Hortorum Cultus. 12(5): 3-16.

Rahimi A. A., K. Mashayekhi, K.H. Hemmati, A. Dardipure. 2008. The effect of nutrient elements and salicylic acid on seed yield and yield components of *Coriandrum sativum*. J. Herb. Prod. Res. 16(4): 149-156.

Reverchon, E., G.D. Porta, and F. Senatore. 1995. Supercritical CO_2 extraction and fractionation of lavender essential oil and waxes. J. Agric. Food Chem. 43(6): 1654-1658.

Rezaei M. B., M. Naderi-Kandi, S.R. Tabaei-Aghdaei. 2003. Mineral elements in different genotypes of *Rosa damascena* in Tehran, Azarbayjan-Sharghi and Golestan. J. Medicinal Aromat. Plants Res. 20(3): 291-299.

Roy R., K.M. Nasiruddin. 2011. Effect of different level of GA_3 on growth and yield of cabbage. J. Environ. Sci. Nat. Resour. 4(2): 79-82.

Sabara D., A. Kunicka-Styczynska. 2009. Lavender oil – flavouring or active cosmetic ingredient. *Scientific Bulletin of the Technical University of Lodz*, *78*, pp.33-41.

Sajjad, Y., M.J. Jaskani, M.Y., Ashraf, M. Qasim, and R. Ahmad. 2014. Response of morphological and physiological growth attributes to foliar application of plant growth regulators in gladiolus' with Prosperity'. Pak. J. Agric. Sci. 51(1).

Salamatbakhsh, M.R., A. Tobe, and E. Taherifard. 2012. Effects of foliar application of micronutrients on yield and yield components of castor bean (*Ricinus communis* L.) varieties. Eur. J. Exp. Biol. 2(4): 975-979.

Salarpour, O., S. Parsa, M.H. Sayyari, and M.J. Alahmadi. 2013. Effect of nano-iron chelates on growth, peroxidase enzyme activity and oil essence of cress (*Lepidium sativum L.*). Int. J. Agron. Plant Prod. 4(S): 3583-3589.

Samadiyan-Sarbangholi, V., B. Abbaszadeh, M.H. Lebaschy, and S.R. Tabaei-Aghdaei. 2013. Investigation the effect of iron chelate and NPK on yield of *Rosa damascena* for the first time in Iran. Int. J. For. Soil Eros. 3(3): 100-103.

Shan, B. Y., M. Cai, H.J. Sun. 2005. Corke, Agric. Food Chem. 53(20): 7749-7759.

Shekofteh, H., S. Shafie, Y. Mahmodi. 2013. A survey on the effects of manure and chemical fertilizers and their mixture on Ajowan's seed yield and its essential oil compositions. Int. J. Agric. 3, 401–408.

Smigielski, K., A.Raj, , K.Krosowiak, R., and Gruska. 2009. Chemical composition of the essential oil of *Lavandula angustifolia* cultivated in Poland. Journal of Essential Oil-Bearing Plants, 12(3):338-347.

Stierlin, É., T. Michel, and X. Fernandez. 2020. Field analyses of lavender volatile organic compounds: performance evaluation of a portable gas chromatography–mass spectrometry device. Phytochem. Anal. 31(6): 778-785.

Sure S., H. Arooie, and M. Azizi. 2012. Influence of plant growth regulators (PGRs) and planting method on growth and yield in oil pumpkin (*Cucurbita pepo* var. *styriaca*). Not. Sci. Biol. 4(2): 101-107.

Taiz, L. and E. Zeiger. 2010. Plant Physiology. Phytochem. Anal. 31(6): 778-785.

Tavallali, V., 2018. Effects of iron nano-complex and Fe-EDDHA on bioactive compounds and nutrient status of purslane plants. Int. Agrophys. 32(3): 411–419.

Tavallali, V., M. Kiani and S. Hojati. 2019. Iron nano-complexes and iron chelate improve biological activities of sweet basil (*Ocimum basilicum L.*). Plant Physiol. Biochem. 144: 445-454.

Touati, B., H. Chograni, I. Hassen, M. Boussa, L. Toumi, N.B. Brahima. 2011: Chemical composition of the leaf and flower essential oils of Tunisian *Lavandula dentata L.* (Lamiaceae). Chem. Biodiver. 8(8): 1560-1569.

Tucker, A.O. 1985. Lavender, spike, and lavandin. The Herbarist. 51: 44-50.

Wang, D., T. Yoshimura, K. Kubota, and A. Kobayashi. 2000. Analysis of glycosidically bound aroma precursors in tea leaves. 1. Qualitative and quantitative analyses of glycosides with aglycons as aroma compounds. J. Agric. Food Chem. 48(11): 5411-5418.

Yadegari, M. 2015. Foliar application of micronutrients on essential oils of borago, thyme and marigold. of Soil Science Plant Nutr, 15(4):1- 20.

Yousefzadeh, S., and N. Sabaghnia. 2016. Nano-iron fertilizer effects on some plant traits of dragonhead (*Dracocephalum moldavica L.*) under different sowing densities. Acta. Agric. Slov. 107(2): 429-437

Yuan L., and D.Q. Xu. 2001. Stimulation effect of gibberellic acid short-term treatment on the photosynthesis re-lated to the increase in Rubisco content in broad bean and soybean. Photosynth. Res. 68(1): 39-47.

Zagorcheva, T., S. Stanev, K. Rusanov, and I. Atanassov. 2013. Comparative GC/MS analysis of lavender (*Lavandula angustifolia* Mill.) inflorescence and essential oil volatiles. Agric. Sci. Technol. 5(4): 1313-8820.

Zehtab-Salmasi, S., F. Heidari, and H. Alyari. 2008. Effects of microelements and plant density on biomass and essential oil production of peppermint (*Mentha piperita* L.). Pl. Sci. Res. 1: 24-26.

Ziaeyan, A., M.J. Malakouti. 1998. Esffects of fertilizer containing micronutrients and timing on their increase Corn production. Iran. J. Soil Res. 12(1): 56-62.

Ziedan, E.H. and E.A. Eisa. 2016. The use of some micronutrients and plant extracts of resistance to powdery mildew and nutrition dill plants in the Gharbiyah Governorate. J. Plant Prot. and Path., Mansoura Univ. 7(9): 579-586.

CHAPTER IV

MYCORRHIZAE AND HUMIC ACID ON *LAVANDULA* ESSENTIAL OIL PRODUCTION

Abstract

The study investigated applications of humic acid and Mycorrhizae on growth and yield of essential oils in lavender. A factorial experiment with 4 rates of humic acid, 3 rates beneficial fungi (Mycorrhizae), and 3 species of lavender in a randomized complete block design with 5 replications was conducted for two successive growth seasons (2019/2020). The study was conducted in glass glazed greenhouses. Treatments included humic acid foliar spray at 0, 1.5, 2.5, or 3.5 g/L, Mycorrhizae fungi at 0, 10g *Trichoderma*, or 5g/L *Mycorrhiza* and 3 species of *Lavandula* (*L.* x *intermedia, L. angustifolia, L. stoechas*). Humic acid at 3.5 g/L resulted in the greatest rate of growth, plant height, number of branches, number of flowers, dry weight, and oil weight. All Mycorrhizae treatments indicated positive synergistic interactions with humic acid on the growth of lavender. The essential oil was extracted by Soxhlet. The composition of the essential oil was determined via GC-MS analyses. The characteristics of the essential oils of three species of lavender were comprehensively investigated by GC-MS. Seventy-seven volatile compounds were identified. The concentration of these compounds was affected by humic acid and mycorrhizal fungi. It was found the lavender plants treated with mycorrhizae had a high essential oils content and increased growth more than the non-mycorrhizae treated plants. The

use of mycorrhizal fungi may increase plant growth and increase the aromatic plants' production of essential oils.

Keywords: Humic acid, *Mycorrhiza, Trichoderma*, Three species of lavender, GC-MS

Introduction

The *Lamiaceae* plant family is represented by about 224 genera and 5600 species across the world. The most important medicinal and aromatic species for this family are mint, thyme, oregano, sage, rosemary, and lavender (Kan, et al., 2006). Oil of lavender is one of the most popular essential oils. It has a delightfully clear, refreshing and yet sweet odor which blends with many other essential oils. It is used as a carminative and spasmolytic in the form of a compound tincture. It is also used as a coloring and flavoring agent. Lavender is used largely in perfumery, toilet waters, soap manufacture and occasionally to cover disagreeable odors in ointments and other preparations. The oil contains chiefly linalool and linalyl acetate. It also contains pinene, limonene, geraniol and sesquiterpene (Guenther, 1961). Lavender essential oil is used in a wide variety of aromatherapy applications (Glenn, 2007). Moreover, lavender essential oil is used as an organic pesticide by mixing with rosemary essential oil (Anonymous, 2009). In addition to both industrial and traditional uses, lavender is valuable as an ornamental and landscaping plant (Kotsiris, et al., 2012; Matysiak and Nogowska, 2016).

In recent times, sustainable farming has become an important production practice in the world (El-Koni, 2002). Compost, with its content of humic and microbial substances, improves the physical, chemical, and microbiological conditions, and reduces the leaching of soil nutrients (Amin et al., 1999). Fertilization and mineral uptake/plant accumulation are among the most important factors in terms of plant growth and production (Marschner, 1995).

Lavender is a small shrub native to the mountains of the Mediterranean, the Arabian Peninsula, Russia, and Africa, and cultivated in southern Europe, the United Kingdom, the United States and Australia (Basch et al., 2004). Since lavender is a perennial plant, harvests can be made at least 15 years from the same plantation of lavender so, it is considered an economical crop. Lavender is produced and propagated in two main ways, sexually and vegetatively. Some types of lavender can be easily and quickly reproduced in both methods (Bajaj, 1988; Baydar, 2010a). Lavender is mostly planted in early spring.

An important aspect of humic acid is it is an organic acid without damaging effects to the environment (Ceylan, 1996). However, more environmentally friendly production should continue to be pursued. Many bio-fertilizers used mainly for plant growth have beneficial effects on the plant and this relates to the development and increased availability of nutrients for the host plants (Vessey, 2003). Environmentally friendly products have been widely used in agricultural applications. Increasing the safe production of food is a global concern and increasing yields is the most important agricultural economic goal for farmers. While fertilization with conventional micro and macro elements has a great effect on plant growth and productivity, it is costly and environmentally dangerous due to the pollution it causes to groundwater and the destruction of beneficial microorganisms (Hilman and Asandhi, 1987). Humic acid enhances plant growth and its physiological activities as well as stimulates microorganisms in the soil such as bacteria and fungi. Humic acid acts as a good chelate helping increase nutrient availability in the soil such as phosphates, calcium and trace elements as well as affecting the soil pH. Humic acids protect against negative changes that may occur due to the use of chemical fertilizers (Leonard, 2008). It is considered an alkali-soluble polymeric organic acid having an aromatic structure substituted

by carboxyl, phenol, hydroxyl, and alkyl groups bound together by ester bonds (Gaines and Yilmaz, 1983).

Essential oils are a mixture of chemical compounds, primarily monoterpenes and sesquiterpenes (Andrys and Kulpa, 2017; Sharopov, et al., 2015), which have antibacterial, antifungal and antioxidant properties (De Rapper, et al., 2016; Mahmoudi, et al., 2020). Essential oils synthesis in plants is likely to occur in different pathways. The formation of these compounds may link the formation of turbines with the conversion of carbohydrates while others link them to the transformation of proteins (Dubey et al., 2003(. They are analyzed by gas mass spectrometry (GC-MS) to assess the contribution of each plant to the mix of VOCs from harvested plants, to identify linalool (Landmann et al., 2007).

Fungal symbiosis plays an essential role in ecosystem sustainability (Bethlenfalvay and Schi.iepp, 1994; Giannenazi and Schiepp, 1994). The innate symbiosis has been shown to be an important determinant of plant diversity, ecosystem diversity and productivity (Van der Hejden et al., 1998). Mycorrhizae activity and humic acids enhance plant growth and therefore may enhance essential oils production. The aim of this study is to investigate the effect of humic acid and mycorrhizal fungi on plant growth and yield of essential oils in three species of lavender.

Materials and methods

Plant material and experimental design

The experiment was conducted twice from February to August 2019-2020. Lavender cuttings were obtained for three cultivars (Fig 4.1) *Lavandula* × *intermedia* 'Provence', *L. angustifolia* 'Hidcote' (Fig. 4.3), and *L. stoechas* 'Otto Quast' (Fig. 4.4) (Emerald Coast Growers, Inc., Pensacola, FL). They were planted in 15 cm (1 L) containers in a peat-based

substrate (Pro Mix BX, Premier Tech Horticulture, Quakertown, PA) when their height was 15 cm. A total of 180 plants were potted in the greenhouse between 2019-2020. No pesticides were used in the study area throughout the trial period. The experiment was carried out in a Randomized Complete Block Design (RCBD) with five replications. The following treatments were applied as a foliar spray: humic acid (Natural Guard, Ferti-lome Voluntary Purchasing Groups, Inc., Bonham, TX) at 0, 1.5, 2.5, or 3.5 g/L and mycorrhizae at 0 g, 10g *Trichoderma* (RootShield PLUS^{+}WP, BioWorks, Victor, NY) or 5g *Mycorrhiza* (Earth-Juice Rooter's Mycorrhizae, Earth Juice, Inc. Chico, CA). Plants were grown in Mississippi State University's Department of Plant and Soil Sciences greenhouses (latitude 27-33 ° N; longitude 88-47 ° W) with the temperature set points at 24/20° C Day/Night. Plants were fertilized twice weekly with 2-5-1 organic fertilizer (Drammatic "O", Dramm Corp., Manitowoc, WI).

L. intermedia ***L. stoechas*** ***L. angustifolia***

Figure 4.1 The three species of lavender used in the study.

Measurements of vegetative growth and essential oils

Plants were grown for 8 months, and the experiment repeated twice. When 50% of the plants were flowering, the plants and flowers were harvested at the soil line. Vegetative growth was recorded at the end of the growing season for all plants, with the following assessments made, Plant height (cm) was measured from the soil to the growing top, number of branches was counted as the number of main branches of each plant, number of florets on each plant at harvest, and shoot dry weight (g). Harvested plants were placed in 16x25x5 cm kraft paper food trays (Specialty Quality Packaging, Scotia, NY) and dried at room temperature (20°C). Samples were turned daily until dried to a continuous weight. After one month the plants were totally dry without any molding problems. Dried samples were weighed to determine the total plant dry weight (Dw-g) (buds, flowers, leaves, and stems) and stored in paper bags (Bettina and Helge, 2015).

Essential oils were extracted from the air-dried samples using a Soxhlet extraction apparatus (Reverchon and Della, 1995). Samples were prepared by placing 5 gm of dried, finely ground leaves and flowers (Cuisinart Spice and Nut Grinder MODEL SG-10, East Windsor, NJ) in a 33 x 80 mm paper thimble filter. 150 mL ethyl alcohol was placed in the boiling chamber and the samples were extracted over 9 hours. After the extraction, the samples were filtered from the plant tissues deposited at the bottom of the flask. Then the samples were transferred to the oven at 68°C to evaporate the ethanol from the extracted oil leaving the extracted oil (about 2 to 3 mL) in the glass flask. After that, the total weight of the oil was taken. Oils were kept in amber glass bottles at temperature room until GC-MS analysis using the method described in Chapter 3.

The chemical components of lavender oil were determined by GC-MS. Treatment samples were compared to pure standards for linalool and linalyl to compare their spectra. Quantitative data were based on the normalization of the peak area without using factor correction.

Statistical analysis

A Randomized Complete Block Design (RCBD) was used in a factorial experiment including three factors: humic acid, mycorrhizal fungi, and lavender species. SAS statistical software (SAS® ver. 9.4, SAS Institute, Cary, NC) was used for statistical analysis. The significant differences between means of experimental units among each treatment were compared using the least significant difference (LSD) at P α = 0.05. Data were subjected to analysis of variance (ANOVA) and separation of means by the LSD test at P $\leq$ 0.05. Normalized (average = 0, variance = 1) data were submitted to principal component analysis (PCA) with the aim of discriminating species based on the studied variable association.

Statistical analysis for GC-MS

The large number of treatments and replications which amounted to 360 samples for two experiments (2019, 2020), required much time and money to complete the essential oil analysis using GC-MS. In this study, duplicates of each treatment were collected, and one homogeneous sample was taken to represent that treatment in the statistical analysis process to estimate the active compounds. For estimating the significance of the statistical differences, the mean of the control parameters was used as a constant for the purpose of the assay using the T test of one sample at a probability level of 0.05. The data were analyzed using the Statistical Package for the Social Sciences (SPSS) version 26 (IBM, Inc, Armonk,

NY). Peak features were considered significant when comprising a signal-to-noise (S/N) ratio greater than 50 (and p-values < 0.05). All the data were analyzed with analysis of variance (ANOVA) using the SAS Statistical Package Program.

Results and Discussion

Effect of humic acid and mycorrhizae on plant growth

There was a significant effect of the experiment factors and their interaction on the plant height for the two experiment seasons (Table 4.1). *L. intermedia* grew taller than the other species in both experiments. Humic acid at 2.5 g/L resulted in taller plants than the control in the first experiment while 3.5 g/L humic acid produced taller plants in the second experiment. Mycorrhizae at a concentration of 5 g *Mycorrhiza* /L increased plant height in both experiments. The results of the two-way interaction between humic acid and species showed plant height was tallest in *intermedia* at 3.5 g/L. The shortest plant height was in *stoechas* at 0 g/L. The two-way interaction of humic acid and mycorrhizae resulted in the tallest plants at 3.5 g/L humic acid and 5 g *Mycorrhiza*/L. The interaction between species and mycorrhizae had *intermedia* with the tallest plants when 5 g *Mycorrhiza* /L was applied for both experiments. The shortest plants were in the treatment *stoechas* x 0 g mycorrhizae/L. There was a three-way interaction where 3.5 g humic acid/L × *intermedia* × 5 g *Mycorrhizae*/L produced the greatest plant height for the two experiments. The shortest plants were with 0 g humic acid/L × *stoechas* × 0 g mycorrhizae/L.

Table 4.1 Effect of humic acid and mycorrhizae on plant height (cm)in three species of lavender.

2019-2020						2020-2021			
Humic Acid (g L^{-1})	Lavender Species	Mycorrhizae (g L^{-1})			Humic Acid × Species	Mycorrhizae (g L^{-1})			Humic Acid × Species
		0g	10 g Trico	5 g Mycor.		0g	10 g Trico	5 g Mycor.	
0.0	*Intermedia*	39.20	40.40	44.00	41.2	34.00	35.00	40.00	36.3
	Angustifolia	31.00	33.80	36.80	33.9	33.00	36.00	39.67	36.2
	Stoechas	21.60	22.50	29.20	24.4	22.83	24.67	27.00	24.8
1.5	*Intermedia*	40.40	42.00	50.20	44.2	39.83	40.67	44.00	41.5
	Angustifolia	34.20	37.60	47.20	39.7	35.33	39.50	43.33	39.4
	Stoechas	22.50	23.00	29.80	25.1	23.33	25.33	28.67	25.8
2.5	*Intermedia*	45.60	48.20	53.10	49.0	40.67	45.17	49.67	45.2
	Angustifolia	36.60	39.60	43.50	39.9	42.33	44.67	47.67	44.9
	Stoechas	25.60	27.00	35.60	29.4	26.33	28.67	31.33	28.8
3.5	*Intermedia*	47.40	56.60	61.10	55.0	50.67	55.00	63.17	56.3
	Angustifolia	39.00	41.80	45.00	41.9	49.33	50.33	54.00	51.2
	Stoechas	28.80	34.60	38.80	34.1	30.00	33.00	34.67	32.6
LSD (0.05)			2.45		1.41		2.37		1.36
					Species Means				Species Means
Species × Mycorrhizae	*Intermedia*	43.15	46.8	52.10	47.35	41.29	43.96	49.21	44.82
	Angustifolia	35.2	38.2	43.12	38.84	40.00	42.62	46.17	42.93
	Stoechas	24.63	26.78	33.35	28.25	25.62	27.92	30.42	27.99
LSD (0.05)			1.22		0.70		1.18		0.68
					Humic Acid				Humic Acid
Humic Acid × Mycorrhizae	0.0	30.60	32.23	36.67	33.17	29.94	31.89	35.56	32.46
	1.5	32.37	34.20	42.40	36.32	32.83	35.17	38.67	35.56
	2.5	35.93	38.27	44.07	39.42	36.44	39.5	42.89	39.61
	3.5	38.40	44.33	48.30	34.68	43.33	46.11	50.61	46.69
LSD (0.05)			1.41		0.81		1.36		0.79
Mycorrhizae Means		34.33	37.26	42.86		35.64	38.17	41.93	
LSD (0.05)			0.70				0.68		

Mycorrhizae have been shown increase uptake of nutrients as well as stimulate plant production of phytohormones (Fitter, 1985). Humic acids make nutrients more accessible to the plant and promote plant growth and development (Amarowicz et al., 2008). Harper et al. (2000), found humic acid, when added to the soil or the plant, leads to an increase in the percentage of absorbed nutrients through the decomposition of organic matter, which in turn affects the acidic function of the soil (pH). These results are agreement with Badri et al. (2015) who reported humic acid improved plant height of lavender. Similar results were also reported in marjoram (*Majorana hortensis*) (El-Khateeb et al., 2017), calendula (*Calendula officinalis*) (Mohammadipour et al., 2012), dill (*Anethum graveolens*) (Said-Al Ahl, et al., 2016), niger (*Guizotia abyssinica*) (Tadayyon et al., 2017), chamomile (*Matricaria chamomilla*) (Mohammed et al., 2019), mint (*Mentha piperita* var. *citrate*) and basil (*Ocimum basilicum*) (Hendawy et al., 2015, Jamali et al., 2015 and Vafaei et al., 2015). Bayat and Belopukhov (2019) found plant height increased with humic acid and mycorrhizae in basil (*Ocimum basilicum L.*). Humic acid increases nutrient absorption, cell division, and photosynthesis. Furthermore, the use of humic acid on leaves and soil works to increase levels of auxin, cytokinin, and gibberellin in plants. Likewise, humic acid acts like a hormone-like substance whose auxin-like activity stimulates cell division and cell elongation (Schnitzer and Weightman, 1974).

Several mechanisms may explain the positive effects of mycorrhizae on plant growth, including enhanced protection from soil pathogens, hormone synthesis or mineralization of soil organic compounds (Mandyam and Jumpponen, 2005; Wu et al., 2010; Newsham, 2011). It has been highly acknowledged that mycorrhizal symbiosis improves the shoot biomass of the plant (Chaudhary et al., 2008; Kapoor et al., 2007; Mandal et al., 2013). Podila et al. (2000) reported mycorrhizae have importance due to its capability to increase plant growth and yield under stress

conditions. Inoculation with mycorrhizal fungi may increase plant growth in basil and mint (Aslani et al., 2014; Gupta et al., 2002). In a study conducted by Zubek et al (2012) mycorrhizae and soil microbial groups had a positive effect of lavender growth. Lavender plants are representative plant species in Mediterranean shrublands and belong to the natural succession in some semiarid Mediterranean ecosystems (Barea et al., 1992). They have been classified as ‘‘obligatory mycorrhizal’’ (Brundrett, 1991) or as ‘‘highly dependent on mycorrhiza’’ (Habte and Manjunath, 1991). More recent studies have confirmed the high mycorrhizal dependency of these plant species (Azcon and Barea, 1997). Mycorrhizal fungi resulted in improved lavender plant growth (Azcón and Barea,1997).

Number of branches/plants

There was a significant interaction of the trial factors on number of branches for the two experiments (Table 4.2). The species *stoechas* had the most branches followed by *angustifolia* and *intermedia*. Humic acid at 3.5 g/L increased number of branches compared to the control. Mycorrhizae at 5 g *Mycorrhiza*/L also increased the number of branches more than the control. The two-way interaction of humic acid and species indicated greatest branch numbers occurred in the treatment 3.5 g humic acid/L and *stoechas.* The least number of branches occurred in the 0 g humic acid/L and *intermedia* treatment. The two-way interaction between humic acid and mycorrhizae resulted in the greatest branch number in the 3.5 g humic acid/L and 5 g *Mycorrhiza*/L treatment. The lowest average branch number was in treatments with 0 g humic acid/L and 0 g mycorrhizae. In the species by mycorrhizae interaction, *stoechas* with 5 g *Mycorrhiza*/L had the greatest number of branches while *intermedia* with 0 g mycorrhizae/L had the least number of branches. There was a significant three-way interaction where 3.5 g humic acid/L × *stoechas* × 5 g *Mycorrhiza*/L produced the greatest number of branches. The least

number of branches was observed in the treatment 0 g humic acid/L × *intermedia* × 0 g mycorrhizae/L for the two experiments.

Table 4.2 Effect of humic acid levels and biofertilizer on number of branches/plants in three species of lavender.

2019-2020						2020-2021			
Humic Acid (g L^{-1})	Lavender Species	Mycorrhizae (g L^{-1})			Humic Acid × Species	Mycorrhizae (g L^{-1})			Humic Acid × Species
		0g	10 g Trico	5 g Mycor.		0g	10 g Trico	5 g Mycor.	
0.0	*Intermedia*	6.7	8.2	8.7	7.9	6.3	8.3	8.0	7.5
	Angustifolia	10.7	12.3	14.0	12.3	10.3	11.3	13.3	11.6
	Stoechas	15.3	16.7	19.3	17.1	13.7	15.5	17.3	15.5
1.5	*Intermedia*	13.7	14.0	15.3	14.3	12.5	13.3	14.2	13.3
	Angustifolia	15.0	17.7	21.0	17.9	14.5	15.7	19.0	16.4
	Stoechas	21.7	22.7	23.3	22.6	20.0	20.7	22.5	21.1
2.5	*Intermedia*	16.3	17.0	20.3	17.9	14.0	15.3	17.3	15.5
	Angustifolia	17.0	20.0	24.7	20.6	16.0	18.0	20.7	18.2
	Stoechas	22.0	23.0	23.2	22.7	20.7	22.3	22.7	21.9
3.5	*Intermedia*	19.7	20.3	23.0	21.0	16.7	18.7	21.0	18.8
	Angustifolia	18.7	22.0	26.0	22.2	25.0	26.0	28.0	26.3
	Stoechas	25.0	29.0	32.3	28.8	24.3	31.7	33.7	29.9
LSD (0.05)			1.93		1.11		1.52		0.88
					Species Means				Species Means
Species × Mycorrhizae	*Intermedia*	14.1	14.9	16.8	15.3	12.4	13.9	15.1	13.8
	Angustifolia	15.3	18	21.4	18.2	16.5	17.8	20.2	18.1
	Stoechas	21.0	22.8	24.6	22.8	19.7	22.5	24.1	22.1
LSD (0.05)			0.96		0.56		0.76		0.44
					Humic Acid				Humic Acid
Humic Acid × Mycorrhizae	0.0	10.9	12.4	14.0	12.4	10.1	11.7	12.9	11.6
	1.5	16.8	18.1	19.9	18.3	15.7	16.6	18.6	16.9
	2.5	18.4	20.0	22.7	20.4	16.9	18.6	20.2	18.6
	3.5	21.1	23.8	27.1	24.0	22.0	25.4	27.6	25.0

Table 4.3 (continued)

LSD (0.05)	**1.11**			**0.64**		**0.88**			**0.51**
Mycorrhizae Means	16.8	18.6	20.9			16.2	18.1	19.8	
LSD (0.05)	0.56					0.44			

Humic acid contains many elements that increase the availability of nutrients and thus affect plant growth (Akinci et al., 2009). The concentration of humic acid was reported to increase shoots in lavender (Badri et al., 2015). Similar results were found by Ahmed et al. (2011) on roselle, Mohammadipour et al. (2012) on marigold, Hendawy et al. (2015) on *Mentha piperita* var. *citrate*, El-Khateeb et al. (2017) on marjoram and Yousif (2018) on garlic. Muhammad et al. (2019) indicated the maximum number of branches of the stevia plant was when plants received the highest level of humic acid. Humic acid increases the porosity of soil and improves growth of the root system which leads to increases in the crop shoot system (Garcia et al., 2008). As humic acid increases soil porosity it also affects root development which in turn can induce shoot growth (Katalinić et al., 2009). Khanam et al. (2012) noted that humic substance has a remarkable impact on physical and chemical properties of the soil and consequently ameliorates nutrient mineral uptake. Humic acid contains many elements which improve the soil fertility and increases the availability of nutrient elements by holding them on mineral surfaces, consequently, affecting plant growth and yield (El-Sharkawy and Abdel-Razzak, 2010).

Mycorrhizae symbiosis modifies root functions and microbial equilibrium in the rhizosphere (Leyval and Berthelin, 1993). Mycorrhizal fungi are usually generalists with the plant hosts they colonize to be functionally equivalent in their effects on a host and on the soil microflora (Hart et al., 2003). However, it has been recently demonstrated that mycorrhizal

fungal growth and species is host specific when different plant species and fungi are grown together (Van der Hejden et al., 1998). Ouahmane et al. (2006) reported native mycorrhizal fungi seemed to have a better capacity to impact lavender seedling growth. They confirmed the role of mycorrhizal fungi as a major factor contributing to the growth and co-existence of each of the lavender species. Gianinazzi-Pearson (1984) and Brundrett (1991) found mycorrhizal dependency is a botanical characteristic. Growth improved due to mycorrhizal performance, such as relative contribution from the root compared to the nutrient mediated by fungi absorption (Gianinazzi-Pearson, 1984; Brundrett, 1991).

Number of flowers per plant

The species *angustifolia* produced the greatest number of florets followed by *intermedia* and *stoechas* (Table 4.3). Treating with humic acid at 3.5 g/L resulted in a greater number of florets compared to the 0 g humic acid/L control. Foliar applications of mycorrhizae at 5 g *Mycorrhiza*/L resulted in a greater number of florets than treatments without mycorrhizae. The two-way interaction between humic acid and the species produced the greatest florets numbers when *angustifolia* was treated with 3.5 g humic acid/L. Plants of *stoechas* did not bloom when treated with any concentration of humic acid in the second experiment. The two-way interaction of humic acid and mycorrhizae resulted in the greatest number of florets when plants received 3.5 g humic acid/L and 5 g *Mycorrhiza*/L. The least number of florets occurred when plants received no humic acid and no mycorrhizae. The species *L. angustifolia* produced the most florets when also treated with 5 g *Mycorrhizae*/L while *stoechas* treated with 0 g mycorrhizae/L had the fewest florets. There was a significant three-way where the greatest number of florets were produced in the treatment 3.5 g humic acid/L × *angustifolia* × 5 g *Mycorrhizae*/L for the two experiments. The species *stoechas* did not bloom with any of treatments.

Table 4.3 Effect of humic acid and mycorrhizae on number of flowers per plant in three species of lavender.

2019-2020						2020-2021			
Humic Acid (g L^{-1})	Lavender Species	Mycorrhizae (g L^{-1})			Humic Acid × Species	Mycorrhizae (g L^{-1})			Humic Acid × Species
		0g	10 g Trico	5 g Mycor.		0g	10 g Trico	5 g Mycor.	
0.0	*Intermedia*	64.7	105.3	118.3	96.1	53.7	89.9	99.3	81.0
	Angustifolia	121.0	210.0	227.0	186.0	113.7	143.7	162.0	139.8
	Stoechas	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
1.5	*Intermedia*	79.0	110.3	128.7	106.0	64.7	94.7	112.7	90.7
	Angustifolia	128.0	161.7	184.3	158.0	116.3	136.7	154.7	135.9
	Stoechas	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
2.5	*Intermedia*	124.3	134.3	156.0	138.2	119.0	129.3	143.3	130.5
	Angustifolia	188.7	203.3	225.0	205.7	164.3	184.7	195.0	181.3
	Stoechas	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
3.5	*Intermedia*	141.0	157.7	169.0	155.9	125.7	140.3	156.0	140.7
	Angustifolia	195.3	233.0	251.7	226.7	174.7	193.7	203.0	190.5
	Stoechas	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
LSD (0.05)		6.52			3.77	6.75			3.90
					Species Means				Species Means
Species × Mycorrhizae	*Intermedia*	102.3	126.9	143.0	124.0	90.8	113.6	127.8	110.7
	Angustifolia	158.2	202.0	222.0	194.0	142.3	164.7	178.7	161.9
	Stoechas	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
LSD (0.05)		3.26			1.88	3.38			1.95
					Humic Acid				Humic Acid
Humic Acid × Mycorrhizae	0.0	61.9	105.1	115.1	94.0	55.8	77.9	87.1	73.6
	1.5	69.0	90.7	104.3	88.0	60.3	77.1	89.1	75.5
	2.5	104.3	112.6	127.0	114.6	94.4	104.7	112.8	104.0
	3.5	112.1	130.2	140.2	127.5	100.1	111.3	119.7	110.4
LSD (0.05)		3.77			2.17	3.90			2.25
Mycorrhizae Means		86.8	109.6	121.7		77.7	92.7	102.2	
LSD (0.05)		1.88				1.95			

Singh and Wasnik (2013) and Jalayerinia et al. (2017) found foliar humic acid in well fertilized rosemary plants produced the greatest number of flowers. Similar results were observed in gerbera where higher rates of humic acid increased the number of harvested flowers (Nikbakht et al., 2008). Similar results were also reported on chamomile (*Matricaria chamimilla*) where humic acid levels increased the number of air-dried flower heads/ plant (g) and per acre (Mohammed et al., 2019).

Humic acids have a reducing capacity with respect to Fe uptake (Sanchez-Sanchez et al., 2002). Humic acid also serves as a catalyst in promoting the activity of microorganisms in the soil (Chen and Aviad, 1990; Sharif et al., 2002). Bohme and Thi Lua (1997) reported on humic acid's beneficial effects on plant nutrient uptake, and micronutrient transport and availability. Hypotheses explaining the stimulatory effects of humic acid are numerous, the most compelling of which is the "direct action" on the plant, which is hormonal in nature, along with the "indirect action" through positive effects on seed germination, seedling growth, root growth, and bud development. Zhang and Ervin (2004) showed humic acid has cytokinin activity. Other hypothetical mechanisms include enhanced uptake of metabolic ions and increases in cell permeability (Chen and Aviad, 1990). Varanini and Pinton (1995) reported that humic acid enhanced absorption of micro- and macro-elements in plants. Humic acid can enhance plant growth by promoting the bioavailability of nutrients (Chen et al. 2004). Consequently, the use of humic substances has often been proposed as a method to improve crop production. Boehme et al. (2005) reported humic acid influences the respiration process, the amount of sugars, amino acids, and nitrate accumulated.

Lavender plants are representative botanical species in Mediterranean shrubs (Barea al., 1992).

It is rated as "compulsory mycorrhizal" (Brundrett, 1991) or as " highly dependent on mycorrhizal fungi" (Habte and Manjunath, 1991). Mycorrhizal fungi can solubilize surrounding weatherable minerals through excretion of organic acids (Landeweert et al., 2001). The results in this study emphasize there is a role for mycorrhizae as a major factor contributing to growth of species of lavender. Golubkina et al. (2020) explained that mycorrhizae stimulated the growth of tarragon and lavender plants. Previous studies show the positive effects of mycorrhiza on plant growth (Ortas and Akpınar, 2006; Miransari et al., 2009; Wu and Zou, 2010). Wu and Zou (2010) indicated the beneficial effects of mycorrhizae could contribute to high chlorophyll and therefore high photosynthetic activity. Therefore, knowledge of the relationship between plants and fungi is of considerable importance for the successful use of mycorrhizae fungi (Tian et al., 2004).

Plant dry weight (g)

The species *stoechas* had the greatest plant dry weight followed by *angustifolia* and *intermedia* (Table 4.4). Humic acid applied at 3.5 g/L led to a significant increase in the plant dry weight over the control. The use of mycorrhizae at 5g *Mycorrhiza*/L also increased dry weight above the *Tricoderma* and the control treatments.

The humic acid/species two-way interaction between 3.5 g humic acid/L and *stoechas* resulted in the greatest dry weight while the least dry weight was in the 0g humic acid/L and *intermedia*. Humic acid also interacted with mycorrhizae where 3.5 g humic acid/L and 5 g *Mycorrhiza*/L resulted in the greatest plant dry weight. The control of 0 g humic acid/L and 0 g mycorrhizae had the least plant dry weight. Species and mycorrhizae interacted whereby

stoechas and 5 g *Mycorrhiza*/L had the greatest plant dry weight. The species *intermedia* receiving 0 g mycorrhizae/L had the least plant dry weight.

The three-way interaction of 3.5 g humic acid/L x *stoechas* x 5 g *Mycorrhiza* /L produced the greatest plant dry weight. The least plant dry weight was observed in the treatment 0 g humic acid/L x *intermedia* x 0 g mycorrhizae/L.

Table 4.4 Effect of humic acid and mycorrhizae on plant dry weight (g) in three species of lavender.

2019-2020						**2020-2021**			
Humic Acid (g L^{-1})	Lavender Species	Mycorrhizae (g L^{-1})			Humic Acid × Species	Mycorrhizae (g L^{-1})			Humic Acid × Species
		0g	10 g Trico	5 g Mycor		0g	10 g Trico	5 g Mycor	
0.0	Intermedia	7.6	9.3	9.6	8.8	6.5	7.9	7.9	7.4
	Angustifolia	10.2	11.2	11.5	11.0	9.3	10.1	10.8	10.1
	Stoechas	12.3	12.8	13	12.7	11.2	11.7	12	11.6
1.5	Intermedia	8.3	9.1	10.6	9.3	7.5	8.2	9.1	8.3
	Angustifolia	11.2	11.9	12	11.7	10	10.5	11.6	10.7
	Stoechas	12.6	12.8	13.1	12.8	11.9	12.3	12.3	12.2
2.5	Intermedia	11.2	11.4	11.7	11.4	10.6	10.7	10.9	10.7
	Angustifolia	11.6	11.9	12.0	11.8	11.2	11.5	11.8	11.5
	Stoechas	12.5	12.8	12.9	12.7	12	12.2	12.4	12.2
3.5	Intermedia	11.3	11.6	11.8	11.6	10.9	11	11.2	11.0
	Angustifolia	11.8	12.0	12.3	12.0	11.4	11.7	11.9	11.7
	Stoechas	12.7	12.8	12.9	12.8	12.1	12.3	12.4	12.3
LSD (0.05)			0.54		0.31		0.33		0.19
					Species Means				Species Means
Species × Mycorrhizae	Intermediate	9.6	10.4	10.9	10.3	8.9	9.5	9.8	9.4
	Angustifolia	11.2	11.8	11.9	11.6	10.5	11	11.5	11.0
	Stoechas	12.5	12.8	13.0	11.9	11.8	12.1	12.3	12.1
LSD (0.05)			0.27		0.16		0.17		0.10
					Humic Acid				Humic Acid
Humic Acid ×	0.0	10.0	11.1	11.3	10.8	9	9.9	10.3	9.7
	1.5	10.7	11.3	11.9	11.3	9.8	10.3	11.1	10.4
	2.5	11.8	12.0	12.2	12.0	11.3	11.5	11.7	11.5

Mycorrhizae	3.5	11.9	12.1	12.3	12.1		11.5	11.7	11.8	11.7

Table 4.5 (continued)

LSD (0.05)	**0.31**			**0.18**		**0.19**			**0.11**
Mycorrhizae Means	11.1	11.6	11.9			10.4	10.8	11.2	
LSD (0.05)	0.16					0.10			

Humic acid improves the physical and chemical properties of the soil increasing the absorption and transfer of elements from the source to the plant (El-Hak et al., 2012). This results in increasing chlorophyll content in the leaves thus increasing the efficiency of photosynthesis and increasing its products carbohydrates (Abdul-Al- Hayani, 2016). The increase in chlorophyll and the subsequent increase in the products of photosynthesis result in an increase in the proportion of synthesized carbohydrates (Al-Ali and Majid, 2013). Badri et al. (2015) reported the consumption of humic acid increased the dry weight of the plant. They used 2.5 g humic acid under normal conditions which had the greatest effect on lavender to increase the yield and quality. Humic acid application at 2 to 5 g/L increased shoot dry weight of *Mentha piperita* var. *citrate,* basil plants, and *Calendula officinalis* (Hendawy et al. 2015; Jamali et al., 2015; Mohammadipour et al., 2012; Vafaei et al., 2015). Furthermore, this result is in harmony with the results of Moraditochaee (2012) and Bakry et al. (2013) who showed significant increases in peanut and wheat grain yield due to the addition of humic acid. Likewise, Dawood et al. (2019) indicated humic acid application at 5% positively affected dry weight in fava bean.

Plant growth promoting mycorrhizae increased dry weight in basil and chamomile (*Matricaria chamomilla*) (Mohammed et al., 2019). Many studies support the beneficial effects of mycorrhizae such as Ameri and Tehranifar (2012) on strawberry (*Fragaria ananasa)*, and Boghdady et al. (2016) on chickpea seedlings. According to Gupta et al. (2002), *Mentha arvensis*

plants inoculated with Mycorrhizae resulted in increased dry weight compared to non-fungal plants. Mycorrhizae inoculation significantly increased dry weight of flaxseed (*Linum usitatissimum*) (Ansari et al., 2016). Inoculation of basil with mycorrhizae not only enhanced growth but improved shoot dry weight, percent of essential oil and composition of essential oils (Aslani et al., 2014). Martinez et al. (2011) explained it is widely accepted mycorrhizal symbiosis creates hormonal changes in the roots of the host plant; however, there is only limited evidence of the system effects of plant-mycorrhizal fungi interactions. Gutjahr (2014) stated mycorrhizae have a broad host range and thus are the most credible group to enhance secondary production metabolites in plants. Swaminathan and Verma (1979) found the greatest benefit of mycorrhizae to the host plant is increased absorption of mineral nutrients, especially phosphorus. Karagiannidis et al. (2011) reported mycorrhiza-treated oregano plants produced double the dry weight compared to the control plants. Similarly, Gupta et al. (2002) reported that treatment of *Mentha arvensis* with mycorrhiza increased fresh and dry biomass. According to Karagiannidis et al. (2012) *Glomus lamellosum* inoculation to *L. angustifolia* resulted in increased plant biomass.

Oil yield /plant

The species *stoechas* had the highest average essential oil yield followed by *angustifolia* and *intermedia* (Table 4.5). Humic acid at 3.5 g/L was beneficial in increasing the oil yield over the control. Mycorrhizae at 5 g *Mycorrhiza*/L increased oil yield over use of *Tricoderma* or no mycorrhizae. The two-way interaction between humic acid and the species indicated 3.5 g humic acid/ L on *stoechas* increased oil yield while 0g humic acid/L on *intermedia* resulted in the least oil yield. The two-way treatment between humic acid and mycorrhizae showed 3.5 g humic acid/L with 5 g *Mycorrhiza*/L recorded the highest oil yield.

The lowest oil yield was from the treatment with 0 g humic acid and 0 g mycorrhizae. In looking at the interaction of species and *mycorrhiza stoechas* with 5 g *Mycorrhiza*/L had the greatest oil yield while *intermedia* with 0 g Mycorrhizae/L produced the least oil. The three-way interaction humic acid, species, and Mycorrhizae was significant. The treatment 3.5 g humic acid/L x *stoechas* x 5 g *Mycorrhiza*/L produced the greatest oil yield. The least oil yield was in the treatment 0 g humic acid/L x *intermedia* x 0 g mycorrhizae/L.

Table 4.5 Effect of humic acid levels and mycorrhizae on oil yield in three species of lavender.

2019-2020							**2020-2021**			
Humic Acid (g L^{-1})	Lavender Species	Mycorrhizae (g L^{-1})			Humic Acid × Species		Mycorrhizae (g L^{-1})			Humic Acid × Species
		0g	10 g Trico	5 g Mycor.			0g	10 g Trico	5 g Mycor.	
0.0	*Intermedia*	18.77	20.03	20.73	19.84		20.1	21.8	23.4	21.8
	Angustifolia	21.40	21.73	21.90	25.19		22.6	23.5	25.4	23.8
	Stoechas	22.17	23.33	24.27	23.87		20.2	22.3	23.6	22.0
1.5	*Intermedia*	23.93	25.43	26.20	24.43		22.3	24.4	26.3	24.3
	Angustifolia	23.73	26.43	28.10	21.68		25.5	26.8	28.4	26.9
	Stoechas	24.20	26.20	27.77	26.09		22.4	23.6	25.7	23.9
2.5	*Intermedia*	23.20	23.67	24.73	25.17		24.3	26.6	27.6	26.2
	Angustifolia	23.07	25.83	26.60	26.16		25.5	26.6	27.7	26.6
	Stoechas	24.03	25.70	26.20	23.26		23.3	26	27.8	25.7
3.5	*Intermedia*	22.00	24.60	26.70	26.06		23.7	27.5	31.3	27.5
	Angustifolia	24.60	26.53	27.33	25.31		24.6	28.5	29.4	27.5
	Stoechas	24.57	26.60	31.00	27.39		25.5	26.5	31.8	27.9
LSD (0.05)			0.96		0.55			0.63		0.36
					Species Means					Species Means
Species × Mycorrhizae	*Intermedia*	21.98	23.43	24.59	23.33		22.6	25.1	27.2	24.9
	Angustifolia	23.20	25.13	25.98	24.68		24.6	26.3	27.7	26.2
	Stoechas	23.74	25.46	27.31	25.96		22.9	24.6	27.2	24.9
LSD (0.05)			0.48		0.27			0.32		0.18
					Humic Acid					Humic Acid

Table 4.5 (continued)

Humic Acid × Mycorrhizae	**0.0**	**20.78**	**21.70**	**22.30**	**21.59**		**21**	**22.5**	**24.1**	**22.5**
	1.5	23.96	26.02	27.36	25.78		23.4	24.9	26.8	25.1
	2.5	23.43	25.07	25.84	24.78		24.4	26.4	27.7	26.2
	3.5	23.72	25.91	28.34	25.99		24.6	27.5	30.9	27.7
LSD (0.05)			0.55		0.32			0.63		0.21
Mycorrhizae Means		22.97	24.68	25.96			23.3	25.3	27.4	
LSD (0.05)			0.27					0.18		

Badri et al. (2015) indicated humic acid increased the conditions for improvement in linalool and geraniol oil production. The use of 2.5 g humic acid under normal conditions had the greatest effect on herbs and lavender essential oils increasing the yield and quality (Hendawy et al., 2015). Noroozisharaf and Kaviani (2018) indicated humic acid positively changed biological activities increasing essential oil content and its major constituents in *Thymus vulgaris* under greenhouse conditions. Juarez et al. (2011) indicated essential oil percentage and yield were higher at the highest humic acid levels compared to control. Humic acid application at 2 to 5 g/l increased essential oil of *Mentha piperita* var. *citrate* and basil plants (Hendawy et al. 2015, Jamali et al. 2015 and Vafaei et al. 2015). Other investigators found spraying dill plants with humic acids increased oil yield (Said-Al Ahl, et al., 2016) and its use on *Guizotia abyssinica* increased plant growth, oil, and protein content (Tadayyon et al., 2017). Humic acids stimulate biological processes in the plant, especially photosynthesis, which is the biocentre of the plant to produce secondary compounds such as glycosides (Zahra et al., 1984; Bowes et al., 2004). Sardashti et al (2012), reported the percentage chemical composition of the essential oil from *A.*

sieberi increased when its root was exposed to humic acid. Essential oil increased as the humic acid level was increased (Bayat and Belopukhov, 2019).

Rhizobacteria have been shown to increase essential oils in basil and chamomile (Mohammed et al, 2019). Karagiannidis et al. (2011) found that oregano and mint treated with mycorrhizal fungi were higher in essential oils and nutrients than non-treated plants. In addition, the essential oil composition of mycorrhizae treated plants differed from that of non-treated plants. Inoculation of basil not only enhanced growth but also improved the percent of essential oil and composition of essential oil (Aslani et al., 2014). Karagiannidis et al. (2011) found mycorrhizal isolates led to a significant increase in plant growth and total production of essential oil in both thyme and mint plants. The results of this study indicate it may be possible to use mycorrhizal fungi to influence essential oil quality and production. Mycorrhizal fungi improve plant development, nutrition, and essential oil content (Nell et al., 2010). There are many reports on improved nutrient uptake, especially of phosphorus, by mycorrhizal plants compared to non-mycorrhizal plants (Cabello et al., 2005; Clark et al., 1999; Kanias et al., 1998; Raˇzi ´c et al., 2005).

According to Gupta et al. (2002) application of mycorrhiza to *Mentha arvensis* increased the production of essential oils. Kapoor et al. (2004) indicated that inoculation with mycorrhizal fungi increased the total essential oil concentration of *Artemisia annua and Lactuca sativum*, especially geraniol and linalool. Khaosaad et al. (2006) and Copetta et al. (2006) reported the qualitative and quantitative improvement of essential oil production elicits a high commercial interest.

Discussion results of GC-MS (Essential oil composition)

Essential oil composition

Lavender oil obtained from three species of lavender treated with humic acid and mycorrhizae was analyzed by GC-MS (Fig 4.2-4.16). The main ingredients of lavender essential oil are cis- linalool, limonene, linalyl acetate, γ -terpinene, and cis-sabinene hydrate sabinene hydrate, sabinene, γ-terpineol, and terpinen-4-ol. The plants treated with humic acid at a high level (3.5 gm) as well as *Mycorrhiza* at 5 g/L were found to be rich in cis-sabinene hydrate, while terpinen-4-ol was found to be abundant in the essential oil of plants treated with at 3.5 g humic acid/L or mycorrhizae. Humic acid at all tested levels increased the proportion of linalool in the oil compared to the control. Sabinene and γ-terpineol decreased with all mycorrhizae treatments compared to the control group. Novak et al (2002) showed the formation of cis-sabinene and acetate hydrates appears to be due to a specific enzymatic mutation in oregano plants.

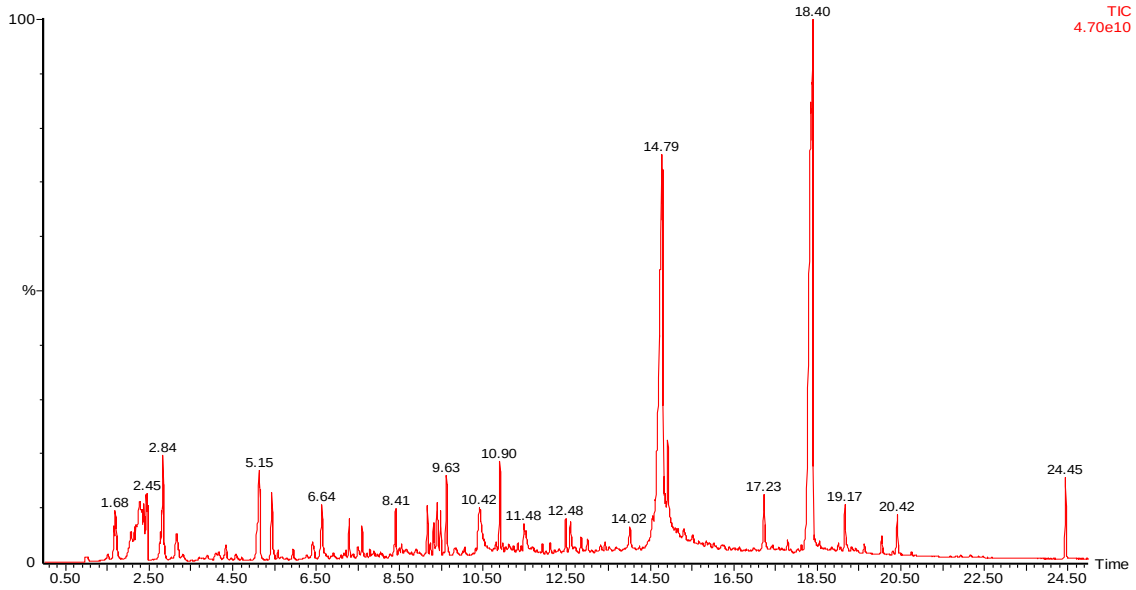

Figure 4.2 Chromatograph of essential oil extracted from *L. angustifulia* treated with 3.5g/l humic acid and 5g *Mycorrhiza*.

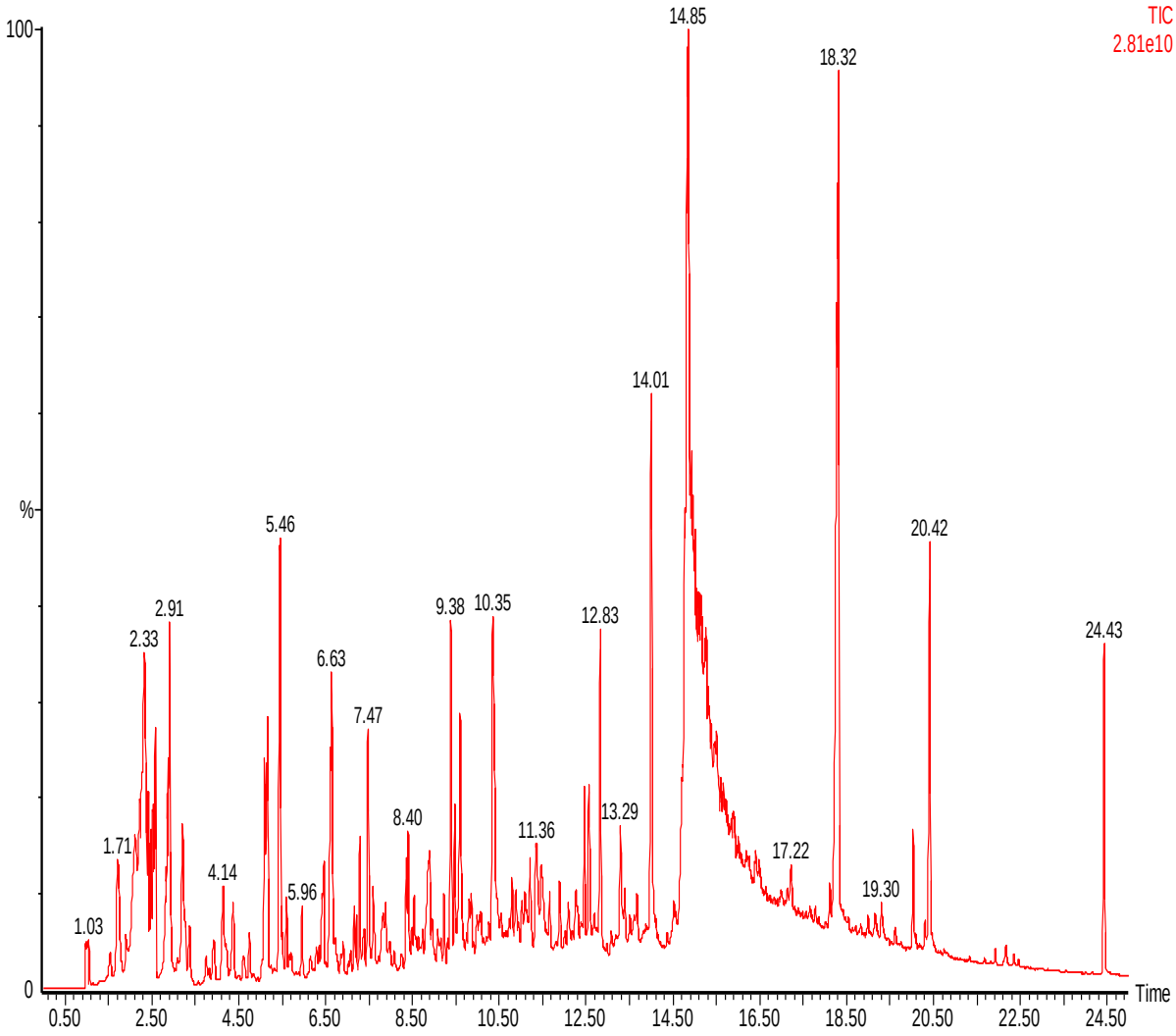

Figure 4.3 Chromatograph of essential oil extracted from *L. stoechas* treated with 3.5g/L humic acid and 10g *Trichoderma*.

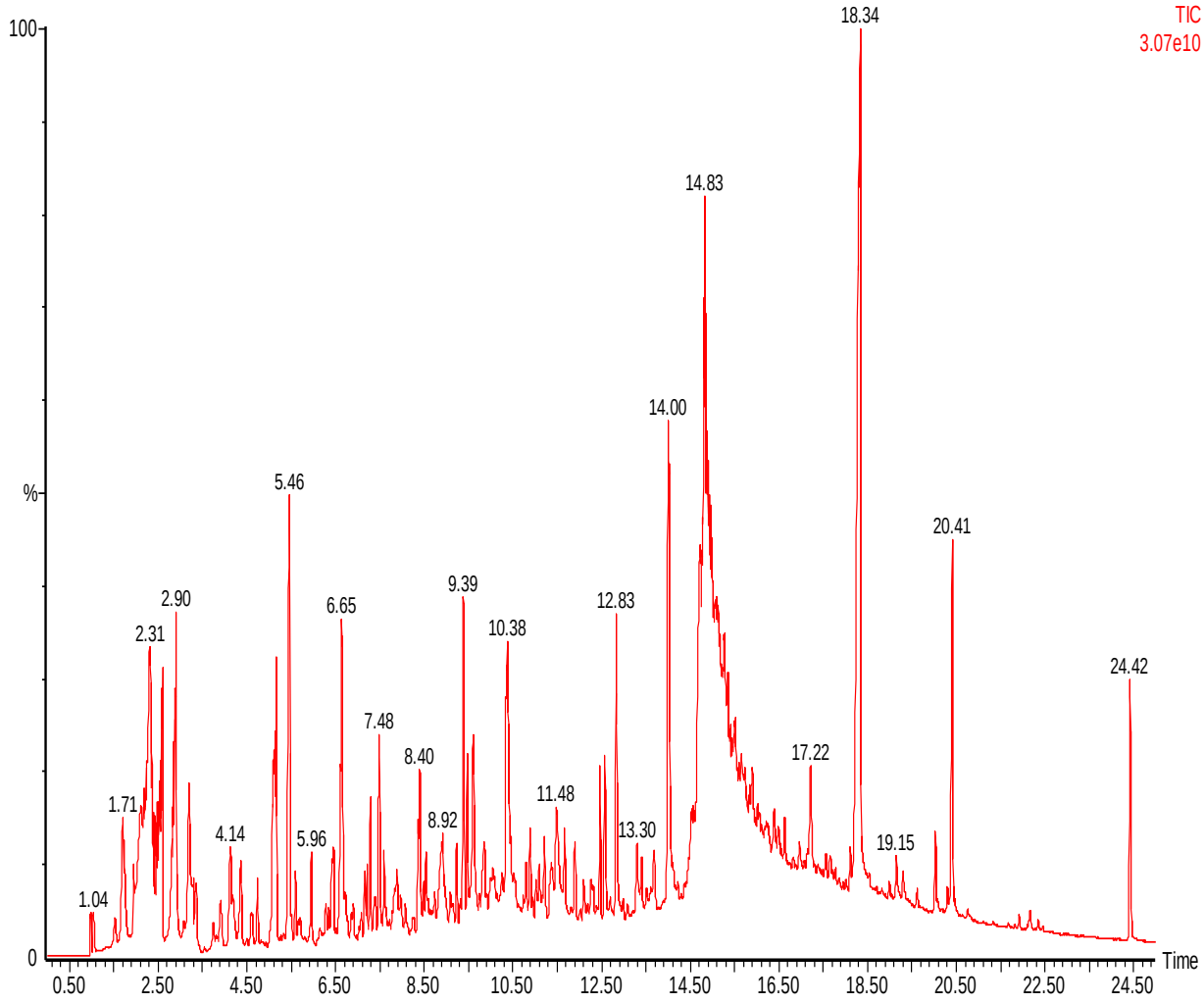

Figure 4.4 Chromatograph of essential oil extracted from *L. stoechas* treated with 2.5 g /L humic acid and 5g/L *Mycorrhiza*.

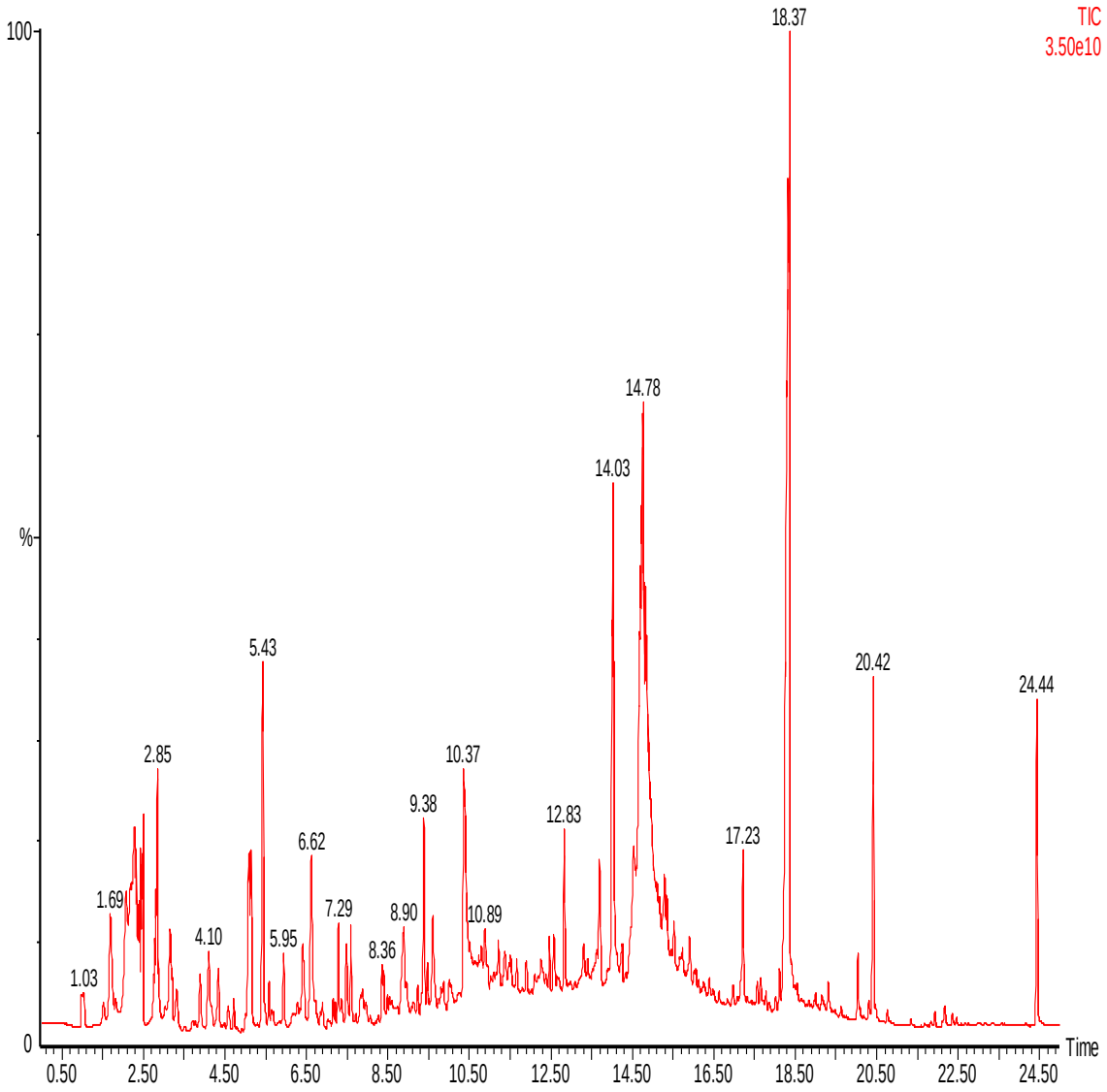

Figure 4.5 Chromatograph of essential oil extracted from *L. stoechas* treated with 3.5 g /L humic acid and 5g/L *Mycorrhiza*.

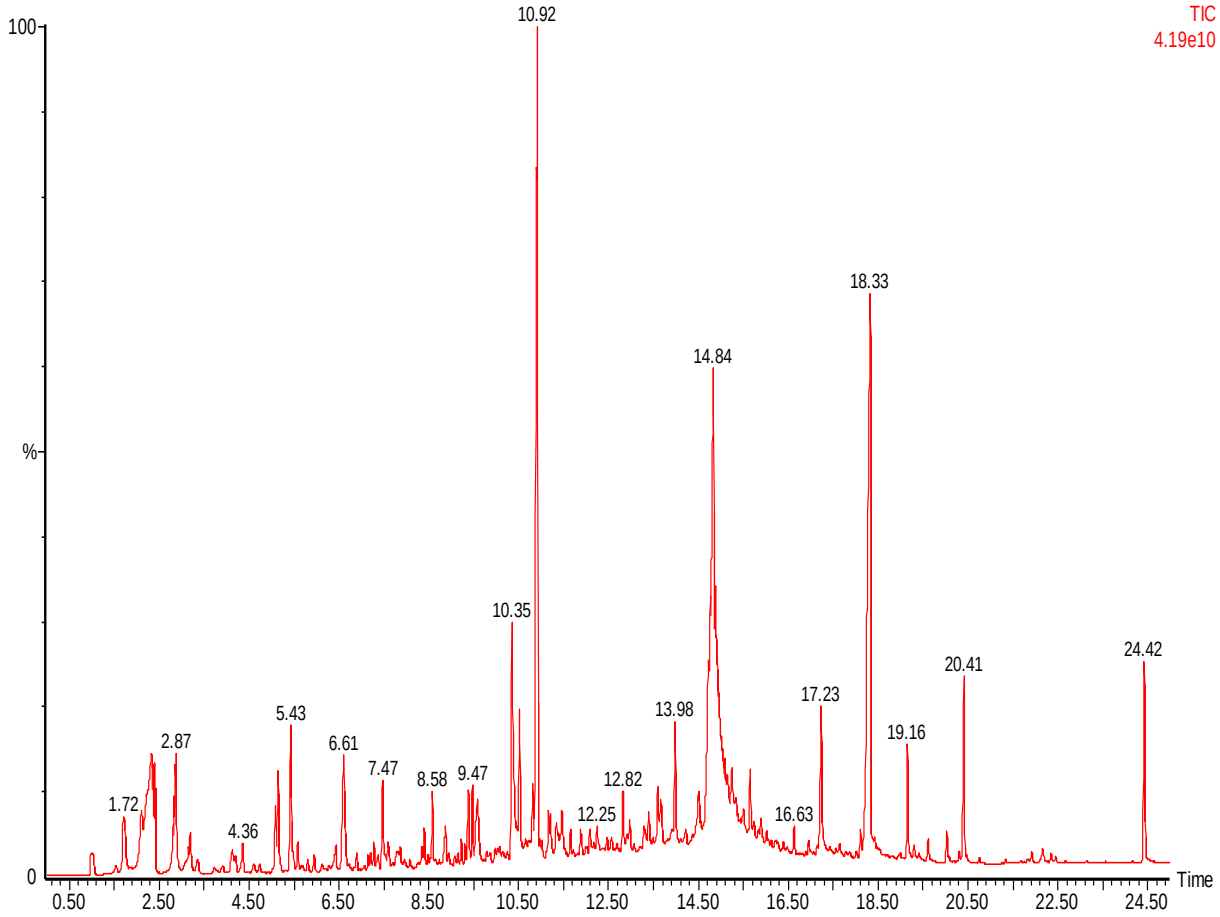

Figure 4.6 Chromatograph of essential oil extracted from *L. stoechas* treated with 0g/L humic acid, 0g/L *Mycorrhiza*.

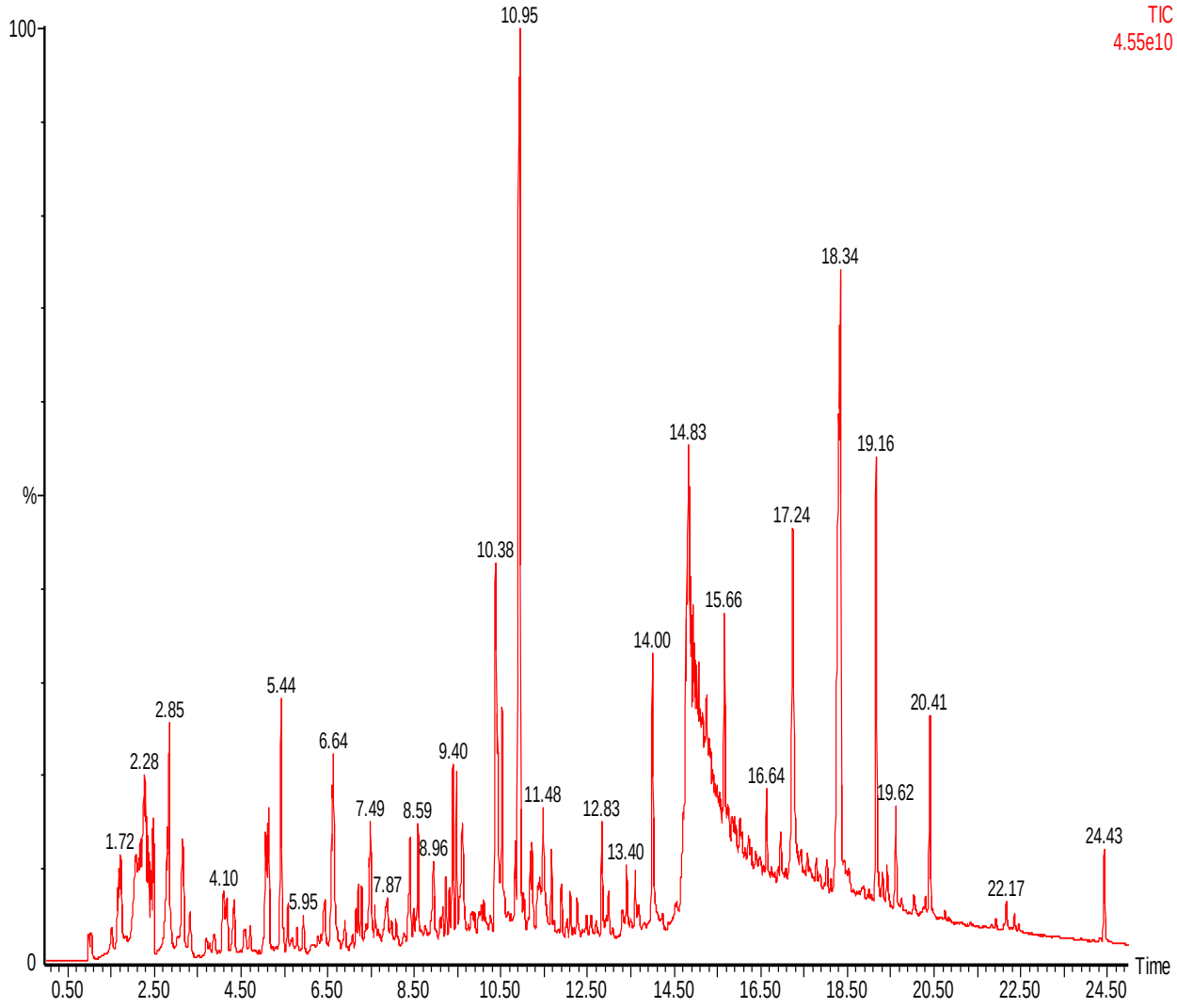

Figure 4.7 Chromatograph of essential oil extracted from *L. intermedia* treated with 1.5g/L humic acid and 5g/L *Mycorrhiza* .

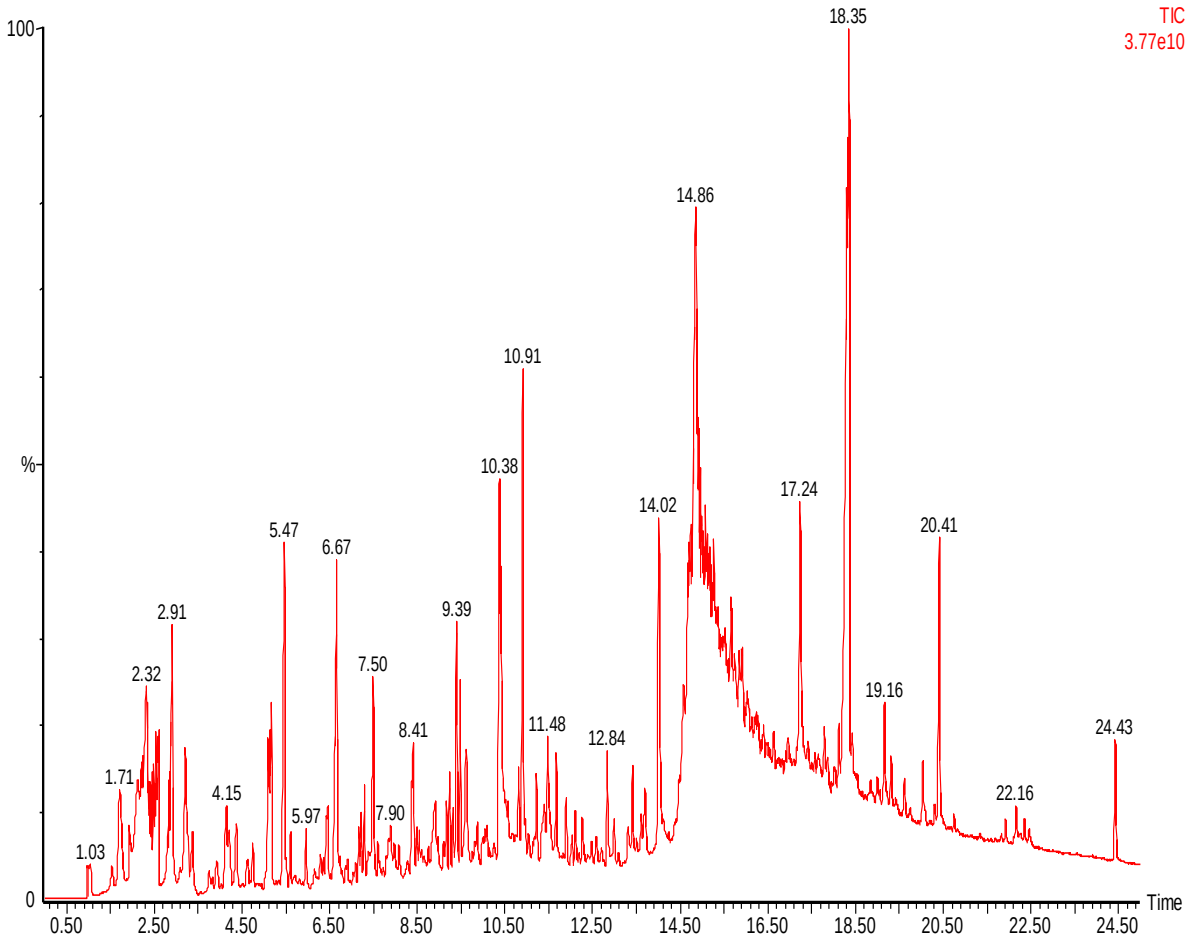

Figure 4.8 Chromatograph of essential oil extracted from *L. intermedia* treated with 0g/L humic acid and 0g/L *Mycorrhiza* .

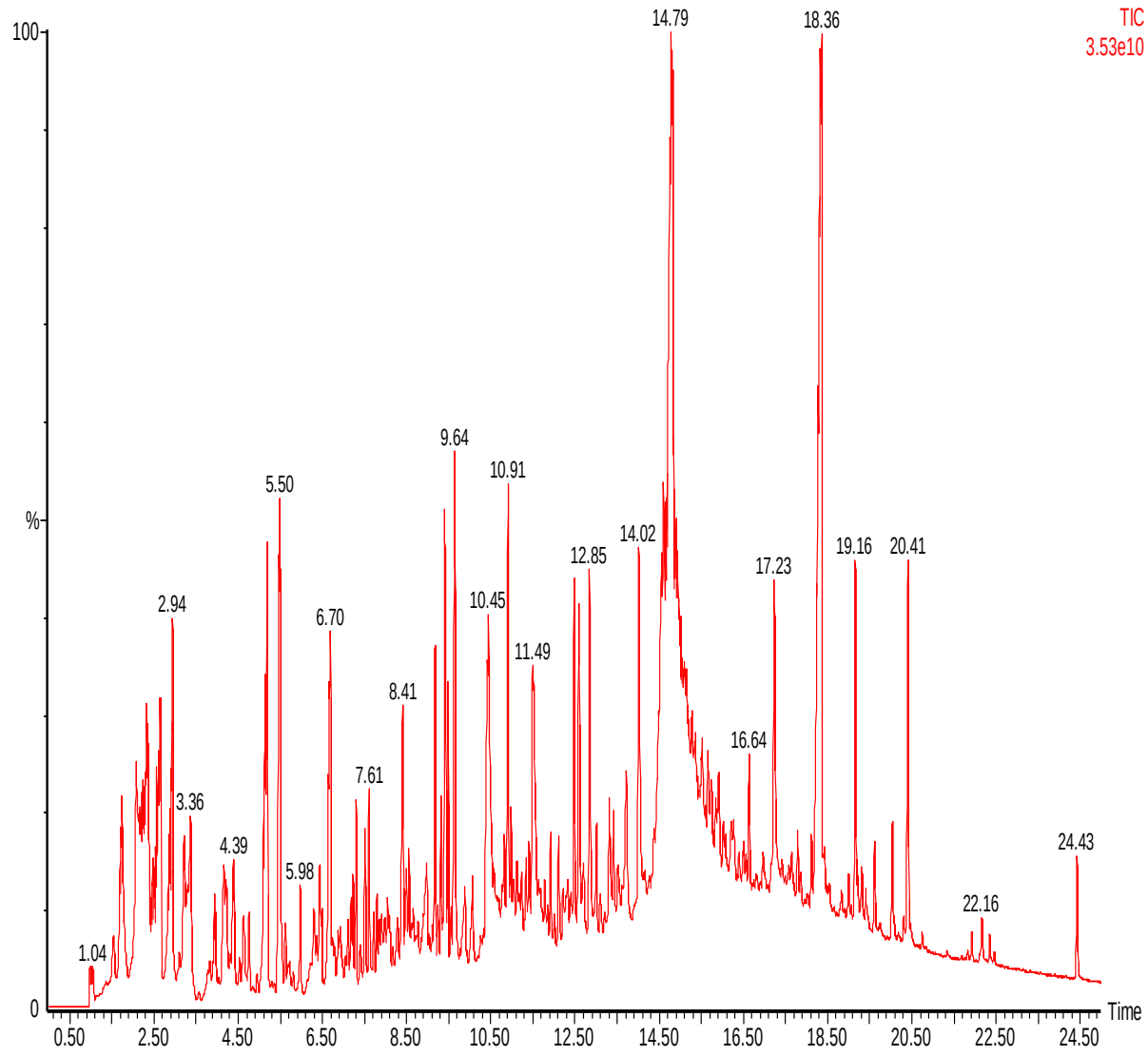

Figure 4.9 Chromatograph of essential oil extracted from *L. angustifulia* treated with 2.5g/L humic acid and 5g/L *Mycorrhiza* .

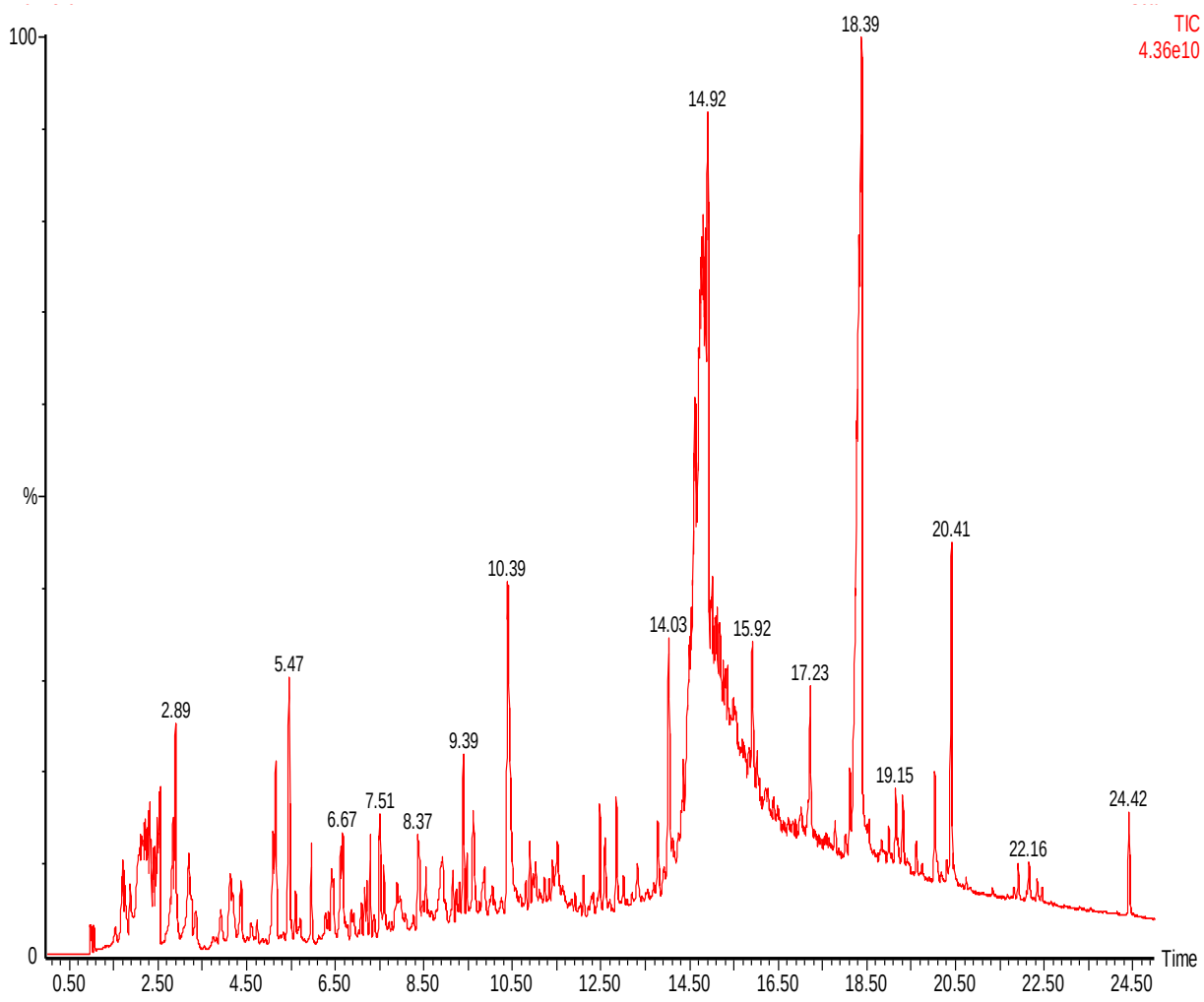

Figure 4.10 Chromatograph of essential oil extracted from *L. intermedia* treated with 2.5g/L humic acid and 5g/L *Mycorrhiza.*

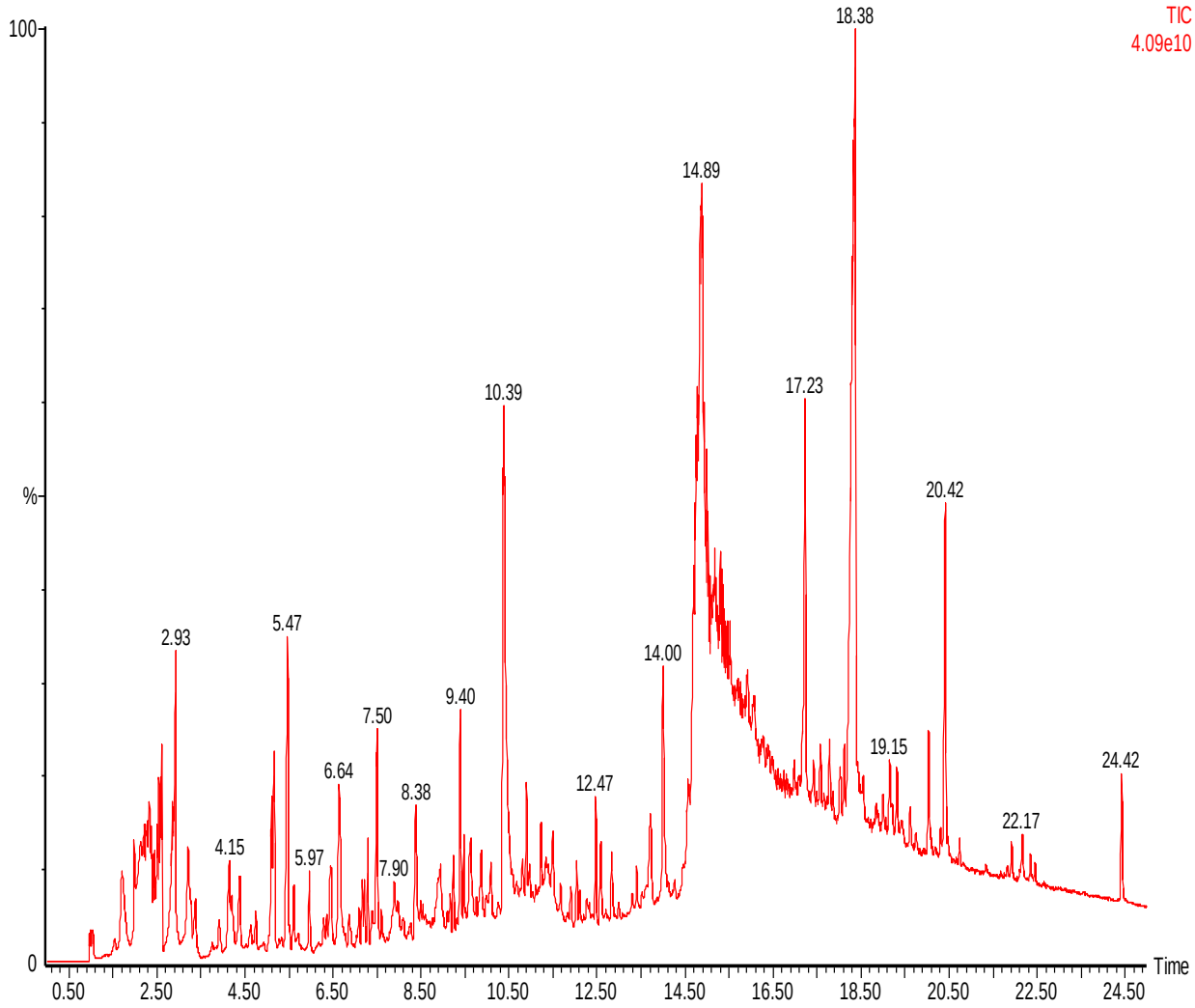

Figure 4.11 Chromatograph of essential oil extracted from *L. angustifulia* treated with 1.5g/L humic acid and10g/L *Trichoderma*.

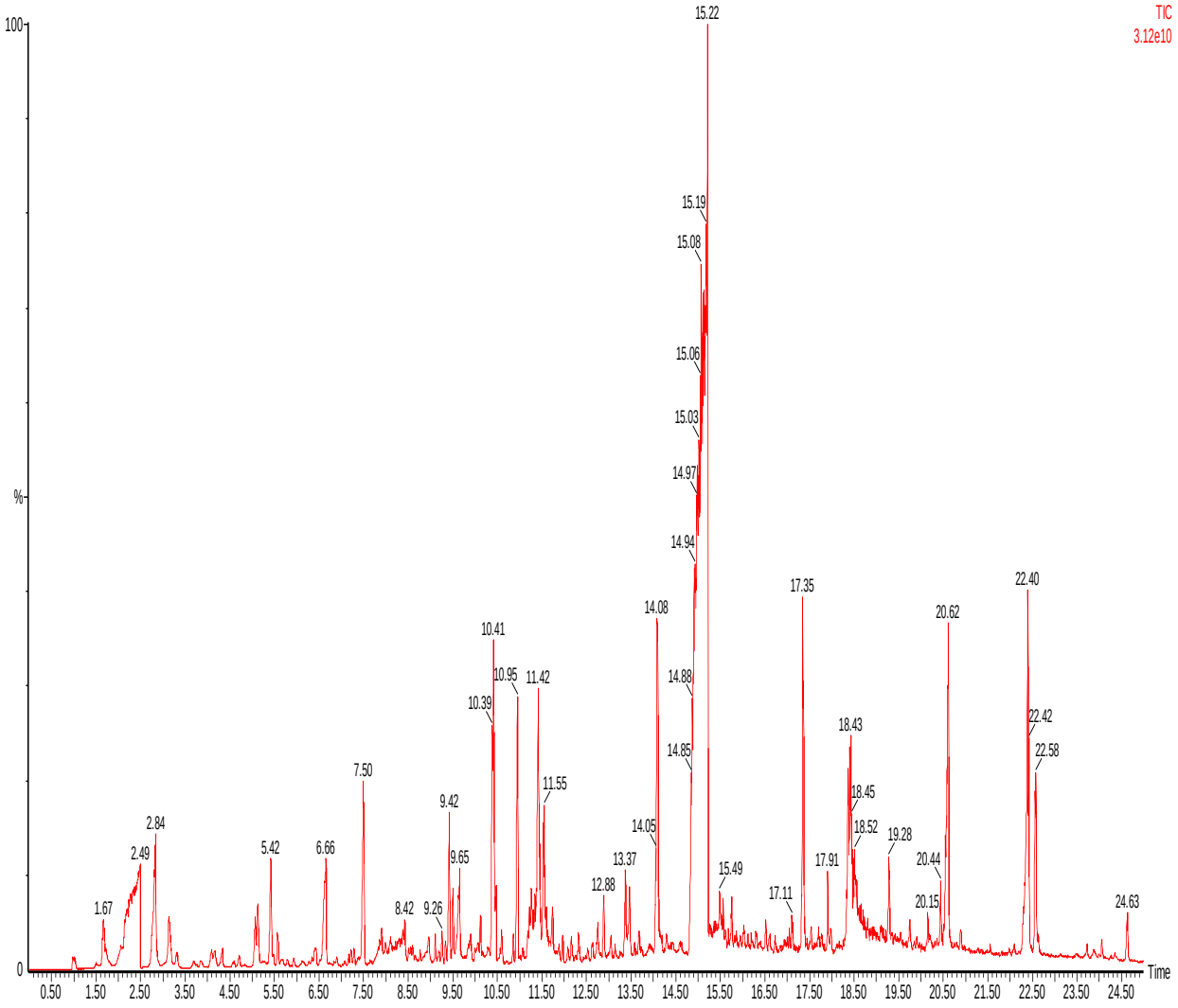

Figure 4.12 Chromatograph of essential oil extracted from *L. angustifulia* treated with 3.5g/L humic acid and 5g/L *Mycorrhiza* .

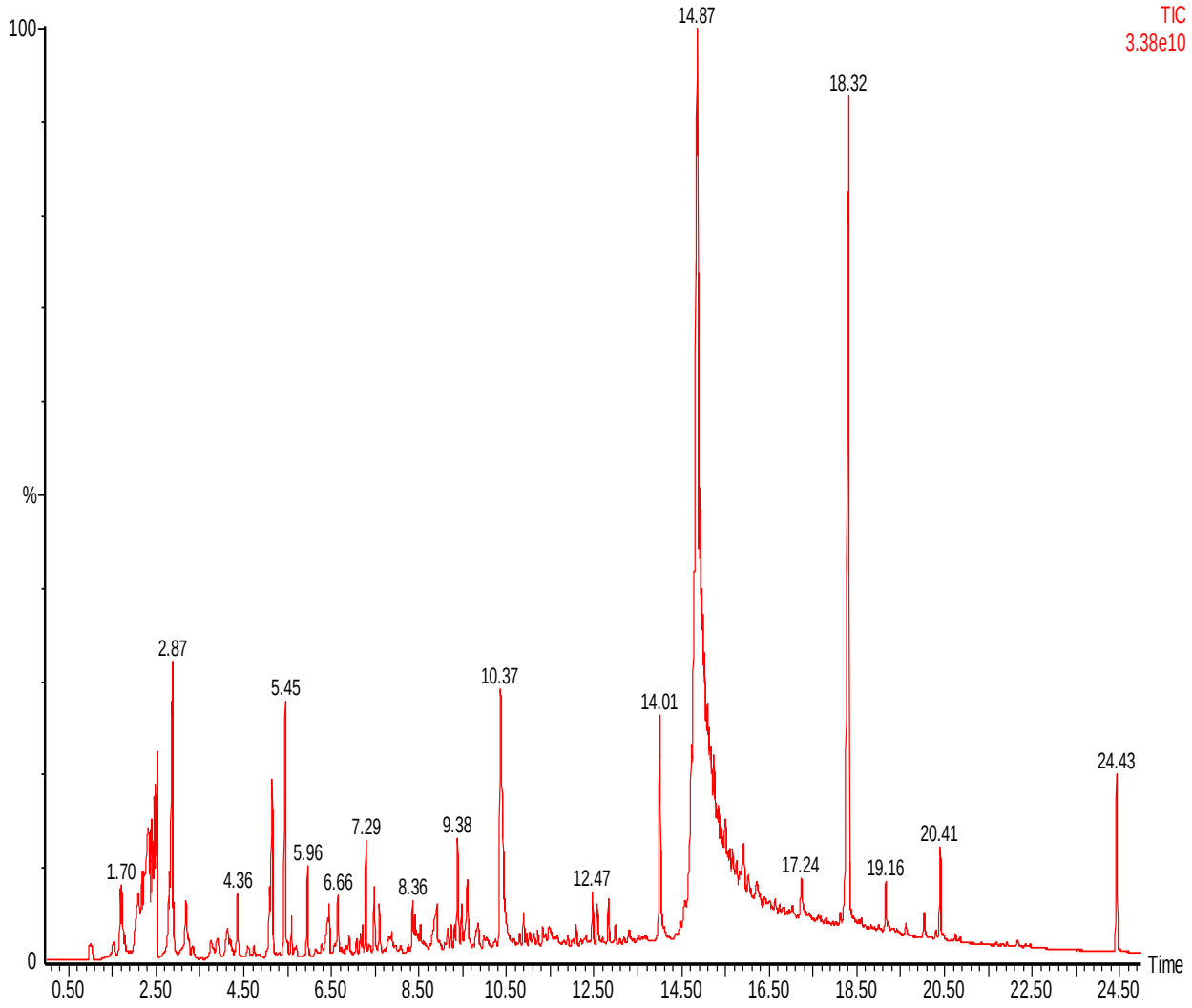

Figure 4.13 Chromatograph of essential oil extracted from *L. intermedia* treated with 3.5g/L humic acid and 5g/L *Mycorrhiza.*

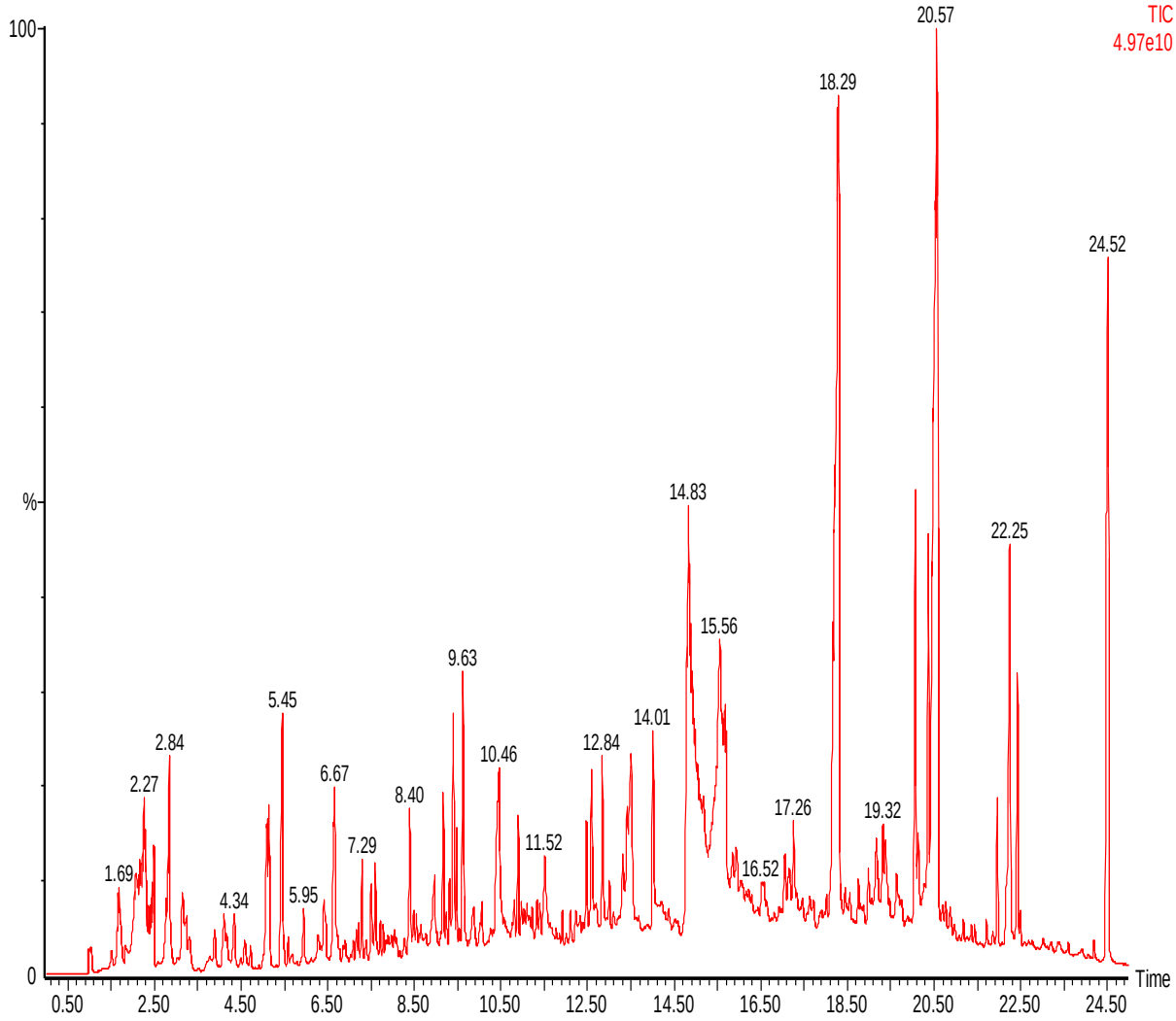

Figure 4.14 Chromatograph of essential oil extracted from *L. angustifolia* treated with 0g/L humic acid and 0g/L *Mycorrhiza*.

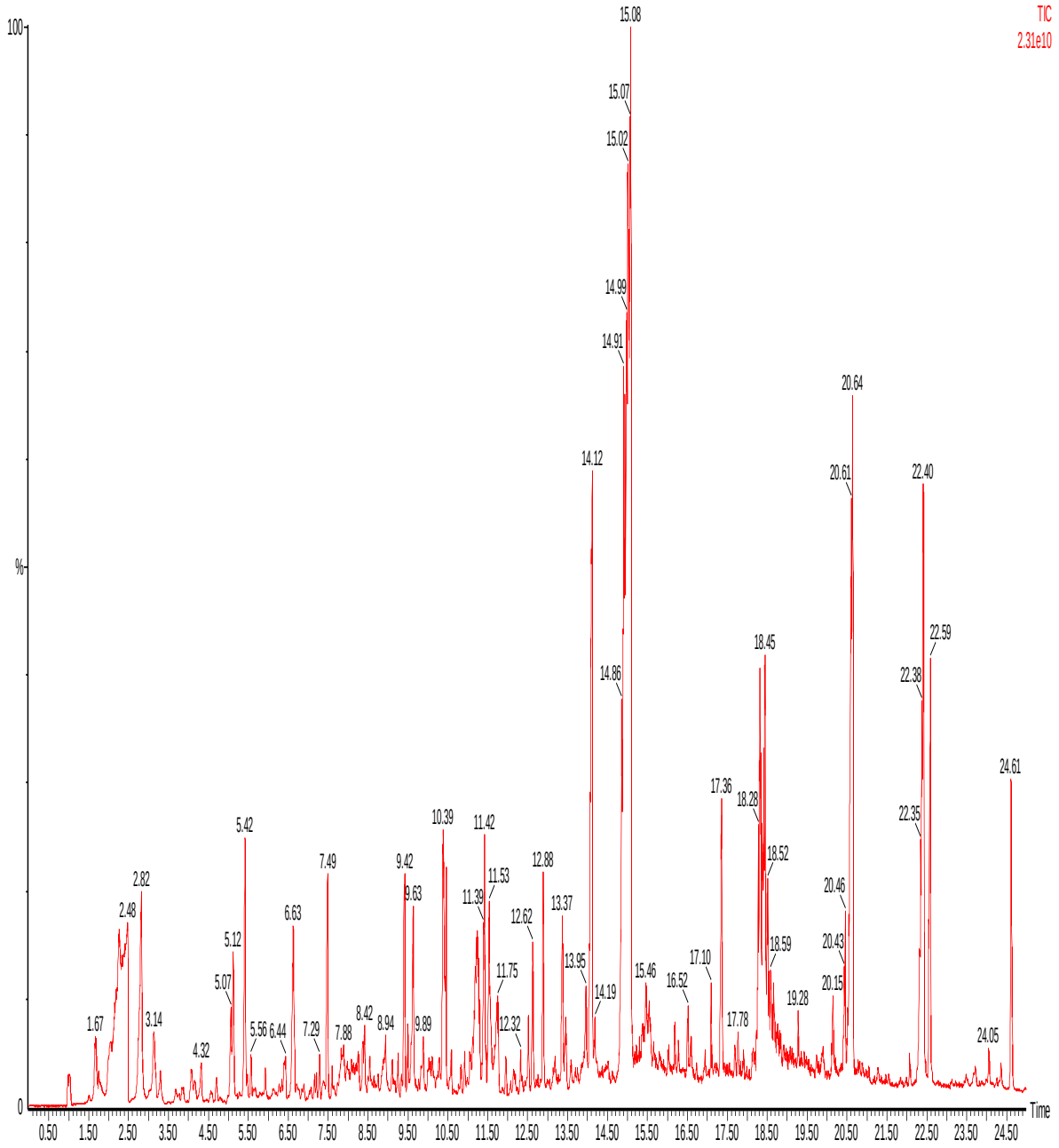

Figure 4.15 Chromatograph of essential oil extracted from *L. angustifulia* treated with 1.5g/L humic acid and 5g/L *Mycorrhiza*.

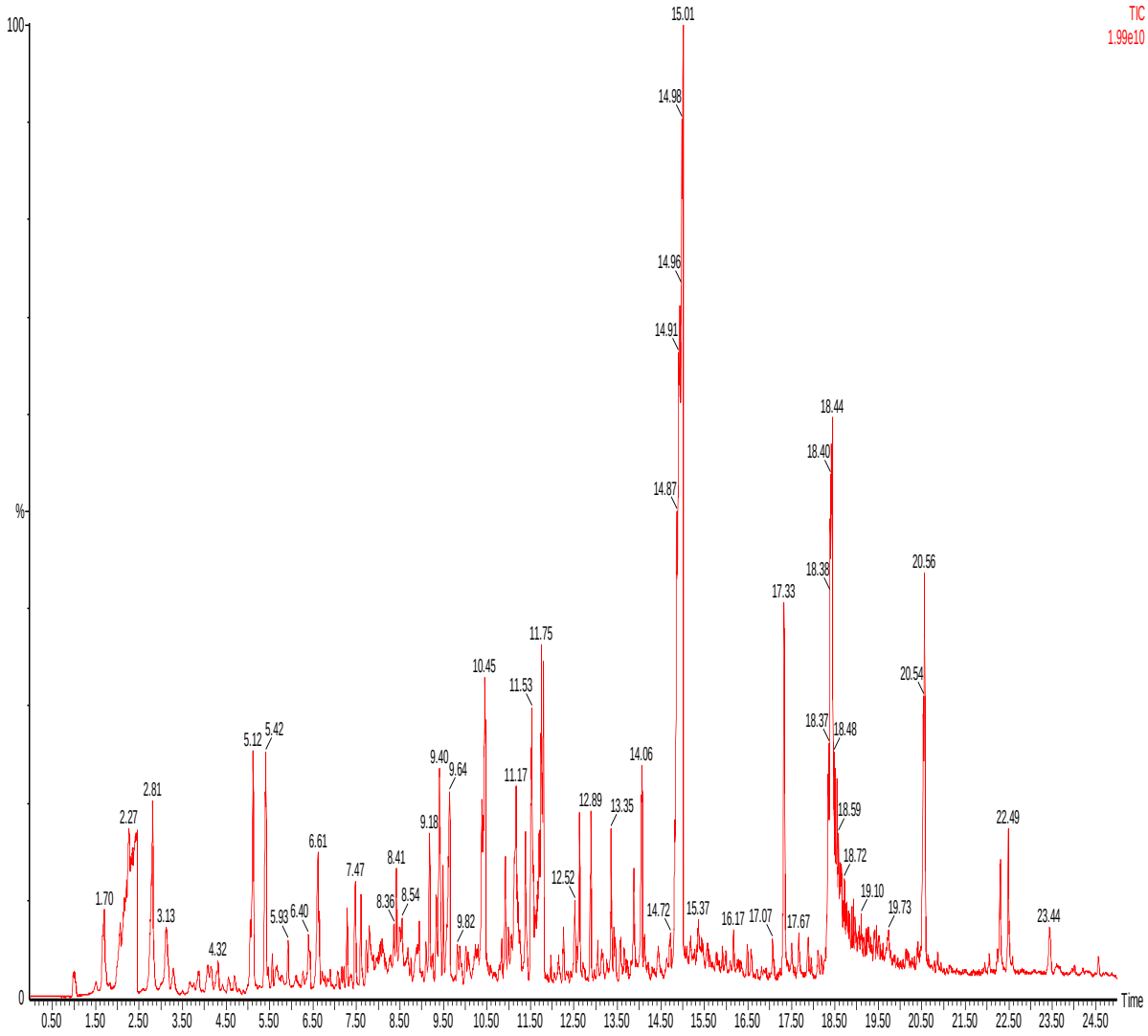

Figure 4.16 Chromatograph of essential oil extracted from *L. angustifulia* treated with 3.5g/L humic acid, 10g/L *Trichoderma*.

GC-MS was used to identify the main compounds in lavender oil which are linalool, linalyl acetate, sabinene hydrate, Trans Linalool, β-Pinene, Cymene, Linalool Oxide, and camphor (Fig 4.2to 4.16). GC-MS assays of the essential oil indicated that monoterpenes were distinguished as the main class of ingredients, followed by a secondary proportion of sesquiterpenes in leaves, flowers, and branches of the lavender species. Hassanpouraghdam et al.

(2011) found the large quantities of monoterpenes containing oxygen in oils support the dynamic assembly of plastid hydroxylases and hydrogen trends involved in the subsequent modification of the primary hydrocarbons.

Chemical identification and quantification

Table 4.3 Table 6: GC-MS identification and relative percentage concentration of components found in the lavender essential oil.

No.	RT(min)	Essential Oil Compound	Formula	percentage %	Molecular class	Identified Methods
1	1.008	Carny acetate	$C_{12}H_{18}O_2$	0.192549812		Pyro-GC-MS
2	1.679	Trans- β-Ocimene	$C_{10}H_{16}$	0.439794	Monoterpene	Pyro-GC-MS
3	2.254	Hexanol	$C_6H_{14}O$	1.332310156	Aliphatic alcohol	Pyro-GC-MS
4	2.491	1-Penten -3-ol	C_6H_{12}	0.628748541	Monoterpene	Pyro-GC-MS
5	2.839	α-Pinene	$C_{10}H_{16}$	1.35064767	Monoterpene	Pyro-GC-MS
6	3.233	β-pinene	$C_{10}H_{16}$	0.669156872	Monoterpene	Pyro-GC-MS
7	4.201	3-Carene	$C_{10}H_{16}$	0.393119149	Monoterpene	Pyro-GC-MS
8	4.685	Cis-β -Ocimene	$C_9H_{16}O_2$	0.258659199	Monoterp.alcohol	Pyro-GC-MS
9	5.099	Myrcene	$C_{10}H_{16}$	1.409498356	Monoterpenoids	Pyro-GC-MS
10	5.447	Cymene	$C_{10}H_{16}$	2.287461117	Monoterpene	Pyro-GC-MS
11	5.951	- terpinene-4-ol	$C_{10}H_{16}O$	0.291386732	Monoterpene	Pyro-GC-MS
12	6.319	Unknown	/	0.409573426	/	Pyro-GC-MS
13	6.455	Sabinene hydrate	$C_{10}H_{18}O$	0.457893432	Monoterpene	Pyro-GC-MS

14	6.662	Limonene	$C_{10}H_{16}$	1.45379587	Monoterpene	Pyro-GC-MS
15	7.217	Lavandulol	$C_{10}H_{18}O$	0.492380375	Monoterp.alcohol	Pyro-GC-MS
16	7.292	γ-Terpinene	$C_{10}H_{16}$	0.616937659	Monoterpene	Pyro-GC-MS
17	7.358	2-methyl-1-butanal	$C_5H_{12}O$	0.346796732	Monoterp. ether	Pyro-GC-MS

Table 4.7 (continued)

18	7.514	Bicyclo (4.1-0)hept-3-ene	C_7H_{8O}	0.557129136	Monoisotopic	Pyro-GC-MS
19	7.6	Terpinolene	$C_{10}H_{16}$	0.781592822	Monoterpene	Pyro-GC-MS
20	7.781	-7-Trans- Linalool oxide	$C_{10}H_{18}O_2$	0.255958234	Monoterp.alcohol	Pyro-GC-MS
21	7.953	Hexyl butyrate	$C_{10}H_{20}O_2$	0.344285033	Aliphatic ester	Pyro-GC-MS
22	8.402	trans-3-Caren-2-ol	$C_{10}H_{16}O$	1.000915334	Monoisotopic	Pyro-GC-MS
23	8.487	Toluene	C_7H_8	0.667027161	Methylbenzene	Pyro-GC-MS
24	8.966	3,4-hexanedione	$C_6H_{10}O_2$	0.588212315	Monoisotopic	Pyro-GC-MS
25	9.103	Linalool oxide	$C_{10}H_{18}O_2$	0.518074081	Monoterp.alcohol	Pyro-GC-MS
26	9.168	(Z)-Cinnamaldehyde	C_9H_8O	1.176005655	Monoisotopic	Pyro-GC-MS
27	9.319	α- Thujone	$C_{10}H_{16}O$	1.022646462	Monoisotopic	Pyro-GC-MS
28	9.405	Limonene oxide	$C_{10}H_{16}O$	2.141247091	Monoterpene	Pyro-GC-MS

29	9.481	Camphene	$C_{10}H_{16}$	1.251525003	Monoterpene	Pyro-GC-MS
30	9.627	Linalool	$C_{10}H_{16}O$	2.233760957	Monoterp.alcohol	Pyro-GC-MS
31	9.788	Sabinene	$C_{10}H_{16}$	0.422233368	Monoterpene	Pyro-GC-MS

Table 4.7 (continued)

32	10.01	Butanoic acid	$C_4H_8O_2$	0.422149502	Monoisotopic	Pyro-GC-MS
33	10.449	Linalyl acetate	$C_{12}H_{20}O_2$	1.861968756	Monoterp. ester	Pyro-GC-MS
34	10.822	α- Terpinene	$C_{10}H_{16}$	0.653615571	Monoterpene	Pyro-GC-MS
35	10.898	Par-Cymenene	$C_{10}H_{14}$	0.928306709	Monoterpene	Pyro-GC-MS
36	10.973	Camphor	$C_{10}H_{16}O$	0.710154277	Monoterp. ketone	Pyro-GC-MS
37	11.508	3-Caren-10-al	$C_{10}H_{14}O$	0.723946095	Monoterpene	Pyro-GC-MS
38	12.396	Cumin aldehyde	$C_{10}H_{12}O$	0.4123527	Monoterpene	Pyro-GC-MS
39	12.481	D-Carvone	$C_{10}H_{14}O$	1.05792457	Monoterpene	Pyro-GC-MS
40	12.597	Geraniol	$C_{10}H_{18}O$	1.385554837	Monoterp. ether	Pyro-GC-MS
41	12.85	-1,8-Cireole	$C_{10}H_{18}O$	1.328351869	Monoterp.alcohol	Pyro-GC-MS

42	13.485	Cis-Linalool oxide	$C_{10}H_{18}O_2$	1.876236291	Monoterp.alcohol	Pyro-GC-MS
43	14.009	Nerol	$C_{10}H_{18}O$	.792425201	Monoterp.alcohol	Pyro-GC-MS
44	14.115	3- Carene	$C_{10}H_{16}$	0.800928203	Monoterpene	Pyro-GC-MS
45	14.846	α-Terpineol	$C_{10}H_{18}O$	4.592931332	Monoterpene	Pyro-GC-MS

Table 4.7 (continued)

46	15.558	Borneol	$C_{10}H_{18}O$	3.293906627	Monoterp.alcohol	Pyro-GC-MS
47	15.986	Caryophyllene oxide	$C_{15}H_{24}O$	0.910607198	Sesquiterpene	Pyro-GC-MS
48	16.576	-Cadinol	15H	0.549516129	Sesquiterpenoid	Pyro-GC-MS
49	17.257	(E,E)-Farnesol	$C_{15}H_{26}O$	0.895903525	Monoisotopic	Pyro-GC-MS
50	17.625	Thuja-2,4(10)-diene	$C_{10}H_{14}$	0.350204307	Monoterpene	Pyro-GC-MS
51	17.918	Cis-α-Bergamotene	$C_{15}H_{24}$	0.290768582	Sesquiterpene	Pyro-GC-MS
52	18.291	Geranyl acetate	$C_{12}H_{20}O_2$	9.767224029	Monoterp. ester	Pyro-GC-MS
53	18.573	Trans-α-Bergamotene	$C_{15}H_{24}$	0.317684787	Sesquiterpene	Pyro-GC-MS
54	18.71	Di isobutyl phthalate	$C_{16}H_{22}O_4$	0.324690403	Monoterp. ester	Pyro-GC-MS

55	19.32	Myrlenyl acetate	$C_{12}H_{18}O_2$	0.961492764	Monoisotopic	Pyro-GC-MS
56	19.693	Neryl acetate	C12H20O2	0.492731271	Monoisotopic	Pyro-GC-MS
57	19.965			0.308380609	-	Pyro-GC-MS
58	20.071	Trans-Linalool oxide	$C_{10}H_{18}O_2$	3.948007842	Monoterp.alcohol	Pyro-GC-MS
59	20.132	α-Caryophyllene	$C_{15}H_{24}$	0.82842743	Sesquiterpene	Pyro-GC-MS

Table 4.7 (continued)

60	20.374	α- Long pinene	$C_{10}H_{16}$	3.634527334	Monoterpene	Pyro-GC-MS
61	20.576	E-Caryophyllene	$C_{15}H_{24}$	10.08404173	Sesquiterpene	Pyro-GC-MS
62	20.818	β-Caryophyllene	$C_{15}H_{24}$	0.265155555	Sesquiterpene	Pyro-GC-MS
63	20.949	1-Tricosene	$C_{23}H_{46}$	0.1780754	Monoisotopic	Pyro-GC-MS
64	21.877	Germacrene D	$C_{15}H_{24}$	0.306505569	Sesquiterpenes	Pyro-GC-MS
65	21.962	Neryl acetate	$C_{12}H_{20}O_2$	0.880270706	Monoterp. ester	Pyro-GC-MS
66	22.063	Benzaldehyde	C_6H_5CHO	0.212411536	Monoisotopic	Pyro-GC-MS
67	22.245	α-Cedrene	$C_{15}H_{24}$	4.166936997	Monoterpenoids	Pyro-GC-MS

68	22.426	Lavandulyl acetate	$C_{12}H_{20}O_2$	2.330746741	Monoterp.alcohol	Pyro-GC-MS
69	22.497	α-Bergamotene	$C_{15}H_{24}$	0.638138505	Sesquiterpene	Pyro-GC-MS
70	22.779	Methoxy cinnamaldehyde	$C_{10}H_{10}O_2$	0.191081033	Aliphatic alcohol	Pyro-GC-MS
71	23.082	α-Bulnesene	$C_{15}H_{24}$	0.19898576	Sesquiterpenoids	Pyro-GC-MS
72	24.086	Eugenol	$C_{10}H_{12}O_2$	0.252126818	Phenylpropene	Pyro-GC-MS
73	24.202	Limonene diol	$C_{10}H_{18}O_2$	0.271911769	Monoterpene	Pyro-GC-MS

Table 4.7 (continued)

74	24.514	Cadala-1(10)-3-8-triene	$C_{15}H_{22}$	7.885120051	Monoisotopic	Pyro-GC-MS
75	24.6	Trans-Cadinene ether	$C_{15}H_{24}$	0.577726498	Monoisotopic	Pyro-GC-MS
76	24.716	α-Element	$C_{15}H_{24}$	0.148524836	Actinium	Pyro-GC-MS

Total chromatograph (GCMS) of lavender (leaves, flowers, and stems) of essential oil, name, and retention time (min) of compounds: linalool, linalyl acetate, cymene, trans linalool, sabinene hydrate, β-pinene, linalool oxide, camphor.

Table 4.4 Qualitative and quantitative GC-MS data for essential oil components identified in lavender.

No	RT	Compound	Treatment											
			1	2	3	4	5	6	7	8	9	10	11	12
1	1.008	Caray acetate		-										
2	1.679	Trans- β-Ocimene	1.28	1.61	1.24	1.32	1.04	0.96	1.23	1.54		0.71	1.78	1.35
3	2.254	Hexanol	3.38	4.09		3.00	2.69	2.23	2.97	2.52	2.19	1.72	2.70	3.70
4	2.491	1-penten-3-ol			2.41			1.10	1.59				2.59	4.34
5	2.839	α-Pinene	2.84	3.37		3.01	1.99	2.16	3.55	2.80	3.87	3.84	3.77	6.66
6	3.233	β-pinene	0.72	1.60	2.02	1.18	0.64	1.22	1.99	1.56	1.42	1.38	0.98	1.00
7	4.201	3-Carene		0.82		0.85	0.29	0.69	1.19	1.10		1.21		
8	4.685	Cis- β-Ocimene		-		0.37	0.18	0.39	0.60	0.60	0.40			
9	5.099	Myrcene	1.42	2.46	3.17	2.69	1.19	1.61	2.67	3.30	2.25	2.85	3.74	
10	5.447	Cymene	2.99	5.50	5.81	5.47	2.35	2.78	5.42	4.02	5.26	4.80	2.31	5.45
11	5.951	-terpinen-4-ol		0.84										
12	6.319													
13	6.455	Sabinene hydrate	-	0.88	1.13	0.86		0.37		0.58	0.91		0.30	0.73
14	6.662	Limonene	2.52	3.34	3.78	2.15	2.01	2.29	3.90	2.58	1.41	1.77	1.76	
15	7.217	Lavandulol		2.16		0.45		0.52					0.39	

Table 4.8 (continued)

16	7.292	γ-Terpinene				0.96		0.56					0.91	1.7
17	7.358	2-methyl-1-butanol	0.41			0.45								
18	7.514	Bicyclo (4.1.0)hept-3				1.03	0.36	0.55	2.09			2.35	1.67	
19	7.6	Terpinolene											0.66	
20	7.781	-7trans-Linalool oxide	0.31	0.51			0.22				1.79		0.93	

21	7.953	Hexyl butyrate			0.59	0.49		0.48	0.59	0.27	0.58	0.79	0.28	
22	8.402	trans-3-Caren-2-ol		0.90	1.59	0.63		0.80	1.50	1.60	1.18	1.57	1.38	0.8
23	8.487	Toluene			0.93			0.79	0.85	0.90			0.55	
24	8.966	3,4-hexanedione	-	1.03		1.28			0.75	0.37	0.97	0.82		0.8
25	9.103	Linalool oxide		3.28	3.30	2.53	1.04	1.43	2.78	3.00	1.93	2.09	1.31	1.5
26	9.168	(Z)-cinnamaldehyde								1.70			0.17	
27	9.319	α-Thujone	1.59										0.81	
28	9.405	Limonene oxide		3.28	3.30	2.53	1.04			3.00			1.56	
29	9.481	Camphene	1.15	1.35	1.65	1.09	1.37	1.56	1.92	1.32			1.22	
30	9.627	Linalool	1.21	2.17	1.68	1,23	1.01	0.78	1.03	3.40			2.55	

Table 4.8 (continued)

31	9.788	Sabinene										0.21	
32	10.01	Butanoic acid				2.35	0.29					0.17	
33	10.449	Linalyl acetate			1.69	1.17		4.82	1.27	2.92	6.35	8.46	1.95	
34	10.822	α-Terpinene	0.77				1.50	1.32		1.37			0.48	
35	10.898	Para-cymenene	10.37				18.27		6.52	3.33			2.95	
36	10.973	Camphor	0,89	0.98		1.10	0.54	14.00		1.42			0.46	
37	11.508	3-Caren-10-al			0.91					2.48	0.83		0.48	
38	12.396	Cumin aldehyde			0.35					0.78			0.19	
39	12.481	D-carvone		0.52	1.44					2.61				
40	12.597	Geraniol		1.54	0.53	1.00				2.63			0.13	

41	12.85	-1,8-Cireole	5.43	3.39	3.34	3.92	4.43		0.74		1.21		0.34	6.7
42	13.485	Cis-Linalool oxide		0.50										
43	14.009	Nerol		6.73	6.95	7.95		3.37	5.37	2.94	4.49	3.49	0.69	5.1
44	14.115	Z-caryophyllene												

Table 4.8 (continued)

45	14.846	α-Terpineol	0.66	0.58	10.31	1.41	11.19	7.23	12.10	9.10		14.56	
46	15.558	Borneol											0
47	15.986	Caryophyllene oxide									4.77		
48	16.576	-Cadinol					0.46	0.95		1.17			0
49	17.257	(E,E)-Farnesol	1.16	0.91	1.60	2.02	2.99	5.22	4.91	2.76	3.55	8.56	1
50	17.625	Thuja-2,4(10)-diene	0.41		0.82	0.61	0.48	0.56	0.95				0
51	17.918	Cis-α Bergamotene	0.54	0.54	0.71	0.46	0.49	0.40					0
52	18.291	Geranyl acetate	14.90	11.32	13.77	15.34			13.64	8.66	18.88	16.65	2
53	18.573	Trans-α-Bergamotene	0.50				0.43		1.78	1.37			0
54	18.71	Diisobutylphthalate			0.53	0.51	0.39						0
55	19.32	Myrlenyl acetate	1.24	0.21	0.54	0.41				2.61			0
56	19.693	Neryl acetate	0.27		0.23	0.19	1.01	0.49		0.34			0
57	19.965												
58	20.071	Trans-Linalool oxide		1.36	0.68	1.15		0.22	0.60			2.14	

Table 4.8 (continued)

59	20.132	α-caryophyllene											0.15	
60	20.374	α-Long pinene								0.46		1.56	1.03	1.46
61	20.576	E-Caryophyllene									1.17	6.18	2.95	
62	20.818	β-caryophyllene											0.14	
63	20.949	1-Tricosene												
64	21.877	Germacrene D												
65	21.962	Neryl acetate		0.52										
66	22.063	Benzaldehyde												
67	22.245	Lavendulol									0.44	0.65	3.29	
68	22.426	Lavandulyl acetate	0.36			0.47	0.41	0.30					0.75	
69	22.497	α-Bergamotene						0.23				0.33	1.29	
70	22.779	Methoxycinnamaldehde												
71	23.082	α -Bulnesene												
72	24.086	-Eugenol												
73	24.202	Limonene diol												

Table 4.8 (continued)

74	24.514	Cadala-1(10)-3-8-triene	6.16	3.02	2.59	4.00	3.69	0.26		0.51			2.43	3.46
75	24.6	Trans-Cadinene ether												
76	24.716	α-Element												

1=3.5g/l humic acid, 5g *mycorrhiza*, with *L. angustifulia* .
2=3.5g/L humic acid, 10g *Trichoderma*, with *L. stoechas*.
3=2.5 g /L humic acid, 5g/L *Mycorrhiza*, with *L. stoechas*.
4=3.5 g /L humic acid, 5g/L *Mycorrhiza* with *L. stoechas*.

5=0g/L humic acid, 0 biofertilizer with *L.stoechas*.
6=1.5g/L humic acid, 5g/L *Mycorrhiza*, with *L. intermedia.*
7=0g/L humic acid, 0g/L biofertilizer with *L. intermedia.*
8=2,5g/L humic acid, 5g/L *Mycorrhiza*, with *L. angustifulia.*
9=2.5g/L humic acid, 5g/L *Mycorrhiza*, with *L. intermedia.*
10=1.5g/L humic acid, 10g/L *Trichoderma,* with *L. angustifulia.*
11=3.5g/L humic acid, 5g/L *Mycorrhiza,* with *L. angustifulia.*
12=3.5g/L humic acid, 5g/L *Mycorrhiza*, with *L. intermedia.*
13=0g/L humic acid, 0g/L biofertilizer, with *L.angustifolia*
14=1.5g/L humic acid, 5g/L *Mycorrhiza*, with *L. angustifulia.*
15=3.5g/L humic acid, 10g/L biofertilizer, with *L.angustifulia*., RT= Retantion Time, unite measure= %

Table 4.5 Humic acid and mycorrhizae effects on essential oils of *Lavandula* analyzed using GC-MS.

Essential oils compounds								
Samples	Linalool	Linalyl acetate	Sabinene Hydrate	Trans Linalool	β-Pinene	Cymene	Linalool Oxide	Camphor
$H^{3.5}$ × *L. angustifolia* × T^{10}	1.21	0.03	0.02	0.31	0.72	2.99	0.05	0,89
$H^{3.5}$ × *L. stoechas* × M^{5}	2.17	0.05	0.88	0.51	1.6	5.5	3.28	0.98
$H^{2.5}$ × *L. stoechas* × T^{10}	1.68	1.69	1.13	0.04	2.02	5.81	3.3	0.02
$H^{3.5}$ × *L. stoechas* × T^{10}	1,23	1.17	0.86	0.3	1.18	5.47	2.53	1.1
L. stoechas	1.01	0.02	0.03	0.22	0.64	2.35	1.04	0.54
$H^{1.5}$ × *L. intermedia* × T^{10}	0.78	4.82	0.37	0.05	1.22	2.78	1.43	14
L. intermedia	1.03	1.27	0.04	0.05	1.99	5.42	2.78	0.06
$H^{2.5}$ × *L. angustifolia* × T^{10}	3.4	2.92	0.58	0.03	1.56	4.02	3	1.42
$H^{2.5}$ × *L. intermedia* × T^{10}	0.02	6.35	0.91	1.79	1.42	5.26	1.93	0.07
$H^{1.5}$ × *L. angustifolia* × M^{5}	0.03	8.46	0.06	0.02	1.38	4.8	2.09	0.07
L. intermedia × T^{10}	2.55	1.95	0.3	0.93	0.98	2.31	1.31	0.46

Table 4.9 (continued)

$H^{3.5}$ × *L. intermedia* × T^{10}	0.04	0.05	0.73	0.03	1.00	5.45	1.56	0.07
L. angustifolia	2.23	1.86	0.45	0.04	0.66	2.28	0.51	0.71
$H^{1.5}$ × *L. stoechas* × T^{10}	1	0.7	0.35	0.24	0.66	1.67	0.03	0.02
$H^{3.5}$ × *L. angustifolia* × M^{5}	3.34	4.02	0.02	0.23	0.42	1.55	0.66	0.61
Constant value	1.42	1.04	0.15	0.07	1.10	3.35	1.44	0.42
(*p* value=0.05)	0.332 ns	0.67 ns	0.013	0.117 ns	0.63 ns	2.54 ns	0.369 ns	0.267 ns
T test	1.004	1.989	2.826	1.668	0.493	1.191	0.876	1.157

H= Humic acid & mycorrhizae (M=*Mycorrhiza* 5 g/l & T=*Trichoderma* 10g/l

Lavandula essential oil components and their concentrations were obtained with GC-MS using linalool and linalyl standards. The oil components and concentrations were analyzed by humic acid and mycorrhizae treatments (Table4.7-4.8) (Fig. 4.17). Linalool, linalyl acetate, sabinene hydrate, trans linalool, β-pinene, cymene, linalool oxide, and camphor were determined to be the main constituents in the essential oil of lavender fresh stem, flowers, and leaves. The highest linalool content was determined in *L. angustifolia* with 3.5 g humic acid/L and 5g *Mycorrhiza* /L. The lowest linalool content was obtained from *L. intermedia* treated with 2.5 g humic acid/L and 10g *Trichoderma*/L. The highest linalyl acetate content was found in *L. angustifolia* treated with humic acid at 1.5 g /L and 5g *Mycorrhizae*/L while *L. stoechas* with no humic acid or mycorrhizae had the least linalyl acetate. Both levels of humic acid increased the concentration of linalool in the oil compared to other treatments. Sabinene hydrate concentration was highest in *L. stoechas* and *L. intermedia* receiving 2.5g humic acid/L and 10g *Trichoderma*/L. The highest trans linalool content was determined in *L. intermedia* treated with 2.5g humic acid/L and 10g *Trichoderma*/L and *L. stoechas* treated with 3.5g humic acid/L and 5g *Mycorrhizae*/L. The lowest trans linalool content was obtained from *L. angustifolia* treated with 1.5g humic acid/L and 5g *Mycorrhiza*/L. The highest β-Pinene content was obtained from *L. stoechas* and *L. intermedia* receiving 2.5g humic acid/L and 10g Trichoderma/L. The lowest β-Pinene content was obtained from *L. angustifolia* treated with 3.5g humic acid/L and 5g *Mycorrhizae*/L. The highest cymene content was determined from *L. stoechas* at 2.5g or 3.5 g humic acid/L and 10 g *Trichoderma*/L. The lowest cymene content was obtained from *L. angustifolia* at 3.5g humic acid/L and 5g *Mycorrhizae*/L. The highest linalool oxide content was determined from *L. stoechas* treated with 3.5g humic acid/L and 5 g *Trichoderma*/L and in *L. stoechas* treated with 2.5 g humic acid/L and 10 g *Trichoderma*/L. The lowest linalool oxide

content was obtained from *L. stoechas* treated at 1.5g humic acid/L and 10 g *Mycorrhizae*/L. The highest camphor content was determined from *L. stoechas* treated with 1.5 g humic acid/L and 0g *Trichoderma*/L and *L. angustifolia* treated with 2.5 g humic acid/L and 10 g *Trichoderma*/L. The lowest camphor content was obtained from *L. stoechas* treated with 2.5 g humic acid/L and 10 g *Mycorrhizae*/L. There are no interspecific differences, humic acid, and mycorrhizae in the concentration of most of the essential oil components identified by GC-MS analysis due to the difference between lavender species.

The specific essential oil components are responsible for the properties of lavender flavor and their biological and therapeutic properties. The essential oil composition of lavender and lavandin can vary depending on the genotype of the plant (Lawrence, 1994), growth stage on the date of harvest, parts of the plant, and time of day or year when harvested (Baydar, 2009; Avcı, 2010) as well as the drying conditions and extraction technology (Pinto et al., 2007). There is a wide variation in the quantitative composition of lavender oil depending on the genotype of the plant and the area of cultivation, and the composition of lavender oil. It has been recognized that they differ significantly according to altitude, and local climate (Lawrence, 1991; 1994). Srinivasan, (2007) and Abbasi et al. (2011) explain the production of secondary metabolites in plants is significantly affected by environmental stress. As biotic and cellular abiotic stresses delay differentiation through the production of reactive oxygen species, which directly destroy cells by producing secondary metabolites.

Verma and Rahman (2010) reported the principal components of lavender essential oils were linalyl acetate, linalool, lavandolyl acetate, alpha-terpineol, geranyl acetate, caryophyllin oxide and 1.8-cineole along with other secondary ingredients including β-caryophyllene,

borneol, epi-α-cadinol, nerol, terpinen-4-ol, β-myrcene, limonene, and 1-octane-3-ol. Shawl et al. (2005) found lavender oil from the Kashmir valley contained large amounts of limonene, citronellol, and α-terpineol and low amounts of linalool. In addition, the application of humic acid caused a significant increase in the content of essential oils and a slight increase in the content of carvacrol and para semin reduced in contents -terpinene, β-bisabolene, and myrcene. Ghelardini et al. (1999) described the chemical composition of the essential oil of *L. angustifolia* of Italian origin, which was characterized by linalool (31.5%), linalyl acetate (43.0%), and -caryophyllene (5.0%) as the main compounds. While Jianu et al. (2013) reported Romanian lavender essential oil was richer in camphor and 1,8-cineole. In the same line, Verma et al. (2010) indicated the essential oil of lavender grown in India consisted of linalool (28.06%) and linalyl acetate (47.5%). Generally, in the present study, linalyl acetate content was higher than linalool content in all the lavender species. According to quality standards of lavender essential oil composition determined by the International Organization for Standardization (ISO 3515:2002), cymene, terpinen-4-ol and camphor should be between 25.0-38.0 %, 25.0-45.0 %, 4.0-10.0%, 2.0-6.0 %, and 0-0.5 %, respectively (Anonymous, 2009). Baydar (2009) stated that a lower camphor content was better quality for lavender essential oil. The camphor content of high-quality lavender oil should be between 0.5 and 1% and and between 5.0 and 10.0% in lavandin.

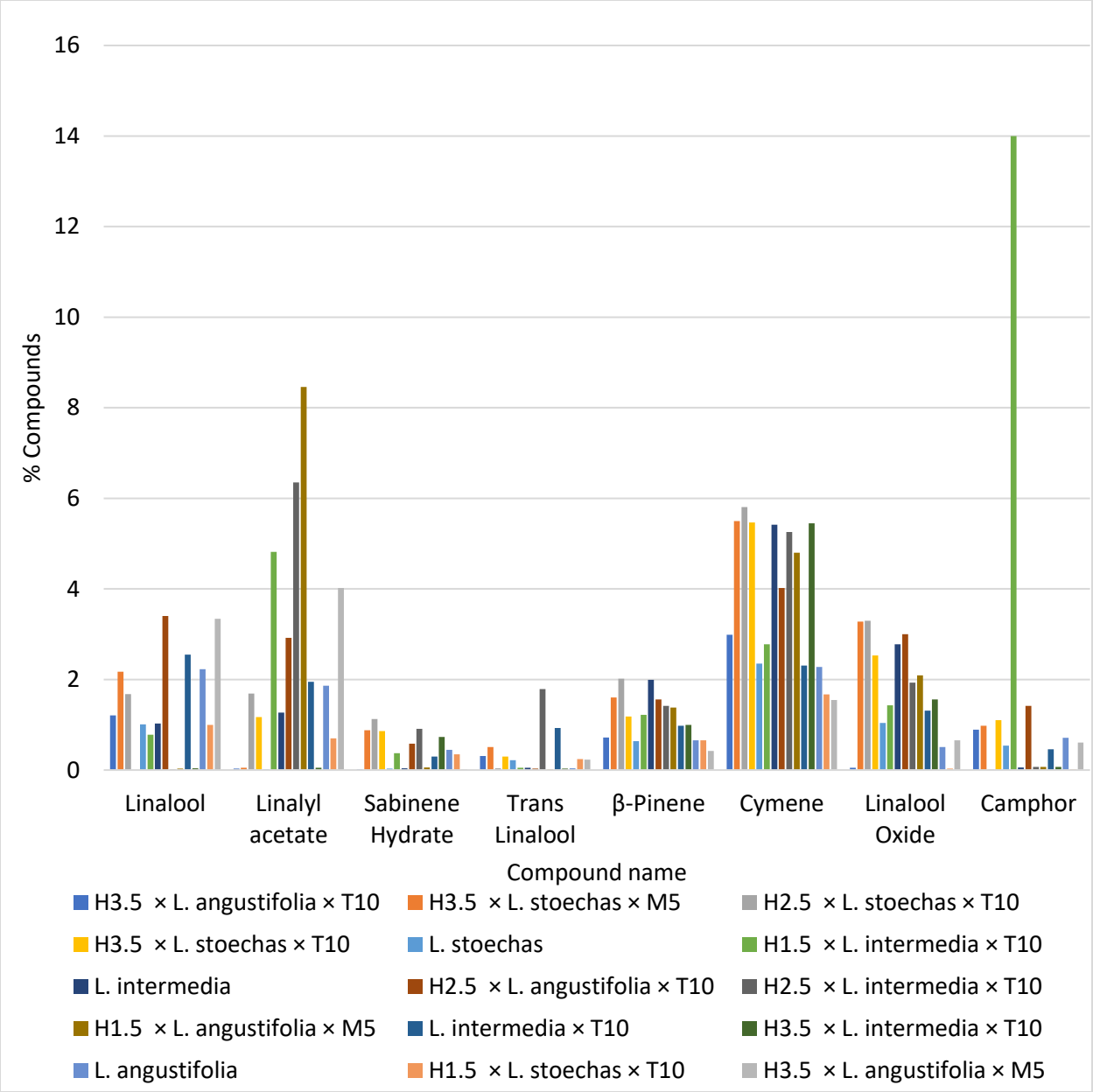

Figure 4.17 Essential oils components of lavender oil from plants treated with humic acid and Mycorrhizae as determined by GC-MS.

Conclusion

Humic acid and mycorrhizae affect essential oil content (quantity and quality of chemical formulations) in lavender. The use of humic acid and mycorrhizae may be a powerful method for enhancing plant growth and essential oil production. Specifically, the highest rate of humic acid (3.5 g/L) and *Mycorrhiza* were beneficial in enhancing essential oil content in lavender. The cultivar *L. angustifolia* was identified as a high-yielding species. The highest content of linalyl acetate was higher than that of linalool. *L. stoechas* tended to have lower linalyl acetate and linalool and higher concentrations of camphor and other compounds noted in lesser quality essential oils.

Literature cited.

Abbasi, B.H., M. Khan, B. Guo, S.A. Bokhari, and M.A. Khan. 2011. Efficient regeneration and antioxidative enzyme activities in *Brassica rapa* var. *turnip*. Plant Cell Tiss. Organ Cult. 105(3): 337-344.

Abdel-Razzak, H.S., and G.A. El-Sharkawy. 2013. Effect of Biofertilizer and humic acid applications on growth, yield, quality, and storability of two garlic (*Allium sativum L.*) Cultivars. Asian J. Crop Sci. 5(1): 48-64.

Abdul-Al-Hayani, A. M. 2016. Effect of origin and spraying of humic acid in carrying citrus leek seedlings for irrigation water salinity: 2- Chemical properties. Basra J. Agric. Sci. 29(2): 545-560.

Ahmed, Y.M., E.A. Shalaby, and N.T. Shanan. 2011. The use of organic and inorganic cultures in improving vegetative growth, yield characters and antioxidant activity of roselle plants (*Hibiscus sabdariffa* L.). Afr. J. Biotechnol. 10(11): 1988-1996.

Akinci, S., T. Buyukkeskin, A. Eroglu, and B. E. Erdogan. 2009. The effect of humic acid on nutrient composition in broad bean (*Vicia faba* L.) roots. J. Nut. Biol. Sci. 1(1): 81-87.

Al-Ali, H. H., and A.H. Abdul Majid. 2013. Effect of spraying of humic acid on some vegetative, physiological, and anatomical characteristics of the plant of the lettuce L. (*Lactuca sativa* L.). J. Euphrates Agric. Sci. 5(2): 26-37.

Amarowicz R, O. Narolewska M. Karamać, A. Kosińska, and S. Weidner. 2008. Grapevine leaves as a source of natural antioxidants. Pol. J. Food Nutr. Sci. 58(1): 73-78.

Ameri, A., and A. Tehranifar. 2012. Effect of humic acid on nutrient uptake and physiological characteristic *Fragaria ananasa* var: Camarosa. J. Biol. Environ. Sci. 6(16): 77-79.

Amin, G.A., M.H. Mostafa, A.A. Amer, and I.R. Refae. 1999. Potential microorganisms for rapid and nutritionally enriched compost. Proc. of 1st Con. On Recent Technologies in Agric., Cairo Univ., Faculty of Agriculture, Egypt, 27-29:804-822.

Andrys, D., and D. Kulpa. 2017. In vitro propagation affects the composition of narrow-leaved lavender essential oils. Acta Chromatogr. 30(4): 225-230.

Anonymous. 2009. Lavender production. Essential oil crops Production guidelines for lavender, Directorate Agricultural Information Services Department of Agriculture, Forestry and Fisheries Private Bag X144, Pretoria, South Africa. 20.

Ansari, A., J. Razmjoo, and H. Karimmojeni. 2016. Mycorrhizal colonization and seed treatment with salicylic acid to improve physiological traits and tolerance of flaxseed (*Linum usitatissimum L.*) plants grown under drought stress. Acta physiol. Plant. 38(2): 34.

Aslani, Z., A. Hassani, M. Rasouli-Sadaghiani, B. Esmailpour, and Z. Rohi. 2014. Effects of arbuscular mycorrhizal (AM) fungi on essential oil content and nutrients uptake in basil under drought stress. J. Medicinal plants By-product. 3(2): 147-153.

Avcı, A.B. 2010. A new record and distribution of the endemic *Thymus cilicicus* Boiss. & Bal. for the squares B3 and C3 in Turkey. Turkish J. Field Crops. 15(1): 40-42.

Azcon, R., and J.M. Barea. 1997. Mycorrhizal dependency of a representative plant species in Mediterranean shrublands (*Lavandula spica L.*) as a key factor to its use for revegetation strategies in desertification-threatened areas. Appl. Soil Ecol. 7(1): 83-92.

Badri, M.S., A. Hani, and L. Hakimi. 2015. Effect of humic acid on performance and nano iron chelated herbs and essential oil of lavender (*Lavandula angustifolia*) volume (4).

Bajaj, E.D. 1988. Biotechnology in Agriculture and Forestry, *Medicinal and Aromatic plants III*, Springer-Verlag, Berlin. 15: 283-310.

Barea, J.M., R. Azcon, and C. Azcon-Aguilar. 1992. The use of 15N to assess the role of VA mycorrhiza in plant N nutrition and its application to evaluate the role of mycorrhiza in restoring Mediterranean ecosystems. In: Read, D.J., D.H. Lewis, A.H. Fitter, I.J. Alexander, I.J. Mycorrhizas in Ecosystems. Structure and Function. CAB International, Wallingford, UK. 190-197.

Bakry, B. A., T.A. Elewa, M.F. El-Kramany, and A.M. Wali. 2013. Effect of humic and ascorbic acids foliar application on yield and yield components of two wheat cultivars grown under newly reclaimed sandy soil. IJAPP. 4(6): 1125-1133.

Basch, E. I. F., R. Liebowitz, J. Nelson, M. Smith, D. Sollars, and C. Ulbricht. 2004. Monograph from natural standard: lavender (*Lavandula angustifolia* Miller) J. Herb. Pharmacother. 4(2): 63-78.

Bayat, H. and S. Belopukhov. 2019. The effect of humic acid, plant growth promoting rhizobacteria and seaweed on growth parameters, essential oil and chlorophyll content in sweet basil (*Ocimum basilicum L.*). Global Sci. J. *7*(7): 19-32.

Baydar, H. 2009. Lavender. Medicinal and Aromatic Plant Science and Technology (3rd press). Suleyman Demirel University Press No: 51: 274-278.

Bethlenfalvay, G.J., and H. Schiiepp. 1994. Arbuscular mycorrhizas and agrosystem stability. In: Gianinazzi, S., Schiiepp, H. (Eds.), Impact of Arbuscular Mycorrhizas on Sustainable Agriculture and Natural Ecosystems. Birkh~iuser Verlag, Basel, 117-131.

Bettina M. and S. Helge. 2015. The essential oil maker's handbook, IV TP958.M27513, 661: 806.

Boehme, M., and J. Schevtschenko, I. Pinker. 2005. Iron supply of cucumbers in substrate culture with humate. Acta Hort. 41(1): 329-335.

Boghdady M.S., A.H. Dalia, and A.H. Selim. 2016. Influence of foliar spray with seaweed extract on growth, yield and its quality, profile of protein pattern and anatomical structure of chickpea plant (*Cicer arientium* L.,). Middle East. J. Appl. Sci. 6(1): 207–221.

Bohme, M. and H. Thi Lua. 1997. Influence of mineral and organic treatments in the rhizosphere on the growth of tomato plants. Acta Hort. 450: 161-168.

Bowes, K.M., V.D. Zheljazkoz, and C.D. Coldwell. 2004. Infuence of seedling date and harvest stage on yields and essential oil composition of three cultivars of dill (*Antheum graveolens L.)* grown in Nova Scotia. Can. J. plant Sci. 84(4): 1155-1160.

Brundrett, M.C. 1991. Mycorrhizas in natural ecosystems. In: Macfayden, A., Begon, M., Fitter, A.H. (Eds.), Advances in Ecological Research, vol. 21. Academic Press Ltd., London, 21: 171-213.

Brundrett, M.C. 1991. Mycorrhizas in natural ecosystems. In: Macfayden, A., Begon, M., Fitter, A.H. (Eds.), Advances in Ecological Research, vol. 21. Academic Press Ltd., London. 21: 171-213.

Cabello, M., G. Irrazabal, A.M. Bucsinszky, M. Saparrat, and S. Schalamuk. 2005. Effect of an arbuscular mycorrhizal fungus, *Glomus mosseae*, and a rock-phosphatesolubilizing fungus, *Penicillium thomii*, on *Mentha piperita* growth in a soilless medium. J. Basic Microbiol. 45(3): 182-189.

Ceylan, A. 1996. Medical Plants-II (Essential Oil Plants). Ege University Agriculture Facult Publications No: 481, İzmir.

Chaudhary, V., R. Kapoor, A.K. Bhatnagar. 2008. Effectiveness of two arbuscular mycorrhizal fungi on concentrations of essential oil and artemisinin in three accessions of *Artemisia annua L*. Appl. Soil Ecol. 40(1): 174-181.

Chen, J., W. Wang, J. Fang, and K. Varahramyan. 2004. Variable-focusing microlens with microfluidic chip. J. Micromech. Microeng. 14(5): 675.

Chen, Y. and T. Aviad. 1990. Effects of humic substances on plant growth. In Humic Substances in Soil and Crop Science: Selected Readings, Edited by: MacCarthy, P., Clapp, C. E., Malcolm, R. L. and Bloom, P. R. 161-186.

Clark, R.B., R.W. Zobel, and S.K. Zeto. 1999. Effects of mycorrhizal fungus isolates on mineral acquisition by *Panicum virgatum* in acidic soil. Mycorrhiza. 9(3): 167-176.

Copetta, A., G. Lingua, and G.E. Berta. 2006. Effects of three AM fungi on growth, distribution of glandular hairs, and essential oil production in *Ocimum basilicum L*. var. Genovese. Mycorrhiza. 16(7): 485-494.

Dawood, M.G., Y.R. Abdel-Baky, M.E.S. El-Awadi, and G.S. Bakhoum. 2019. Enhancement quality and quantity of faba bean plants grown under sandy soil conditions by nicotinamide and/or humic acid application. Bull. Natl. Res. Cent. 43(1): 1-8.

De Rapper, S., A. Viljoen, and S. Van Vuuren. 2016. The in vitro antimicrobial effects of *Lavandula angustifolia* essential oil in combination with conventional antimicrobial agents. Evid. Based Complement. Altern, Jan 1. Volume 2016 . Article ID 2752739.

Dubey, V.S., R. Bhalla, and R. Luthra. 2003. Sucrose mobilization in relation to essential oil biogenesis during palmarosa (*Cymbopogon martini* Roxb. Wats. Var. *motia*) inflorescence development. J. Biosci. 28(4): 479-487.

El-Hak, S. H. G. and A.M. Ahmed. 2012. Effect of foliar application with two antioxidants and humic acid on growth , yield and yield components of peas (*Pisum sativum L* .). IJHSOP. 4(3): 318-328.

El-Khateeb, M.A., A.B. El-Attar. and R.M. Nour. 2017. Application of plant biostimulants to improve the biological responses and essential oil production of marjoram (*Majorana hortensis,* Moench) plants. Middle East J. Agric. Res. 6(4): 928-941.

El-Kouny, H.M. 2002. Effect of organic fertilizer in different application rates under salinity stress Goudvion on soil and plant. International Symposium and Optimum Resources Utilization in Salt-Affected Ecosystems in Arid and Semi-Arid Regions. 8-11.

El-Sharkawy, G.A. and H.S. Abdel-Razzak. 2010. Response of cabbage plants (*Brassica oleraceae* var. capitata L.) to fertilization with chicken manure, mineral nitrogen fertilizer and humic acid. Alex. Sci. Exch. J, 31, pp.416-432.

Fitter, A.H. 1985. Functioning of vesicular-arbuscular mycorrhizas under field conditions. New Phytol. 99(2): 256-265.

Gaines I, and A. Yilma,1983. Comparison of five humic acids. Fuel. 62(9): 373-379.

Garcia, M. C. V., F. S. Estrella, M. J. Lopez, and J. Moreno. 2008. Influence of compost amendment on soil biological properties and plants. Dynamic Soil. Dynamic Plant. 1: 1-9.

Ghasemi, P.A., M. Momeni, and M. Bahmani. 2013. Ethnobotanical study of medicinal plants used by Kurd tribe in Dehloran and Abdanan districts, Ilam province, Iran. Afr. J. Tradit. Complement. Altern. Med. 10(2): 368-385.

Ghelardini, C., N. Galeotti, G. Salvatore, and G. Mazzanti. 1999. Local anaesthetic activity of the essential oil of *Lavandula angustifolia*. Planta medica. 65(8): 700-703.

Gianinazzi, S., and H. SchUepp, 1994. Impact of Arbuscular Mycorrhizas on Sustainable Agriculture and Natural Ecosystem. Brikh~iuser, Basel.

Gianinazzi-Pearson, V., 1984. Host-fungus specificity, recognition, and compatibility in mycorrhizae. In Genes involved in Microbe-plant interactions (pp. 225-253). Springer, Vienna.

Glenn, L. 2007. Lavender one of the Safest and Most Versatile Essential Oils American Botanical Council.

Golubkina, N., L. Logvinenko, M. Novitsky, S. Zamana, S. Sokolov, A. Molchanova, O. Shevchuk, A. Sekara, A. Tallarita, and G. Caruso. 2020. Yield, essential oil and quality performances of *Artemisia dracunculus*, *Hyssopus officinalis* and *Lavandula angustifolia* as affected by arbuscular mycorrhizal fungi under organic management. Plants. 9(3): 375.

Guenther, E. 1961. The Essential Oils. III, 440-492. Van Nostrand Reinhold, London.

Gupta, M.L., A. Prasad, M. Ram, S. Kumar. 2002. Effect of the vesicular–arbuscular mycorrhizal (VAM) fungus *Glomus fasciculatum* on the essential oil yield related characters and nutrient acquisition in the crops of different cultivars of menthol mint (*Mentha arvensis*) under field conditions. Bioresour. Technol. 81(1): 77-79.

Gutjahr, C. 2014. Phytohormone signaling in arbuscular mycorhiza development. Curr. Opin. Plant Biol. 20: 26-34.

Habte, M., and A. Manjunath. 1991. Categories of vesicular arbuscular mycorrhizal dependency of host species. Mycorrhiza. 1(1): 3-12.

Harper, S.M.; G.L. Kerven; D.G. Edwards and Z. Ostatekboczyski. 2000. Characterization on fulvik and humic acids from leaves of (*Eucalyptus comaldulensis*) and from decomposed hay. Soil Biol. Biochem. 32(10): 1331-1336.

Hart, M.M., R.J. Reader, , J.N. Klironomos. 2003. Plant coexistence mediated by arbuscular mycorrhizal fungi. Trends Ecol. Evol. 18(8): 418-423.

Hendawy, S.F., M.S. Hussein, A.E. El-Gohary and M.E. Ibrahim.2015. Effect of foliar organic fertilization on the growth, yield, and oil content of *Mentha piperita* var. citrate. Asian J. Agric. Res. 9(5): 237-248.

Hilman, Y., and A.A. Asandhi, 1987. Effect of several kinds of foliar fertilizer and plant growth regulator on the growth and yield of garlic *Allium sativum L*. cv. LumbuHijau in the rainy season. Bull. Penelitian Hortikult. 151(2): 267-272.

Jalayerinia, N., S.M.N. Kalat, and R.S. Abadi Haghighi. 2017. The effect of vermicompost and spraying with humic acid and fertilizer on quantitative and qualitative characteristics of rosemary. Asian J. Biol. Life Sci. 6(1).

Jamali, Z. S., A.R. Astaraei and H. Emami, 2015. Effect of humic acid, compost, and phosphorus on growth characteristics of basil herb and concentration of micro elements in plant and soil. J. Sci. and Tech. 6(2): 187-205.

Jianu, C.; G. Pop, A.T. Gruia,; and F.G. Horhat. 2013. Chemical composition and antimicrobial activity of essential oils of lavender (*Lavandula angustifolia*) and lavandin (*Lavandula* x *intermedia*) grown in Western Romania. Int. J. Agric. Biol. 15(4): 772-776.

Juarez, R.C.R., L.E. Craker, M.N.R. Mendoza and J.A. Aguilar-Castillo. 2011. Humi substances and moisture content in the production of biomass and bioactive constituents of *Thymus vulgaris* L. Rev. Fitotec. Mex. 34 (3): 183-188.

Kan, Y., N. Arslan, L. Altun, and M. Eagle. 2006. The economic importance of the culture of medicinal and aromatic plants in Turkey. 15. The meeting of herbal medicines raw materials conference. 4(3): 213-219.

Kanias, G.D., C. Souleles, , A.Loukis, , and E.Panou-Filotheou. 1998. Statistical study of essential oil composition in three cultivated Sage species. J. Essent. Oil Res. 10(4): 395-403.

Kapoor R, B Giri, and KG .Mukerji. 2004. Improved growth and essential oil yield and quality in *Foeniculum vulgare* mill on mycorrhizal inoculation supplemented with P-fertilizer. Bioresour. Technol. 93(3): 307-311.

Kapoor, R., V. Chaudhary, and A.K.E. Bhatnagar. 2007. Effects of arbuscular mycorrhiza and phosphorus application on artemisinin concentration in *Artemisia annua L*. Mycorrhiza. 17(7): 581-587.

Karagiannidis, N., T. Thomidis, and E.E. Panou-Filotheou. 2012. Effects of *Glomus lamellosum* on growth, essential oil production and nutrients uptake in selected medicinal plants. J. Agric. Sci. 4(3): 137-144.

Karagiannidis, N., Thomidis, T., Lazari, D., Panou-Filotheou, E. and C. Karagiannidou,., 2011. Effect of three Greek arbuscular mycorrhizal fungi in improving the growth, nutrient concentration, and production of essential oils of oregano and mint plants. Sci. Hortic. 129(2): 329-334.

Katalinić V., I. Generalić, D. Skroza, I. Ljubenkov, A. Teskera, et al. 2009. Insight in the phenolic composition and antioxidative properties of *Vitis vinifera* leaves extracts. Croat. J. Food Sci. Technol. 1(2): 7-15.

Khanam U., S. Oba, E. Yanase, and Y. Murakami. 2012. Phenolic acids, flavonoids, and total antioxidant capacity of selected leafy vegetables. J. Func. Foods. 4(4): 979-987.

Khaosaad, T., H. Vierheilig, K. Zitterl-Eglseer, and J. Novak. 2006. Arbuscular mycorrhiza alters the concentration of essential oils in oregano (*Origanum sp.*, Lamiaceae). Mycorrhiza. 16(6): 443-446.

Kotsiris G., P.A Nektarios., and A.T. Paraskevopoulou. 2012. *Lavandula angustifolia* growth and physiology is affected by substrate type and depth when grown under Mediterranean semiintensive green roof conditions. Hort. Sci. 47(2): 311-317.

Landeweert, R., E. Hoffland, R.D. Finlay, T.W. Kuyper, and N. Van Breemen. 2001. Linking plants to rock: ectomycorrhizal fungi mobilize nutrients from minerals. Trends Ecol. Evol. 16(5): 248-254.

Landmann C, Fink B, Festner M, Dregus M, Engel K-H, Schwab W.2007.Cloning and functional characterization of three terpene synthases from lavender (Lavandula angustifolia). Arch. Biochem. Biophys. 465(2): 417-429

Lawrence B. M., Essential Oils. 1991–1994. Allured Publishing Corp., Wheaton, IL.

Lawrence, D.H., 1994. The complete poems of DH Lawrence. Wordsworth Editions.

Leonard, A. G. 2008. Humic acid: 100% natural, many uses. Goldend Harvest organic. LLCTM.

Leyval, C., and J. Berthelin. 1993. Rhizodeposition and net release of soluble compounds by pine and beech seedlings inoculated with rhizobacteria and ectomycorrhizal fungi. Biol. Fertil. Soils 15(4): 259-267.

Mahmoud, M. J. 2008. Chemistry of medicinal plants. Anwar Al-Dejla Press. Baghdad, Iraq.

Mahmoudi, R., S. Aghaei, Z. Salehpour, A. Mousavizadeh, S.S. Khoramrooz, M. Taheripour Sisakht, G. Christiansen, M. Baneshi, B. Karimi, and H. Bardania. 2020. Antibacterial and antioxidant properties of phyto-synthesized silver nanoparticles using *Lavandula stoechas* extract. Appl. Organomet .Chem. 34(2): 5394.

Mandal, S., H. Evelin, B. Giri, V.P. Singh, and R. Kapoor. 2013. Arbuscular mycorrhiza enhances the production of stevioside and rebaudioside-A in *Stevia rebaudiana* via nutritional and non-nutritional mechanisms. Appl. Soil Ecol. 72: 187-194.

Mandyam, K., and A. Jumpponen. 2005. Seeking the elusive function of the root-colonising dark septate endophytic fungi. Stud. Mycol. 53: 173-189.

Marschner H. 1995. Mineral nutrition of higher plants. 2nd Edition. Academic Press. 889.

Martinez-Medina, A., A. Roldan, A. Albacete, and J.A. Pascual. 2011. The interaction with arbuscular mycorrhizal fungi or *Trichoderma harzianum* alters the shoot hormonal profile in melon plants. Phytochem. 72(2-3): 223-229.

Massada, Y. 1976. Analysis of essential oil by gas chromatograph and spectrometry, John Wiley, and Sons, New York.

Matysiak B., and A. Nogowska. 2016. Impact of fertilization strategies on the growth of lavender and nitrates leaching to environment. Hort. Sci. (Prague). 43(2): 76-83.

Miransari, M., H.A. Bahrami, F. Rejali, and M.J. Malakouti. 2009. Effects of soil compaction and arbuscular mycorrhiza on corn (*Zea mays L.*) nutrient uptake. Soil till. Res. 103(2): 282-290.

Mohammadipour, E., A. Golchin, J. Mohammadi, N. Negahdar and M. Zarchini .2012. Effect of humic acid on yield and quality of marigold (*Calendula officinalis* L.). Ann. Biolog. Res. 3(11): 5095-5098.

Mohammed, A.E.M.M., A.A.A., Mewead, A.S.H., Gendy, and M.A.I. Abdelkader, M.A.I. 2019. Influence of humic acid rates and application times on vegetative growth and yield components of chamomile (*Matricaria chamomilla, L.*) plants grown under reclaimed sandy soil conditions. Zagazig J. Agric. Res. 46(6): 2171-2181.

Mohammed, M.H.M., A.A. Meawad, E.E.A. M. El-Mogy and M.A.I. Abdelkader. 2019. Growth, yield components and chemical constituents of *Stevia rebaudiana* Bert. as affected by humic acid and NPK fertilization rates. Zagazig J. Agric. Res. 46(1): 1-14.

Moraditochaee, M. 2012. Effects of humic acid foliar spraying and nitrogen fertilizer management on yield of peanut (*Arachis Hypogaea L.*) in Iran. ARPN J. Agric. Biol. Sci. 7(4): 289-293.

Nell, M., C. Wawrosch, S. Steinkellner, H. Vierheilig, B. Kopp, and A. Lossi. 2010. Root colonization by symbiotic Arbuscular Mycorrhizal fungi increases sesquiterpenic acid concentrations in *Valeriana officinalis L*. Planta. Med. 76: 393-398.

Newsham, K.K. 2011. A meta-analysis of plant responses to dark septate root endo-phytes. New Phytol. 190(3): 783-793.

Nikbakht, A., M. Kafi, M. Babalar, Y.P. Xia, A. Luo, and N.A. Etemadi. 2008. Effect of humic acid on plant growth, nutrient uptake, and postharvest life of gerbera. J. Plant Nutr. 31(12): 2155-2167.

Noroozisharaf, A. and M. Kaviani. 2018. Effect of soil application of humic acid on nutrients uptake, essential oil and chemical compositions of garden thyme (*Thymus vulgaris L.*) under greenhouse conditions. Physiol. Mol. Biol. Plants. 24(3): 423-431.

Novak, J., B. Lukas, , K.Bolzer, , S.A.B.I.N.E. Grausgruber-Gröger, and J. Degenhardt. 2008. Identification and characterization of simple sequence repeat markers from a glandular *Origanum vulgare* expressed sequence tag. Molecular Ecology Resources, 8(3), pp.599-601.

Ortas, I. and C. Akpınar. 2006. Response of kidney bean to arbuscular mycorrhizal inoculation and mycorrhizal dependency in P and Zn deficient soils. Acta. Agr. Sc. and. B-Soil Plant Sci. 56(2): 101-109.

Ouahmane, L., M. Hafidi, C. Plenchette, M. Kisa, A. Boumezzough, J. Thioulouse, and R. Duponnois. 2006. *Lavandula* species as accompanying plants in *Cupressus replanting* strategies: effect on plant growth, mycorrhizal soil infectivity and soil microbial catabolic diversity. Appl. Soil Ecol. 34(2-3): 190-199.

Pinto, J.G., E.L. Fröhlich, G.C. Leckebusch, and U. Ulbrich. 2007. Changing European storm loss potentials under modified climate conditions according to ensemble simulations of the ECHAM5/MPI-OM1 GCM. Nat. Hazards Earth Syst. Sci. 7(1): 165-175.

Podila, G.K. and D.D. Douds. 2000. Current advances in mycorrhizae research. APS press.

Razic, S., A. Onja, S. Dogo, C.L. Slavkovi. And C.A. Popovi. 2005. Determination of metal content in some herbal drugs – empirical and chemometric approach. Talanta. 67(1): 233-239.

Reverchon, E., G.D. Porta, and F.Senatore. 1995. Supercritical CO_2 extraction and fractionation of lavender essential oil and waxes. Journal of Agricultural and Food Chemistry, 43(6), pp.1654-1658.

Said-AlAhl , H.A., A.G. El-Gendy and E.A. Omer. 2016. Humic acid and indole acetic acid affect yield and essential oil of dill grown under two different locations in Egypt. Int. J. Pharm. Pharmaceutical Sci. 8(8): 146-157.

Sanchez -Sanchez, A., J. SanchezAndreu, M. Juarez, J. Jorda, and D. Bermudez. 2002. Humic substances and amino acids improve effectiveness of chelate FeEDDHA in lemon trees. J. Plant Nutr. 25(11): 2433-2442.

Sardashti, A., A. Ganjali, and A. Kordi. 2012. Effect of humic substances on the quality of essential oils of medicinal plants. J. Med. Plant Res. 6(13): 2644-2654.

Sardashti, A., A. Ganjali, and A. Kordi. 2012. Effect of humic substances on the quality of essential oils of medicinal plants. J. Med. Plant Res. 6(13): 2644-2654.

Schnitzer, E.A. and F. Wightman .1974. Metabolism of auxin in higher plants. Ann. Rev. Plant Physiol., 25: 487-513.

Seidler-Ło, K., K. Zykowska, W.A. Kucharski, and R. Mordalski. 2005. Ekologiczna uprawa ro´slin zielarskich. Rolnictwo ekologiczne. Instytut Ro´slin i Przetworów Zielars-kich, Pozna´ n.

Sharif, M., R.A. Khattak, and M.S. Sarir. 2002. Effect of different levels of lignitic coal derived humic acid on growth of maize plants. Comm. Soil Sci. Plant Anal. 33(19-20): 3567-3580.

Sharopov, F.S., H. Zhang, M. Wink, and W.N. Setzer. 2015.Aromatic medicinal plants from Tajikistan (Central Asia). *Medicines*, *2*(1), pp.28-46.

Shawl, A. S. T. Kumar, S. Shabir, N. Chishti, and Z. A. Kaloo. 2005. Lavender – a versatile industrial crop in Kashmir. Indian Perfumer, 49(2): 235-238.

Singh, M., and K. Wasnik. 2013. Effect of vermicompost and chemical fertilizer on growth, herb, oil yield, nutrient uptake, soil fertility, and oil quality of Rosemary. Comm. Soil Sci. Plant Anal. 44(18): 2691-2700.

Srinivasan, K. 2007. Black pepper and its pungent principle-piperine:a review of diverse physiological effects. Critical Rev. Food Sci. Nutr. 47(8): 735-748.

Swaminathan K, and B.C. Verma. 1979. Responses of three crop species to vesicular arbuscular mycorrhizal infection on zinc-deficient Indian soils. New Phytol. 82(2): 481-487.

Tadayyon, A. S. Beheshti and M. Pessarakli. 2017. Effects of sprayed humic acid, iron and zinc on quantitative and qualitative characteristics of Niger plant (*Guizotia abyssinica L.*). J. Plant Nutr. 40(11): 1644-1650.Tan, K.H. 2011. Principles of soil chemistry. 4th ed. New York (USA): Marcel Dekker.

Tian, C.Y., G. Feng, X.L. Li, and F.S. Zhang. 2004. Different effects of arbuscular mycorrhizal fungal isolates from saline or non-saline soil on salinity tolerance of plants. Appl. Soil Ecol. 26(2): 143-148.

Vafaei, N., S. Samavat and A.R. Ladan. 2015. Effects of different levels of humic acid, amino acid and fulvic acid on the growth and yield of basil plants Intern. Res. J. Appl. Basic Sci. 9(10): 1732-1734.

Van der Hejden, M.G.A., J.N. Klironomos, M. Ursic, P. Moutoglis, R. Streitwolf-Engel, T. Boller, A. Wiemken, and I.R. Sanders. 1998. Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. Nature. 396(6706): 69-72.

Varanini, Z., R. Pinton. 1995. Humic substances and plant nutrition, progress in botany. Springer, Berlin. 97-117.

Verma, R.S., L.U. Rahman, C.S. Chanotiya, R.K. Verma, A. Chauhan, A. Yadav, A. Singh, and A.K. Yadav. 2010. Essential oil composition of *Lavandula angustifolia* Mill. cultivated in the mid hills of Uttarakhand, India. J. Serb. Chem. Soc. 75(3): 343-348.

Vessey, K.J. 2003. Plant growth promoting rhizobacteria as biofertilizers. Plant Soil. 255: 571-586.

Wu, Q.S., and Y.N. Zou. 2010. Beneficial roles of arbuscular mycorrhizas in citrus seedlings at temperature stress. Sci. Hortic. 125(3): 289-293.

Yousif, K.H. 2018. Effect of humic acid and seaweeds extracts on growth, yield and nutrient content of garlic (*Allium sativum* L.). J. Univ. Duhok. 21(1): 8-18.

Zahra, M.K.M., S.I. Monib, S. Abdel, and A. Heggo. 1984. Significance of soil inoculation with silicate bacteria. Zentralblatt Fur Mikrobiologie .139(5): 349-357.

Zhang, X., E.H. Ervin, and R.E. Schmidt. 2003. Plant growth regulators can enhance the recovery of Kentucky bluegrass sod from heat injury. Crop Sci. 43(3): 952-956.

Zubek, S., A.M. Stefanowicz, J. Błaszkowski, M. Niklińska, and K. Seidler-Łożykowska. 2012. Arbuscular mycorrhizal fungi and soil microbial communities under contrasting fertilization of three medicinal plants. Appl. Soil Ecol. 59: 106-115.

CHAPTER V

DIAMMONIUM PHOSPHATE AND POTASH AFFECT GROWTH, FLOWERING AND ESSENTIAL OILS CONTENT OF LAVENDER

Abstract

Production of aromatic plants is subject to cultural practices such as management, irrigation, nutrition, and soil fertility, in addition to external factors which may affect the chemical composition of the secondary metabolites. The aim of this study is to evaluate the effects of diammonium phosphate (DAP) and potash fertilizers on plant growth and essential oils content in three varieties of lavender. An experiment was conducted in a greenhouse at during Spring 2020 to study the effect of DAP fertilizer (0, 1 g, or 2 g/L) and potash fertilizer (0 or 2 g/L) on the growth and essential oils composition of lavender (*Lavandula angustifolia* 'Hidcote', *L. angustifolia* 'Platinum Blonde', or *L. intermedia "province*). DAP fertilizer increased plant height, branch number, flower length, essential oil content, and essential oil components. Potash at 2g/L increased growth but decreased quantitative and qualitative essential oil production. Essential oil extracts were analyzed by high performance liquid chromatography (HPLC) to determine the composition of the volatile fraction, the base oil compounds, and the volatiles selected.

Keywords: *Lavandula angustifolia* 'Hidcote', *L. angustifolia* 'Platinum Blonde', or *L. intermedia "provence*, fertilizer, HPLC

Introduction

Lavender *(Lavandula spp.)* is an evergreen perennial shrub which can survive for 10-15 years. Lavender is grown for its flowers or inflorescences, from which the essential oils is obtained by distillation (Biesiada et al., 2008; Crummitt, Keenen, and Dan Drost, 2012). All *Lavandula* (Lamiaceae) species and hybrids are aromatic plants which produce essential oils in a complex formation of glands on the surface of flowers and leaves (Lis-Balchin, 2002). Lavender should be pruned every year after flowering to stimulate new growth and a more vibrant plant. This will lead to a longer life span and improved productivity (Crummitt, Keenen, and Dan Drost, 2012).

Recently, interest in medicinal and aromatic plants has increased in both industry and scientific research due to their strong pharmacological properties, in addition to the fact they contain many natural antioxidants, vitamins, carotenoids, chlorophyll, phytoestrogens, and minerals (Parejo et al., 2002). Lavender and its essential oils are used in alternative medicine and aromatherapy (Woronuk et al., 2011). Linalyl acetate (25-46%) and linalool (20-45%) are the main components of lavender oil, which have soothing, carminative, and antimicrobial properties attributed to them (Biesiada et al., 2008). Due to the many uses of lavender oil, it is an important part of the essential oils industry, and its share is increasing in the global herbal market (Touati et al., 2011). The annual world production of lavender oil is about 200 tons (Curtis, 2005).

Lavender does not require a lot of fertilizer so, it grows well in soil not suitable for most other crops. However, to obtain a high yield of foliage and quality essential oils, fertilization is

important. Optimum mineral fertilization is essential to the successful production of lavender (Klados and Tzortzakis, 2014). The type of fertilizer and dosage used greatly affects the quality of the oil extracted from the plant. Plant fertilization and mineral absorption/accumulation are the most important factors to increase plant production. Nitrogen (N), phosphorous (P) and potassium (K) affect plant growth and synthesis of essential oils in medicinal plants (Sell, 2003).

Of all the essential nutrients, nitrogen, phosphorous, and potassium have the greatest influence on lavender growth and formation of essential oils. Fertilization has a positive effect on the function and level of enzymes involved in terpene biosynthesis (Hafsi et al., 2014). Nitrogen has been reported to increase essential oil yield in thyme (*Thymus vulgaris L.*) (Baranauskienne et al., 2003) and in cumin (*Cuminum cyminum*) (Azizi and Kahrizi, 2008). Diammonium phosphate (DAP) [(NH_4) $2HPO_4$)] is a water-soluble ammonium phosphate salt produced when ammonia reacts with phosphoric acid (John, 1958). Diammonium phosphate is the most widely used phosphate fertilizer (Robertsa, 2014). It provides phosphorus necessary for all living organisms, as nuclear proteins form the basic materials for the cell and allows the cell to divide and develop meristematic tissues.

Potash is produced in large quantities worldwide (K_2O) by electrolysis of potassium hydroxide (Knight, 1992). It is also obtained from sedimentary rocks. The total potassium content in different potash fertilizers is expressed in equivalent weight (by weight) % of K_2O. (Anderson, 1985; Rawashda and Maxwell, 2014). Potassium is the monovalent cation used by all living cells and is found in high concentrations in plants. The plant needs a large amount of potassium fertilizer for healthy growth, as well as to increase crop production. Due to the lack of

studies on the effects of fertilizers on the essential oils of lavender plants, this study was conducted.

The aim of this research was to study the effect of DAP and potash fertilizer on the vegetative growth and essential oil yield of lavender. This study investigated the effects of nitrogen and phosphorous, and potassium on growth characteristics, and the quality and quantity of the essential oil of three varieties of lavender. High-Performance Liquid Chromatography (HPLC) was used to determine the composition of volatile substances, including linalool, in lavender essential oil.

Material and methods

Lavender cuttings were obtained for three cultivars, *Lavandula angustifolia* 'Platinum Blonde', *L. angustifolia* 'Hidcote', and *L.* x *intermedia* 'Provence' (Emerald Coast Growers, Inc., Pensacola, FL). The experiment was conducted summer 2020. The cuttings were planted and grown in 15 cm (1 L) containers in a peat-based substrate (Pro Mix BX, Premier Tech Horticulture, Quakertown, PA) until when their height was 15 cm before starting treatments. No pesticides were used in the study area throughout the trial period. The experiment was carried out in a Randomized Complete Block Design (RCBD) with 3 replications. The following treatments were applied: DAP (Ferti-lome; Voluntary Purchasing Groups, Inc., Bonham, TX) at 0, 1, or 2, g/L and foliar application of potash at 0 or 2 g/L. Plants were grown in Mississippi State University's Department of Plant and Soil Sciences greenhouses (latitude 27-33 ° N; longitude 88-47 ° W) with the temperature set points at 24/20° C Day/Night. Plants were fertilized twice weekly with 2-5-1 organic fertilizer (Drammatic "O", Dramm Corp., Manitowoc, WI).

Plants were grown for 8 weeks before the plants and flowers were harvested at the soil line in August 2020. Vegetative growth was recorded at harvest for all plants with the following assessments made: Plant height (cm) was measured from the soil to the growing top, number of branches was counted as the number of branches of each plant greater than 5 cm, and number of florets on each plant at harvest. Harvested plants were placed in 16x25x5 cm unbleached paper food trays (500 kraft food trays, Specialty Quality Packaging, Scotia, NY) and dried at room temperature (20°C). Samples were turned daily until dried to a continuous weight. After one month the plants were totally dry without any molding problems. Dried samples were weighed to determine the total plant dry weight (Dw-g) (buds, flowers, leaves, and stems) and stored in paper bags (Bettina and Helge, 2015).

Essential oils were extracted from the air-dried samples using a Soxhlet extraction apparatus (Reverchon and Della, 1995). Samples were prepared by placing 5 gm of dried, finely ground leaves and flowers (Spice and Nut Grinder MODEL SG-10 Cuisinart, Stanford, CT in a 33 x 80 mm paper thimble filter. 150 mL ethyl alcohol was placed in the boiling chamber and the samples were extracted over 9 hours. After the extraction, the samples were filtered to remove plant tissues deposited at the bottom of the flask. Then the samples were transferred to the oven at 68°C to evaporate the ethanol from the extracted oil leaving the extracted oil (about 2 to 3 mL) in the glass flask. After that, the total weight of the oil was taken. Oils were kept in glass bottles at room temperature until HPLC analysis. The chemical components of lavender oil were determined by HPLC (Fig. 5.1). Treatment samples were compared to pure standards for linalool and linalyl.

Analysis of Linalool and linalyl acetate

Each oil sample was dissolved in methanol, and the dissolved samples were analyzed by 1100 Agilent High Performance Liquid Chromatography (HPLC) equipped with ZORBAX eclipse plus a C18 (5 µm, 4.6×150 mm) column connected to a UV detector (Fig. 5.1). The HPLC tests were ran using an autosampler with the flow rate set to 1 mL/min with UV detection at 210 nm. The column temperature remained constant at RT, and the mobile phase was 70% acetonitrile–water. 10 µL was injected for 60 minutes with an interval of 1 minute between samples, where only the mobile phase was injected. The concentrations of linalool and linalyl acetate were calculated based on the area of each peak in the chromatograph. The peak positions of linalool and linalyl acetate were detected based on the injection of prepared standards of linalool and linalyl acetate. The concentrations of the separated compounds were calculated by the following:

Concentration (mg/kg) = standard (ng) x sample peak area x total sample volume (ml).

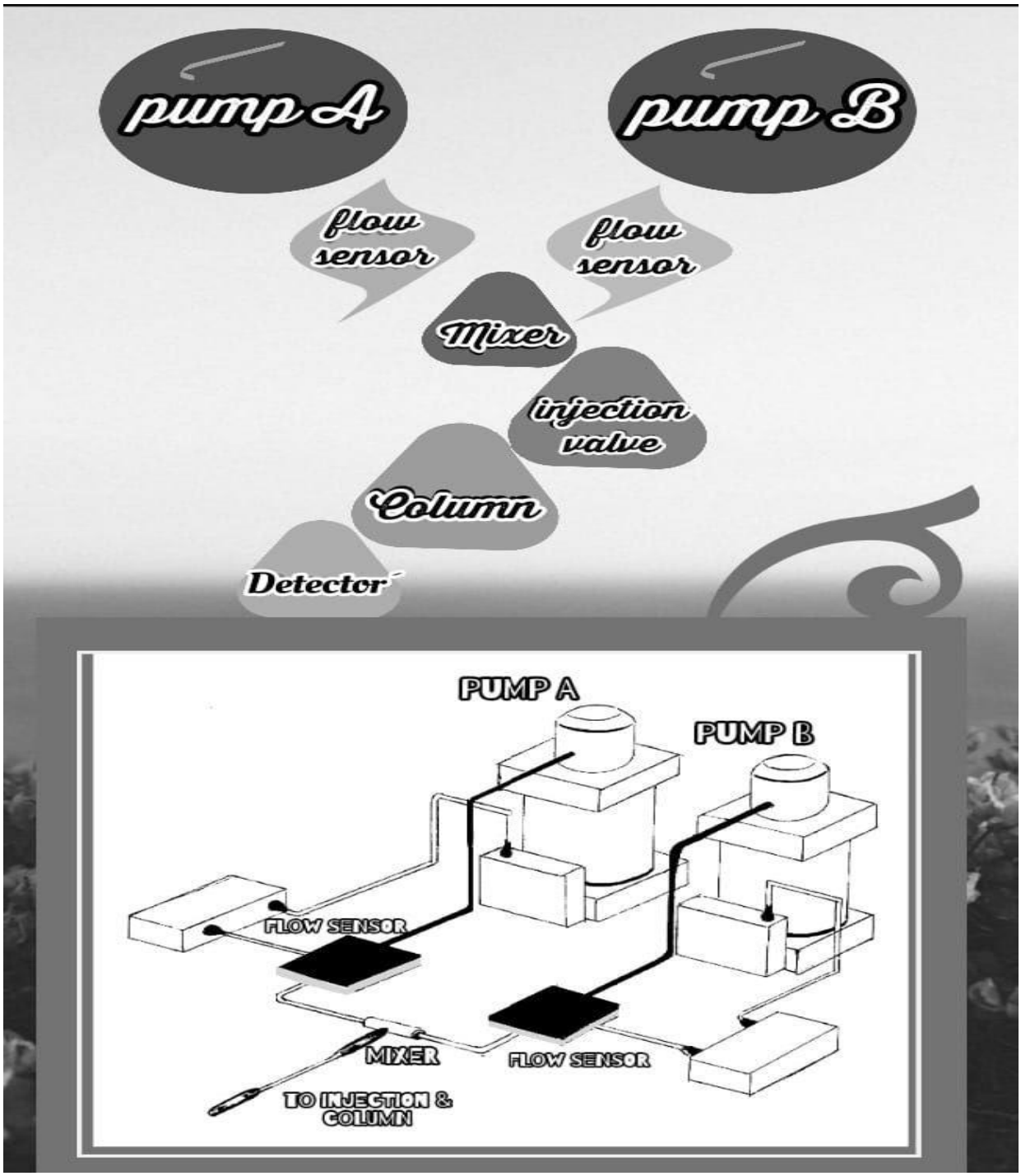

Figure 5.1 Schematic diagram of the HPLC (Credit AL-Garallaa).

Statistical analysis

A Randomized Complete Block Design (RCBD) was used in a factorial experiment including the factors: DAP, potash, and lavender varieties. Statistical software (SAS® ver. 9.4, SAS Institute, Cary, NC) was used for statistical analysis. Data were subjected to analysis of variance (ANOVA). The treatment means were compared using the least significant difference (LSD) at p $\alpha = 0.05$ (Maher, 2019).

Statistical analysis for HPLC

Three replications of each treatment were analyzed for statistical analysis to estimate the active compounds. For estimating the significance of the statistical differences, the mean of the control parameters was used as a constant for the purpose of the assay using the T test of one sample at a probability level of 0.05. Along with the use of the Statistical Package for the Social Sciences (SPSS) version 26 to carry out these analyses (Daniel, 1974).

Results and discussion

Effect of DAP fertilization and potash on plant growth in three lavender varieties.

Plant height (cm)

DAP and Potash fertilizers increased lavender plant height compared to control during both seasons (Table 5.1). The variety Provence was taller than the varieties Hidcote and Platinum Blonde. Potash at 2 g/L led to an increase in plant height compared to the control. DAP at 2 g/L had greater plant height than at 0 g/L, but it did not differ from 1 g/L. In the two-way interaction between the variety and potash, 'Provence' with 2 g potash/L was taller than the rest of the treatments. The shortest plants were 'Platinum Blonde' at 0 g potash/L. In the two-way

interaction between DAP and variety, the tallest plants were from the interactions between 'Provence' and 2 g DAP/L and 'Provence' and 1 g DAP/L. In the two-way interaction between DAP and potash, the tallest plants were in the treatment 2 g DAP/L and 2 g potash/L while the shortest plants were in the treatment 0 g DAP/L and 0 g potash/L. In the three-way interaction, plant height was greatest in the treatment 'Provence' with 2 g DAP/L and 2 g potash/L. The least plant height was in 'Platinum Blonde' with 0 g DAP/L and 0 g potash/L.

Table 5.1 Effect of DAP fertilization and potash on plant height (cm) in three species of lavender.

Lavender Species	Potash g L^{-1}	DAP g L^{-1}			SpeciesX Potash	Species Means
		0	1	2		
'Provence'	2	64.0	64.5	73.7	67.4	57.8
	0	34.2	53.3	57.2	48.2	
'Hidcote'	2	41.7	42.2	42.8	42.2	57.8
	0	40.2	41.3	40.5	40.7	
'Platinum Blonde'	2	21.7	22.8	23.5	22.7	57.8
	0	20.2	21.7	25.0	22.2	
DAP X Species	'Provence'	49.1	58.9	65.4	Potash Means	
	'Hidcote'	40.9	41.8	41.67		
	'Platinum Blonde'	20.9	22.2	24.3		
DAP X Potash	2	42.4	43.2	46.7	44.1	
	0	31.5	38.8	41.7	37.1	
DAP Means		37.0	41.0	43.8		
LSD (0.05)		2.93				

Nitrogen application at different rates significantly affected the plant height rate of lavender. Elshorbagy et al. (2020) found that fertilizer levels of nitrogen, phosphorous and potassium (NPK) increased lavender plant height. Moreover, plant height increase was gradual by increasing the level of NPK fertilization. Biesiada and Kucharska (2008) also indicated an impact of nitrogen dose on the growth and development of lavender, as the plants fertilized with nitrogen at a rate of 200 kg N ha^{-1} were taller and wider than those planted in plots with a total nitrogen dose of 50-100 kg N ha. Karik et al. (2017) found plant height of lavender can be affected by environmental factors and genetic potential. Ceylan et al. (1996) and Arabaci and

Bayram (2005) confirmed that plant height evolves according to taxa and environmental factors. The results of this study agree with Westervelt's (2003) findings that the height of the rosemary plant (*Rosmarinus officinalis*), which belongs to the same family as lavender, increases with increasing nitrogen fertilization.

Number of branches/plants

The greatest branch number for each variety of lavender was observed with the highest level of DAP (Table 5.2). 'Hidcote' produced more branches than 'Platinum Blonde' or 'Provence'. Adding potash fertilizer increased the number of branches per plant compared to no additional potash. Fertilization with DAP at 2 g/L increased the number of branches more than 1 or 0g/L. In the two-way interaction between varieties and potash, 'Hidcote' treated with 2 g potash /L had the greatest number of branches. 'Provence' without adding potash resulted in the fewest branches. In the two-way interaction between DAP and varieties, the greatest number of branches was from the interaction between 'Hidcote' and 2g DAP/L, which had more branches than most other treatments. 'Provence' with 0 g DAP /L produced the least number of branches. In the two-way interaction between DAP and potash, the greatest number of branches was in plants receiving 2 g DAP/L and 2 g potash/L. In the three-way interaction, 'Hidcote' receiving 2g DAP/L and 2 g potash/L had the greatest number of branches. 'Provence' at 0 g DAP/L and 0 g potash/L had the fewest branches.

Table 5.2 Effect of DAP fertilization and potash on number of branches/plants in three species of lavender.

Lavender Species	Potash g L^{-1}	DAP g L^{-1}			Species X Potash	Species Means
		1	2	3		
'Provence'	2	16.3	37.0	36.7	30.0	22.9
	0	8.7	14.0	25.0	15.9	
'Hidcote'	2	28.3	38.3	44.7	37.1	33.6
	0	23.3	33.3	34.0	30.2	
'Platinum Blonde'	2	27.3	23.0	29.0	26.4	25.2
	0	24.7	20.7	26.7	24.0	
DAP X Species	'Provence'	12.5	25.5	30.8	Potash Means	
	'Hidcote'	25.8	35.8	39.3		
	'Platinum Blonde'	26.0	21.8	27.8		
DAP X Potash	2	24.0	32.8	36.8	31.2	
	0	18.9	22.7	28.6	23.34	
DAP Means		21.4	27.7	32.7		
LSD (0.05)		0.79				

Nitrogen promotes abundant vegetative growth through its effect on photosynthesis and protoplasmic construction (Kelly and Thomson, 1957). The increase in the number of main branches of the plant may be the additional fertilizer, nitrogen, phosphorus, and potash, stimulating new growth from lateral shoots (Krideum and Leopold, 1975). The increased levels of phosphorus may also enhance carbohydrate production from the process of photosynthesis to help in the formation and division of cells and the formation of amino acids and proteins (Devasagayam and Jayapaul, 1997). Nitrogen and phosphorous also assist in producing zeatin which stimulates cell division and branching (Thimann et al., 1971). These results are in accordance with those of Elshorbagy (2020) which showed in general, all fertilization levels

increased the number of branches in lavender by 40 to 44%. Potassium has an especially important role in improving plant growth and metabolism contributing to the survival of plants subject to various abiotic stresses. It is an essential nutrient and is also the most abundant cation in plants. In addition, it has essential roles in enzyme activation, protein synthesis, photosynthesis, osmoregulation, stomatal movement, energy transfer, phloem transport, cation anion balance, and stress resistance (Marschner, 2012; Pettigrew, 2008; Shabala and Pottosin, 2010). The availability of potassium at the branching and flowering stages of growth is necessary to obtain good crop yields (Tisdale et al., 1985; Fuentes et al., 2008). Yasemin et. al. (2017) indicated lavender plant growth is especially affected by available nitrogen. Biesiada et al. (2008) confirmed the vegetative growth of *L. angustifolia* was enhanced by nitrogen applications at 50 to 200 kg N/ha, while stating the most appropriate use of nitrogen was an average nitrogen level of 100 kg N/ha. The results of the study mirror those found by Cheang et al. (1987) in mint where branch number increased as the levels of nitrogen fertilization increased.

Number of flowers/ plants

'Hidcote' produced more florets than 'Provence' while 'Platinum Blonde' did not flower (Table 5.3). The addition of potash at 2 g/L increased florets number per plant as did the addition of 2 g DAP /L. In the two-way interaction between variety and potash, it was found 'Hidcote' with or without potash had more florets than the other treatments. In the two-way interaction between DAP and variety, the greatest number of florets was with 'Hidcote' and 2 g DAP /L. In the two-way interaction between DAP and potash, the highest levels of fertilization, 2g DAP /L and 2 g potash/L, resulted in the greatest number of florets per plant while the lowest number of flowers (64.89 flowers) was from the treatment adding the least amount of additional fertilizer, 0g DAP/L and 0 g potash/L. In the three-way interaction, 'Hidcote' treated with 2 g DAP/L and

2 g potash/L increased florets numbers more than in other treatments. ‘Platinum Blonde’ did not flower in this study.

Table 5.3 Effect of DAP fertilization and potash on number of flowers/plants in three species of lavender.

Lavender Species	**Potash g L^{-1}**	**DAP g L^{-1}**			**Species X Potash**	**Species Means**
		1	2	3		
‘Provence’	2	102.7	110.0	322.7	178.4	127.1
	0	35.7	63.3	128.7	75.9	
‘Hidcote’	2	179.0	320.7	587.3	362.3	323.8
	0	159.0	276.0	420.7	285.2	
‘Platinum Blonde’	2	0.0	0.0	0.0	0.0	
	0	0.0	0.0	0.0	0.0	
DAP X Species	‘Provence’	69.1	86.7	225.7	Potash Means	
	‘Hidcote’	169.0	298.3	504.0		
	‘Platinum Blonde’	0.00	0.00	0.00		
DAP X Potash	2	93.9	143.6	303.3	180.3	
	0	64.9	113.1	183.1	120.4	
DAP Means		79.3	128.3	243.2		
LSD (0.05)		27.33				

The increase in flower number may be related to the increased vegetative growth and the number of branches per plant. Since the flowers are terminal, the increased branch number in ‘Hidcote’ would be expected to also show increased flower numbers (Hamman et al, 1996). These results agree with Biesiada and Kucharska (2008) where the rate of nitrogen increased the lavender flowers. Chrysargyris et al. (2016) stated medium phosphorous levels were more growth promotive than low or high phosphorous levels in lavender. Ma et al. (2001) reported

limited levels of phosphorous slowed down the development of floral structures of lavender plants. According to Erbas et. al. (2017), an increase in phosphorus improved such characteristics as the flower length and the number of florets in lavender plants. Previous experiments on the effects of nitrogen and phosphorous levels on lavender revealed the necessity of examining an optimal potassium application (Chrysargyris et al., 2016). Potassium shortage during plant growth can lead to a decrease in chlorophyll content and photosynthetic rate (Gerardeaux et al., 2010), enzymes activation, poor growth, and reduced yield (Kanai et al., 2007). Sanghamitra et al. (2015) indicated high levels of potassium improved the growth and production of *Tagetes erecta* flowers. Silva et al. (2017) also found that using DAP improved flower yield for *L. dentata*.

Dry weight/plant (g)

'Platinum Blonde' had a greater dry weight than 'Provence' which was heavier than 'Hidcote' (Table 5.4). Adding potash at 2 g/L did not affect plant dry weight. The addition of DAP fertilizer at 1 and 2 g/L increased plant dry weight more than 0 g DAP/L. In the two-way interaction between variety and potash, 'Platinum Blonde' treated with 2 g potash/L had the greatest dry weight while the least dry weight was in the treatment 'Hidcote' and 0 g potash/L. In the two-way interaction between DAP and variety, the greatest dry weight was in the treatments 'Platinum Blonde' and 1or 2 g DAP/L and 'Provence' at 2 g DAP/L. 'Hidcote' treated with 0 g DAP/L had the least dry weight. In the two-way interaction between DAP and potash, the highest rates of DAP and potash, 2g/L of DAP and 2 g potash, resulted in the greatest dry weight while the least dry weight was in the treatment with the least fertilizer, 0 g DAP/L and 0 g potash/L. In the three-way interaction, 'Platinum Blonde' treated with 2 g DAP/L and 2 g potash/L had greater dry weight than any of the DAP and potash treatments for 'Provence' and

'Hidcote' but was like 1 g DAP/L and 2 g potash/L. 'Hidcote' at the lowest DAP and potash treatments had the least dry weight.

Table 5.4 Effect of DAP fertilization and potash on plant dry weight in three species of lavender.

Lavender Species	**Potash g L^{-1}**	**DAP g L^{-1}**			**Species X Potash**	**Species Means**
		1	2	3		
'Provence'	2	14.4	17.8	19.8	17.4	17.0
	0	12.89	17.8	19.2	16.6	
'Hidcote'	2	9.6	12.2	13.1	11.6	11.0
	0	7.8	11.1	11.9	10.3	
'Platinum Blonde'	2	15.6	20.8	24.0	20.1	20.0
	0	15.7	21.8	19.6	19.2	
DAP X Species	'Provence'	13.7	17.8	19.5	Potash Means	
	'Hidcote'	8.7	11. 7	12.5		
	'Platinum Blonde'	15.6	21.3	21.8		
DAP X Potash	2	13.2	17.0	18.9	16.4	
	0	12.1	17.0	16.9	15.3	
DAP Means		12.7	17.0	16.92		
LSD (0.05)		1.08				

Nitrogen has an important role in the structure of living plants and comprises 1.5-2.0% of plant dry matter (Huston and Pinchak, 1991). Kokate and Varme(1971) and Van Ginkel (1972) found adding nitrogen fertilizer to lemongrass plants at rates of 80-100 kg/ha led to a 60% increase in dry weight. Growing in a greenhouse may lead to greater growth and higher production of lavender (Morais, 2009). The soil conditions, precipitation and light period are controlled which may lead to a change in the yield and chemical composition of the essential

oils. Biesiada et al. (2008) found that providing 100 kg N/ha in the form of ammonium nitrate increased inflorescence fresh and dry weight in *L. angustifolia*. A potassium deficiency during plant growth leads to a decrease in chlorophyll content and photosynthetic rate (Gerardeaux et al., 2010). Chrysargyris et. al. (2016) reported that the application of 50 mg/L P increased weight and dry matter content. Phosphorous affects many metabolic events such as chlorophyll, protein, carbohydrates, and oil formulations, which contributes to an increase in yield (Rathore et al., 1985; Ramezani et al., 2009).

Oil Yield /Plant

'Platinum Blonde' produced more essential oil than 'Hidcote' or 'Provence' (Table 5.5). Adding potash fertilizer at 2 g/L decreased the oil content from the 0 g potash treatment. Oil dry weight increased as DAP fertilizer concentration increased. In the two-way interaction of variety and potash, 'Platinum Blonde' without the addition of potash produced more oil than the other treatments while the least oil was produced in 'Provence' receiving 2 g potash/L. In the two-way interaction between DAP and variety, greatest oil yield was from 'Platinum Blonde' regardless the DAP treatment and 'Hidcote' and 'Provence' at 2 g DAP/L. The least oil yield was from 'Provence' and 'Hidcote' receiving 0g DAP/L. In the two-way interaction between DAP and potash, the greatest oil yield was from plants receiving the highest DAP and lowest potash treatments, 2g DAP/L and 0 g potash/L. As potash increased and DAP decreased, oil yield decreased. In the three-way interaction, as DAP decreased, and potash increased oil yield decreased in every variety.

Table 5.5 Effect of DAP fertilization and potash on oil yield in three species of lavender.

Lavender Species	Potash g L^{-1}	DAP g L^{-1}			Species X Potash	Species Means
		1	2	3		
'Provence'	2	20.1	22.4	23.3	21.9	23.5
	0	22.7	24.3	28.2	25.0	
'Hidcote'	2	21.8	22.9	24.3	23.0	23.7
	0	22.2	24.3	26.7	24.4	
'Platinum Blonde'	2	22.5	25.3	23.5	23.8	25.4
	0	25.6	25.7	29.5	26.9	
DAP X Species	'Provence'	21.4	23.3	25.7	Potash Means	
	'Hidcote'	22.0	23.6	25.5		
	'Platinum Blonde'	24.1	25.5	26.5		
DAP X Potash	2	21.5	23.5	23.7	22.9	
	0	23.5	24.8	28.1	25.5	
DAP Means		22.5	24.2	25.9		
LSD (0.05)		0.93				

Nitrogen fertilizer increases vegetative growth of plants, increases flowering, and increases the concentration of secondary metabolites. Essential oils are secondary metabolic compounds that increase synthesis with the increase and accumulation of carbohydrates (Dickes and Nicholas, 1978). Also, nitrogen is included in the composition of many compounds found in essential oils (Duhan et al., 1975; Chang et al., 1987). Corrêa et al. (2010) explained the accumulation of secondary metabolites is directly related to the plant genotype and indirectly related to fertilization (types of fertilizer and rates) and environmental variables such as light, temperature, and photoperiod. High-quality lavender oils used in perfumery should contain high levels of linalool and linalyl acetate. High levels of camphor reduce the quality of the product

(Adam, 2006). Fertilization with phosphorous in some species of Apiaceae resulted in an increase in total carbohydrates, total dissolved sugar, total oil, and vegetative productivity of plants (Khaled, 2012). Similar results were found in chamomile (Naderidarbaghshahi et al., 2011), sweet basil (Sharafzadeh et al., 2011), mint (Alsafar and Al.-Hassan (2009), and sage (Lu et al., 2004). Phosphorous plays an important role in the formation of aromatic compounds because it is the building block of the phosphoinol pyruvate (PEP) molecule (Qadry, 2010). Hussain et al. (1996) stated essential oil content increased with the addition of foliar applied phosphorous in lavender. Lis-Balchin (2002) noted that monoterpenes are the main group of essential oils of lavender, followed by the hydrocarbon group. Turbines with lower molecular weights result from the condensation of isopentenyl diphosphate (IPP, C5) and dimethyl allyl diphosphate (DMAPP, C5), the universal terpene precursor. While monoterpenes are generally synthesized from geranyl diphosphate (GPP), sesquiterpenes are synthesized from farnesyl diphosphate (FPP) (Woronuk et al., 2011). Khaled (2013) stated that potassium is necessary to activate the essential oil enzyme; however, in the current study, addition of potassium reduced oil content. Ramzani et al. (2009) stressed that phosphorous use significantly increased the basil essential oil content, but the fresh and dry weight of the herb was not affected. Potassium affects the growth and synthesis of essential oils in medicinal plants, and these components influence levels of enzymes in the biosynthesis of terpenoids (Sell, 2003).

HPLC Discussion Results

High-performance liquid chromatography (HPLC) chromatographs of lavender (flowers, leaves, and stems) samples were generated for each treatment (Fig5.2-5.10) and peaks were linalool, linalyl acetate, trans linalool, Cis-linalool acetate, limonene only 5 compounds listed ,

respectively. The peaks before 3 min were recognized as solvent peaks. Peak identification: 1 – linalool; 2 – linalyl acetate (Table 5.6).

Effect of DAP and potash fertilizer on essential oils of *Lavandula*

Lavender essential oils contain 24 main molecules determined from the HPLC analysis on chemical composition and homology ratios (Table 5.7), for essential oils for typical chromatography. Nine samples were subjected to quantification, all of them showing a common composition pattern. The composite tariffs were performed using stabilization indicators and co-saturation with the parameters of the 24 examined compounds. Differences in the composition, values, and mean ranges of five compounds of lavender essential oils were found (Table 5.8-5.9).

The results of separation and diagnosis in the data received using HPLC indicate the oil extracted from the lavender plant contained several oil compounds. Linalool and linalyl were identified and the percentage of these compounds was affected by the fertilization with DAP and potash. The essential oil compounds increased in general with DAP when compared with the control treatment. Linalool increased at the level 1g/L DAP and 0 g potash/L. As for the linalyl acetate compound, its percentage increased at the 0g/L DAP and 2g/L potash level.

The effect of fertilizing with DAP and potash on increasing the concentrations of the compounds that make up the essential oil of lavender may be due to increasing plant growth. Note that essential oils are secondary metabolites, and their synthesis increases with the increase in sugars and carbohydrate accumulation (Dhan et al., 1975). Cheang et al. (1987) noted nitrogen is included in the composition of many compounds found in volatile oils. The results of the study demonstrated that high quantities of linalool and linalyl present in three species of lavender. Nine of HPLC chromatographs were developed from the aqueous extract of

Lavandula (Figs. 5.2-5.10). Five compounds were identified in greatest abundance (Table. 5.9) (Fig.5.11). All analyzed samples showed wide diversity in both the number and concentration of the essential oils. The chromatographic profiles ranged from simple (containing as little as one or two components) to complex (containing more than ten components). Many other peaks were detected, such as camphor and a-pinene. Such diversity in composition resulted in the generation of HPLC fingerprints that were unique for almost every one of the analyzed samples.

Linalool was increased with DPA 1g/L × 'Hidcote' × Potash 2g/L. The lowest linalool content was obtained from treatments with the lowest fertilization in 'Platinum Blonde'. The highest linalyl acetate content was determined in the essential oil for plants from DAP 1g/L × 'Provence' × potash 2 g/L and DAP 0g/L× 'Hidcote' × potash 2 g/L. The lowest linalyl acetate content was obtained from DAP 1g/L × 'Provence'. The highest content of cis-linalool was obtained from 'Platinum Blonde' while the lowest Cis-linalool acetate content was obtained from DAP 2g/l × 'Provence'. The highest trans linalool content was obtained from 'Platinum Blonde' . The lowest trans-linalool content was obtained from DAP 1g/l × 'Provence'. The highest limonene content was determined from DAP 0g/l × 'Provence'. The lowest limonene content was obtained from DAP 1 g/l× 'Hidcote' × Potash 2 g/l. However, there were differences between the varieties and trans-linalool, and limonene compound of essential oils of lavender. Depending on the different varieties of lavender, differences in chemical composition have been found. These results agree with Lesage et al. (2015) that the most abundant components were linalool and linalyl acetate, followed by eucalyptol and terpinen-4-ol, while lavandulyl acetate and borneol were identified as secondary compounds. Wellwood and Cole (2004) and MunnéBosch et al. (1999) indicated that variability obtained for content may express the dependence of these compounds' generation on their geographical and agronomic parameters.

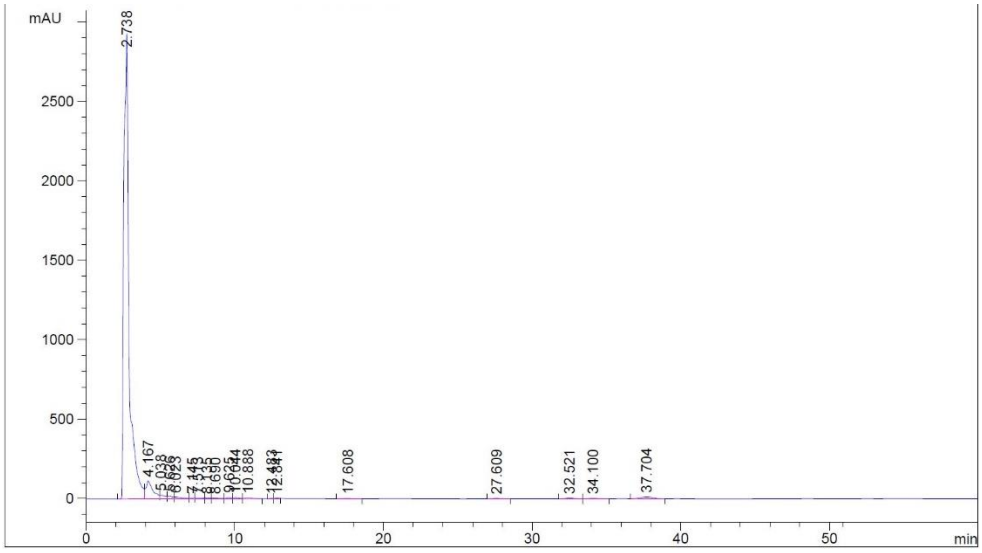

Figure 5.2 Chromatograph of *Lavandula angustifolia* 'Hidcote' oil extract from 2g/l DAP, 2g/l potash.

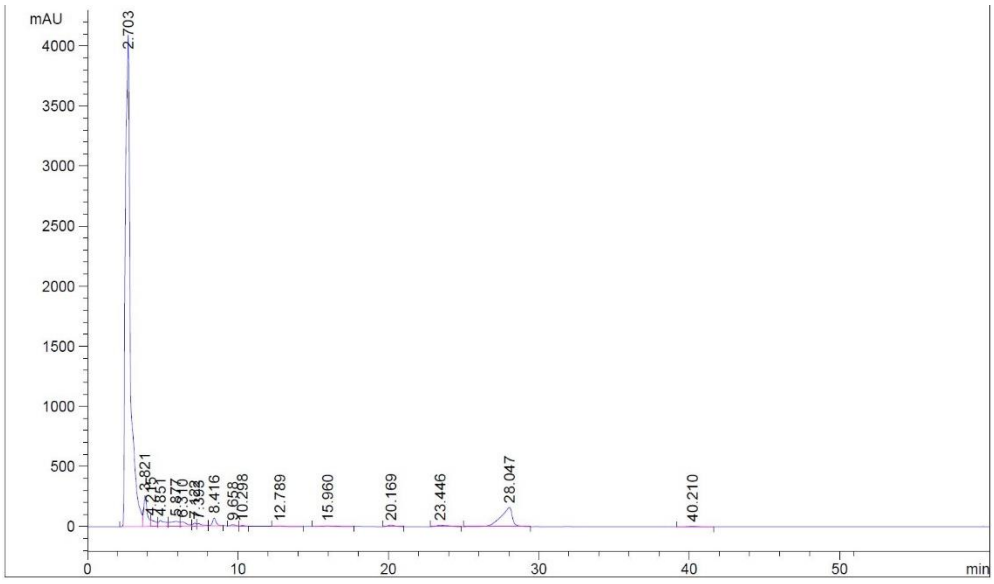

Figure 5.3 Chromatograph of *Lavandula angustifolia* 'Platinum Blonde' oil extract from 1g/L DAP, 1g/L potash.

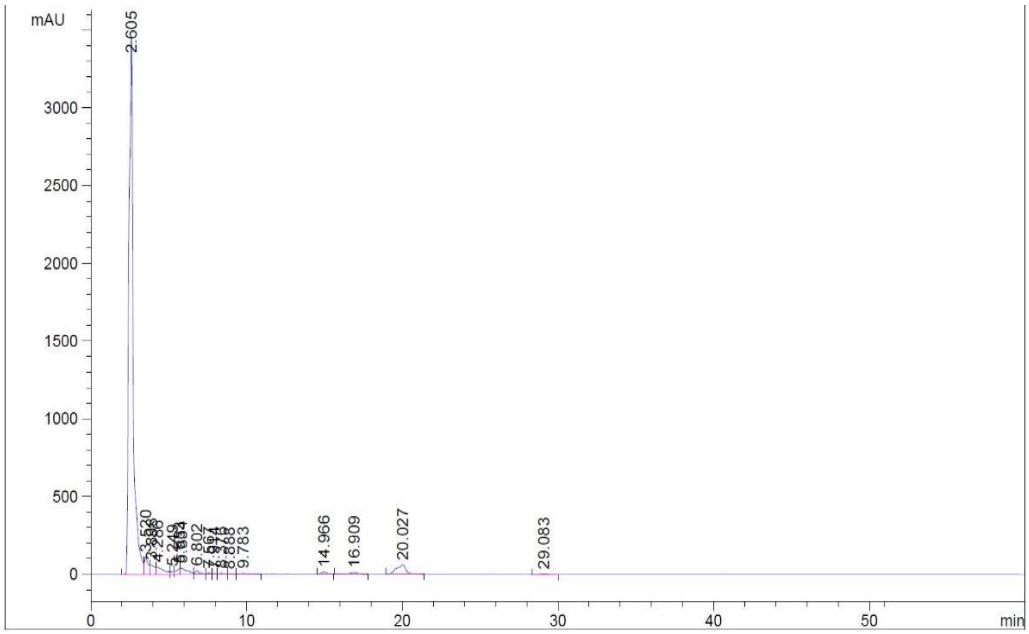

Figure 5.4 Chromatograph of *Lavandula* x *intermedia* 'Provence' oil extract from 2g/l DAP, 0g/l potash.

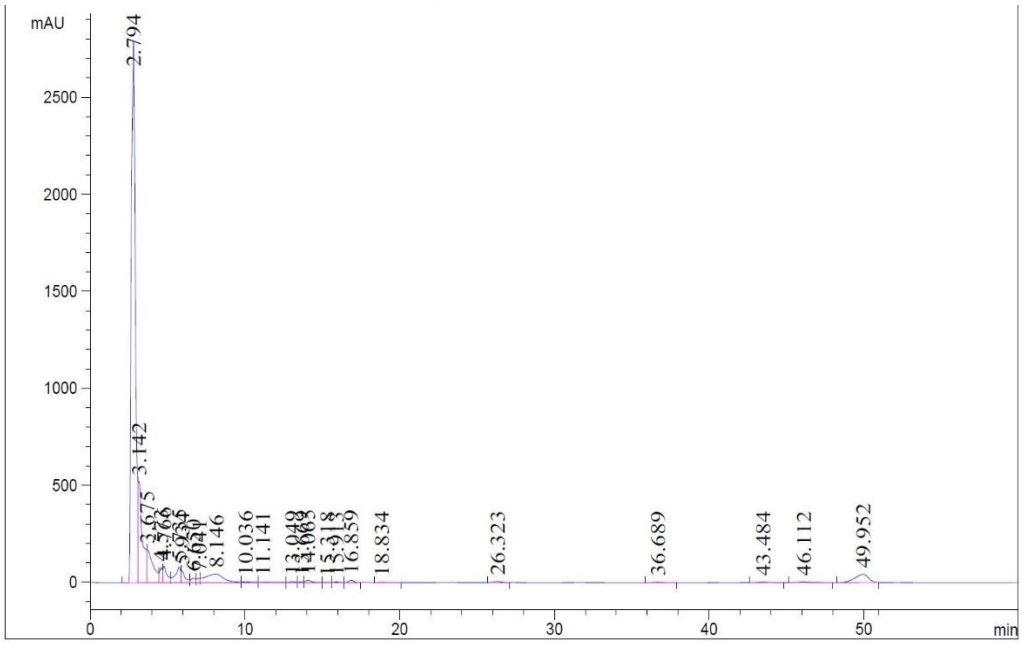

Figure 5.5 Chromatograph of *Lavandula angustifolia* 'Platinum Blonde' oil extract from 0g/l DAP, 0g/l potash.

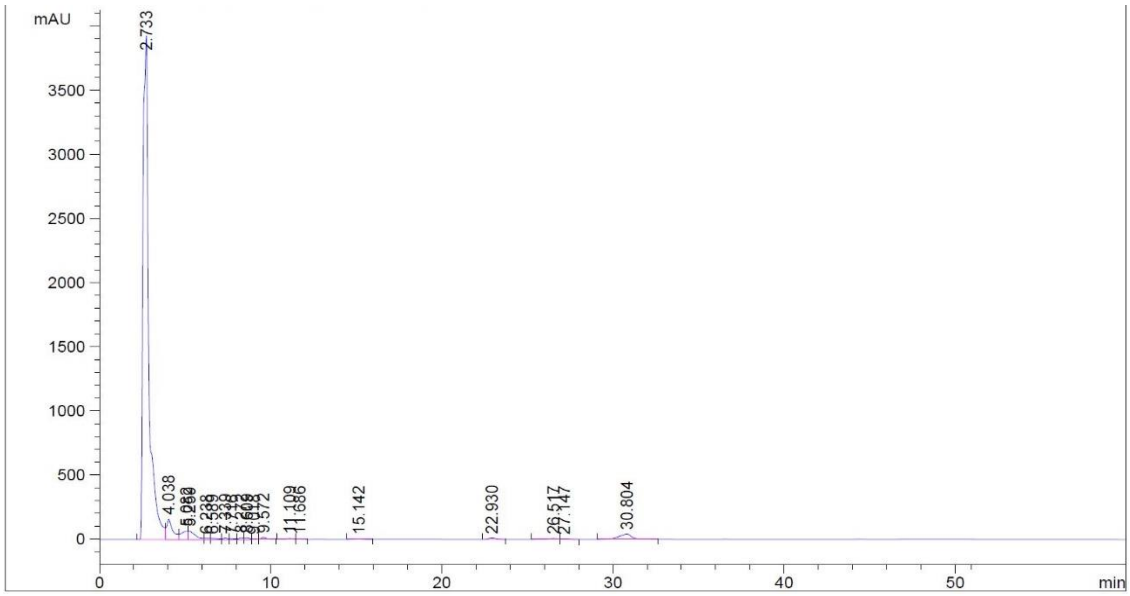

Figure 5.6 Chromatograph of *Lavandula angustifolia* 'Hidcote' oil extract from 0g/l DAP, 0g/l potash.

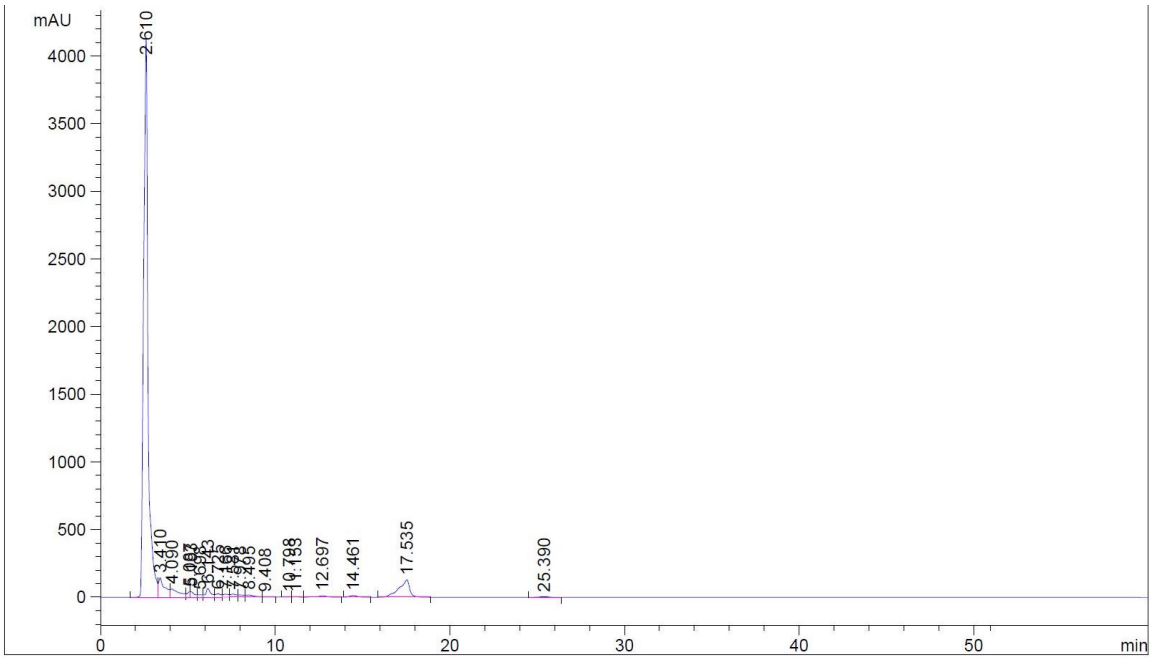

Figure 5.7 Chromatograph of *Lavandula angustifolia* 'Platinum Blonde' oil extract from 2g/l DAP, 1g/l potash.

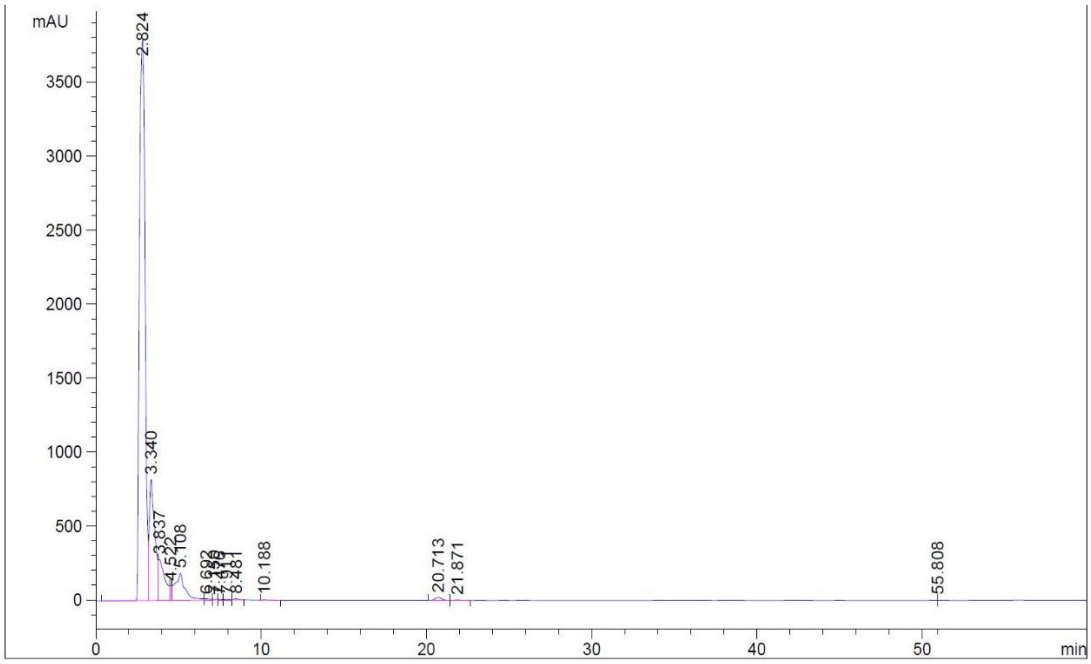

Figure 5.8 Chromatograph of *Lavandula* x *intermedia* 'Provence' oil extract from 1g/l DAP, 1g/l potash.

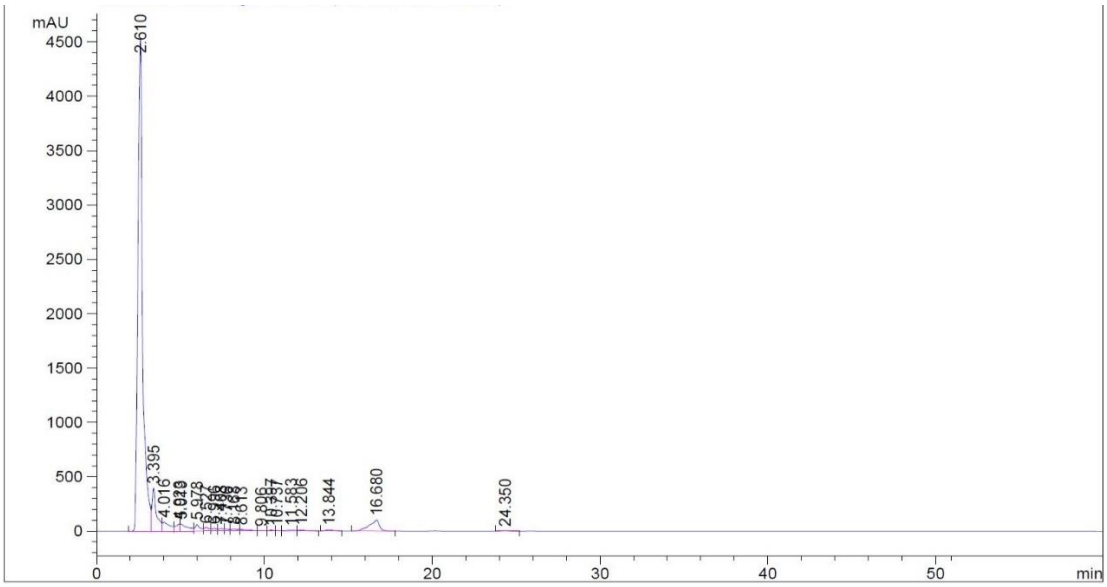

Figure 5.9 Chromatograph of *Lavandula* x *intermedia* 'Provence' oil extract from 0g/l DAP, 0g/l potash.

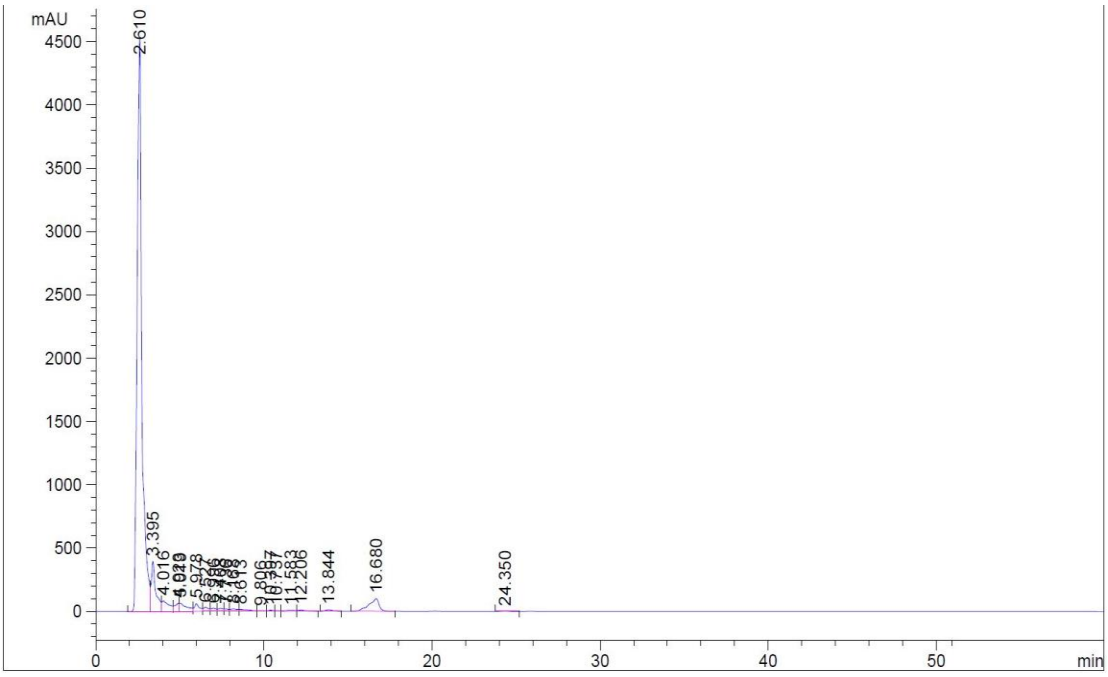

Figure 5.10 Chromatography of *Lavandula angustifolia* 'Hidcote' oil extract from 1g/l DAP, 1g/l potash.

Table 5.6 Quantification of linalool and linalyl acetate in three *Lavandula* species extract by HPLC.

Compound	RT	Concentration % *L. intermedia* 'Provence'	Concentration % *L. angustifolia* 'Hidcote'	Concentration % *L. angustifolium* 'Platinum Blonde'
Linalool	2.73	71.21	90.32	81.33
Linalyl acetate	3.52	15.06	4.70	3.62

Table 5.7 Chemical components identified by HPLC and quantitative composition for the lavender essential oil extracts.

No	RT	Compound	Percentage %	Formula
1	2.79	linalool	63.47	$C_{10}H_{16}O$
2	3.14	Linalyl acetate	11.74	$C_{12}H_{20}O_2$
3	3.67	luteolin 5-O-glucoside	5.81	$C_{21}H_{20}O_{11}$
4	4.57	tyrosol	1.18	$C_8H_{10}O_2$
5	4.76	Chlorogenic acid	2.09	$C_{16}H_{18}O_9$
6	5.73	Vanillic acid	2.39	$C_8H_8O_4$
7	5.94	Terpinene-4-ol	1.27	$C_{10}H_{12}O_2$
8	6.64	Limonene	0.57	$C_{10}H_{16}$
9	7.04	p-couma	0.42	$C_{19}H_{16}O_4$
10	8.14	Ocimene	4.63	$C_{10}H_{16}$
11	10.03	Cis-linalool acetate	0.31	$C_{10}H_{18}O_2$
12	11.14	Trans linalool	0.34	$C_{10}H_{18}O_2$
13	13.04	vanillin	0.20	$C_8H_8O_3$
14	13.66	Stearic acid	0.11	$C_{18}H_{36}O_2$
15	14.06	Trans caryophyllene	0.46	$C_{15}H_{24}$
16	15.31	α-phellandren-8-ol	0.08	$C_{10}H_{16}O$
17	15.91	Unknown	/	/
18	16.85	Eugenol	0.38	$C_{10}H_{12}O_2$
19	18.83	Trans carveol	0.08	$C_{10}H_{16}O$
20	26.68	β -citronellol	0.27	$C_{10}H_{20}O$
21	36.68	Sabinene	0.17	$C_{10}H_{16}$
22	43.48	β -caryophyllene	0.18	$C_{15}H_{24}$
23	46.11	α -pinene	0.32	$C_{10}H_{16}$
24	49.95	α -cedrene	3.26	$C_{10}H_{20}O$

Table 5.8 HPLC data for essential oil components identified in lavender.

No	RT	Compound	Treatment								
			1	2	3	4	5	6	7	8	9
1	2.79	Linalool	90.32	81.33	85.25	63.47	88.81	80.23	71.21	79.47	71.21
2	3.14	Linalyl	4.70	3.62	2.56	11.74	3.78	3.96	15.06	6.54	15.06
3	3.67	luteolin 5-O-gluco side			1.62	5.81			5.89		5.89
4	4.57	Tyrosol		1.02		1.18			0.56		0.56
5	4.76	Chlorogenic acid		1.35		2.09				1.06	
6	5.73	Vanillic acid	0.49		0.82	2.39	1.71	0.37			
7	5.94	Terpinene-4-ol		1.49	1.59	1.27				1.11	
8	6.64	Limonene		0.90	0.58	0.57	0.19	0.63	0.17	0.64	0.17
9	7.04	p-couma	0.18			0.42		0.59	0.08		
10	8.14	Ociemne	0.12			4.63	0.18			0.51	
11	10.03	Cis-linalool acetate	0.24			0.31			0.03	0.15	0.03
12	11.14	Trans linalool				0.34	0.12	0.08			
13	13.04	Vanillin				0.20					
14	13.66	Stearic acid				0.11				0.28	
15	14.06	Trans caryophyllene				0.46					
16	15.31	α-phellandren-8-ol		0.25		0.08	0.07				
17	15.91	Unknown									
18	16.85	Eugenol			0.53	0.38				3.71	
19	18.83	Trans carveol				0.08					
20	26.68	β -citronellol				0.27	0.19	0.18			
21	36.68	Sabinene	0.90			0.17					
22	43.48	β -caryophyllene		0.12		0.18					
23	46.11	α -pinene				0.32					
24	49.95	α -cedrene				3.26					

Sample 1 = *Lavandula angustifolia* 'Hidcote' oil extract from 2g/l DAP, 2g/l potash,
2=*Lavandula angustifolia* 'Platinum Blonde' oil extract from 1g/L DAP, 1g/L potash.
3= *Lavandula x intermedia* 'Provence' oil extract from 2g/l DAP, 0g/l potash.
4= *Lavandula angustifolia* 'Platinum Blonde' oil extract from 0g/l DAP, 0g/l potash.
5= *Lavandula angustifolia* 'Hidcote' oil extract from 0g/l DAP, 0g/l potash.
6= *Lavandula angustifolia* 'Platinum Blonde' oil extract from 2g/l DAP, 1g/l potash.
7= *Lavandula x intermedia* 'Provence' oil extract from 1g/l DAP, 1g/l potash.
8= *Lavandula x intermedia* 'Provence' oil extract from 0g/l DAP, 0g/l potash.
9= *Lavandula angustifolia* 'Hidcote' oil extract from 1g/l DAP, 1g/l potash.

RT= Retention Time.

Unit measure = %

Table 5.9 Effect of DAP and potash fertilizer in diagnosing essential oils of *Lavandula* using high performance liquid chromatography (HPLC).

Essential oils extraction					
Treatment	linalool	Linalyl acetate	Cis-linalool acetate	Trans linalool	Limonene
DAP[2] × 'Hidecote' × P[2]	90.32	4.7	0.24	0.03	0.02
DAP[1] × 'Platinum Blonde' × P[2]	81.33	3.62	0.04	0.05	0.9
DAP[2] × 'Provence'	85.25	2.56	0.03	0.02	0.58
'Platinum Blonde'	63.47	11.74	0.31	0.34	0.57
'Hidcote'	88.81	3.78	0.02	0.12	0.19
DAP[2] × 'Platinum Blonde' × P[2]	80.23	3.96	0.03	0.05	0.63
DAP[1] × 'Provence' × P[2]	71.21	15.06	0.03	0.08	0.17
DAP[1] × 'Provence'	79.47	6.54	0.15	0.06	0.64
DAP[1] × 'Hidecote' × P[2]	71.21	15.06	0.03	0.03	0.17
Constant value	77.25	7.35	0.15	0.15	0.47
(*p* value=0.05)	0.564 ns	0.956 ns	0.138 ns	0.045	0.033
T test	0.601	0.057	-1.649	-2.368	2.569

DAP=Diammonium phosphate g/l P=Potash g/L
Unit measure= %

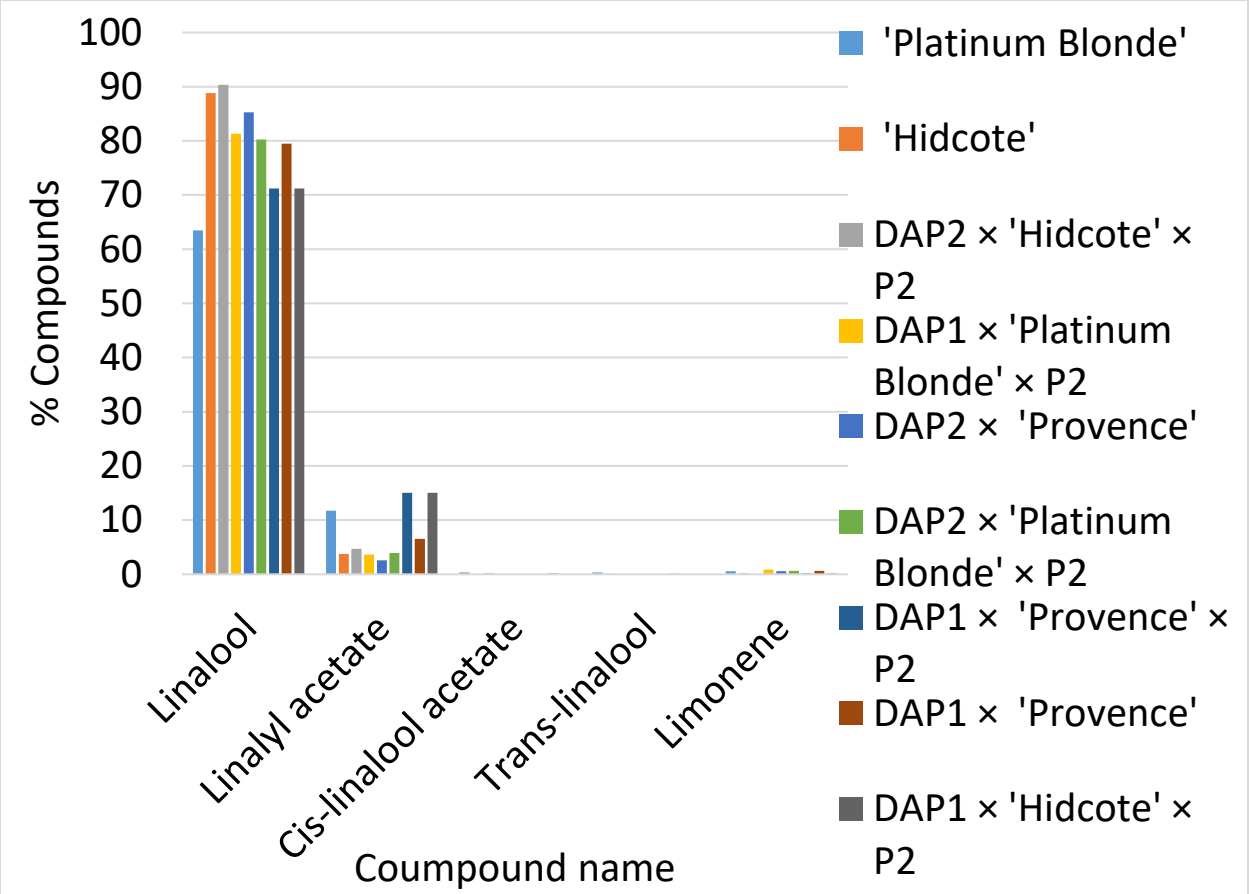

Figure 5.11 Results of HPLC of essential oils compounds in three varieties of lavender treated with DAP and potash.

Conclusions

HPLC is suitable for detecting components of essential oils in lavender. Fertilization with DAP increased growth and essential oil content as DAP concentration increased. Potash increased growth but decreased essential oil production. 'Platinum Blonde' produced more oil than the other varieties; however, the plants did not flower, and the oil was of lower quality. The present results highlight that the growth of aromatic plants and the production of essential oils may be affected positively or negatively by the percentage and quantity of minerals. The HPLC method was successful in identifying and quantifying linalool and linalyl-acetate. The method

was a simple, fast, and efficient method for determining linalool and linalyl in lavender samples. The results showed that all varieties of lavender tested contained a relatively high concentration of linalool and linalyl.

Literature Cited

Adam, K.L. 2006. Lavender Production, Products, Markets, and Entertainment Farms National Center for Appropriate Technology, Butte, Montana, USA.

Anderson, D.L. 1985. Market power and the Saskatchewan potash industry. Can. Public Policy. 11: 321-328.

Arabaci, O. and A. Ceylan. 1990. Research on yield and ontogenetical variability of some perfume plants plants (*Lavandula angustifolia* Mill., *Melissa officinalis L*., *Salvia sclerea L*.). Journ. E.U. Sci. Fac. 1(1): 233-236.

Arabaci, O. and E. Bayram. 2005. Effect of plant density and nitrogen fertilizers on some agronomic and quality characteristics of lavander (*Lavandula angustifolia* Mill.) under the ecological conditions of Aydın. ADU. J. Agr. Fac. 2(2): 13-19.

Azizi, D. Kahrizi. 2008. Effect of nitrogen levels, plant density and climate on yield and quality in cumin (*Cuminum cyminum*) Asian J. Plant Sci. 7: 710-716.

Alsafar, M.S. and Y.M. Al-Hassan. 2009. Effect of nitrogen and phosphorus fertilizers on growth and oil yield of Indigenous mint (*Mentha longifolia* L.). Biotechnol. 8(3): 380-384.

Baranauskienne, P.R., P. Venskutonis, and E. Viskelis. 2003. Dambrausience Influence of nitrogen fertilizer on the yield and composition of thyme (*Thymus vulgaris*) J. Agr. Food Chem. 51(26): 7751-7758.

Biesiada A, A. Sokoi-Letowska, A. and A Kucharska. 2008. The effect of nitrogen fertilization on yielding and antioxidant activity of lavender (*Lavandula angustifolia* Mill). Acta. Sci. Pol. Hortorum. Cult. 7 (2): 33-40.

Bettina M. and Helge S. 2015. The essential oil maker's handbook, IV TP958.M27513 661: 806.

Corrêa, R. M., J.E.B.P., Pinto, E.S. Reis, L.C.B. Costa, P.B. Alves, E.S. Niculan, and R.S. Brant. 2010. Organic fertilization on phytomass production and essential oil conten and quality of oregano (*Origanum vulgare L*.) under protected cultivation. Rev. Bras. Plant. Med. 12(1): 80-89.

Chrysargyris, A., C. Panayiotou, and N. Tzortzakis. 2016. Nitrogen and phosphorus levels affected plant growth, essential oil composition and antioxidant status of lavender plant (*Lavandula angustifolia* Mill.). Ind. Crop Prod. 83: 577-586.

Crummitt, K. and Drost, D., 2012. English Lavender in the Garden. USA, Utah State, published of May 8 and June 30, 1914

Ceylan, A., E. Bayram and N. Ozay. 1996. Effects of different plant densities and nitrogen doses on some agronomic and technological characteristics of Lavender (*Lavandula angustifolia* Mill.). Turk. J. Agri. Forest. 20: 567-572.

Curtis, B. 2005. Lavender Production and Marketing Washington State University (WSU) Cooperative Extension Bulletin.

Cheang, K.H., J.H. Hwan, K.B. Park, D.J. Kang, and Y.S. Lee. 1987. Effect of application levels of nitrogen fertilizer on the growth and chemical compositions of mint. Research reports of the rural development administration crops Korea R. 29(1):289-293.

Daniel, W.W., 1974. Biostatistics; a foundation for analysis in the health sciences. USA, New York : 448 p.m.

Dickes, G.J., P.V. Nicholas. 1978. Gas chromatography in food analysis. Butterworths, London.

Devasagayam, M.N. and Jayapaul. 1997. Varietal's response to graded levels of nitrogen in sesame. Sesame and Safflower. Newsletter. 12: 37-40.

Duhan, S.P.S., B.C. Gulati and A.K. Bhattacharya. 1975. Effect of nitrogen and rew spacing on the yield and quality of essential oil in Japanese mint. Indian J. Agronomy (India). 20(1): 14-16.

Elshorbagy, A. I. 2020. Effect of some amino acid concentration on growth and yield of lavender (*Lavandula officinalis*, CHAIX) Plant under different NPK fertilization levels. Zagazig J. Agric. Res. 47(4): 895-907.

Erbas, S., Z. Kucukyumuk, H. Baydar, I. Erdal, and A. Sanli. 2017. Effects of different phosphorus doses on nutrient concentrations as well as yield and quality characteristics of lavandin (*Lavandula× intermedia* Emeric ex Loisel. var. Super). Turkish J. Field Crops. 22(1): 32-38.

Fuentes, B., M. De La Luz Mora, N.S. Bolan, and R. Naidu. 2008. Assessment of phosphorus bioavailability from organic wastes in soil. Dev. Soil Sci. 32: 363-411.

Daniel, W.W., 1974. Biostatistics; a foundation for analysis in the health sciences (No. 574.0182 D3).

Gerardeaux, E., L. Jordan-Meille, J. Constantin, S. Pellerin, and M. Dingkuhn. 2010. Changes in plant morphology and dry matter partitioning caused by potassium deficiency in *Gossypium hirsutum* (L). Environ. Exp. Bot. 67(3): 451-459.

Huston, J.E., and W.E. Pinchak. 1991. Range animal nutrition. In: Grazing Management Ecological Perspective. (Eds, R.K: Heitshmidt and J.W. Stuth). Timber Press. Oregon. P. 27-63.

Hussain, M.J., M. Peakman, H. Gallati, S.S.S., Lo, M. Hawa, G.C. Viberti, P.J. Watkins, R.D.G. Leslie, and D. Vergani. 1996. Elevated serum levels of macrophage-derived cytokines precede and accompany the onset of IDDM. Diabetologia. 39 (1): 60-69.

Hafsi, C., Debez, A., and Abdelly, C. 2014. Potassium deficiency in plants: effects and signaling cascades. Acta Physiol. Plant. 36(5): 1055-1070.

Kelly, K.O. and R. H.K. Thomson. 1957. Solubility studies on arachin. Journal of Applied Chemistry, 7(8), pp.432-435.

Khalid, A.K. 2013. Effect of potassium uptake on the composition of essential oil content in *Calendula officinalis* L. flowers. Emir. J. Food Agric. 25: 189-195.

Knight, D. 1992. Humphry Davy: Science and Power. Oxford: Blackwell.

Karik, U., F. Cicek and O. Cınar. 2017. Determination of morphological, yield and quali characteristics of lavender (*Lavandula spp*.) species and varieties under ecological conditions. ANADOLU, J. AARI. 27(1): 17-28.

Khalid, K.A. 2012. Effect of phosphorous fertilization on anise, coriander, and sweet fennel plants growing under arid region conditions. Med. Aromat. Plant. Sci. Biotechnol. 6 (1): 127-131.

Klados, E. and N. Tzortzakis. 2014. Effects of substrate and salinity in hydroponically grown *Cichorium spinosum*. J. Soil Sci. Plant Nutr. 14 (1): 211-222.

Kokate, C.K. and K.C. Varma. 19971. Cultivation of lemon grass and cirtronella grass Roudle of sagar of Madhya Pradesh. Indian J. Agric. Sci. 41(4): 382-385.

Krideum, M. and B. Leopold. 1975. Anise oils progress in essential oils. 25: 25-27.

Kanai, S., K. Ohkura, J.J. Adu-Gyamfi, P.K. Mohapatra, N.T. Nguyen, H. Saneoka, K. Fujita. 2007. Depression of sink activity precedes the inhibition of biomass production in tomato plants subjected to potassium deficiency stress. J. Exp. Bot. 58(11): 2917-2928.

Lis-Balchin, M. 2002. Lavender essential oil: Standardisation, ISO; adulteration and its detection using GC, enantiomeric columns, and bioactivity. In *Lavender*. CRC Press. 131-137.

Lu, H.T., H.B. Li, F. Chen, M.H. Wong, and Y. Jiang. 2004. Determination of triphenyltin and its metabolite diphenyl tin in culture medium by high-performance liquid chromatography with UV detection. Chromatographia. 60(1): 113-116.

Morais, L.A.S. 2009. Influência dos fatores abióticos na composição química dos óleos essenciais. Hortic. Bras. 27(2): 4050-4063.

Marschner, P. 2012. Marschner's Mineral Nutrition of Higher Plants, 3rd ed.; Academic Press: London, UK. 178-189.

Ma, J.F., Y. Miyake, and E. Takahashi. 2001. Silicon as a beneficial element for crop plants. Studies Plant Sci. 8: 17-39.

Maher, H.S. 2019. GenStat for Analyzing Agricultural Experiments, AL-Qassim Green University- Agriculture College. (Book).

Munné-Bosch, S., S. Nogués, L. Alegre. 1999. Diurnal variations of photosynthesis and dew absorption by leaves in two evergreen shrubs growing in Mediterranean field conditions. New Phytol. 144(1): 109-119.

Naderidarbaghshahi, M. S.M. Monemian H. Zeynali and B. Bahari. 2011. Effects of different levels of nitrogen, phosphorus, and potassium fertilizers on some agro-morphological and biochemical traits of German chamomile (*Matricaria chamomilla* L.). J. Med. Plants Res. 6(2): 277-283.

Parejo, F., J. Viladomat, A. Bastida, N. RosasRomero, J. Flerlage, C. Burillo. 2002. Coding Comparison between the radical scavenging activity and antioxidant activity of six distilled and nondistilled Mediterranean herbs and aromatic plants. J. Agric. Food Chem. 50(23): 6882-6890.

Pettigrew, W.T. 2008**.** Potassium influences on yield and quality production for maize, wheat, soybean, and cotton. Physiol. Plant. 133(4): 670-681.

Qadry, J.S. 2010. Pharmacognosy.16th Ed., Somya Printers. Delhi-5. Edn. 2010. 15: 305-15.

Rawashdeh, R.A and P. Maxwell. 2014. Analysing the world potash industry. Resour. Policy. 41: 143-151.

Ramezani, M.R., P. Rezaei, and P. Sotoudehnia. 2009. Improved growth, yield and essential oil content of basil grown under different levels of phosphorus sprays in the field J. Appl. Biol. Sci. 3(2): 105-110.

Reverchon, E. G. Della Porta. 1994**.** Food Chem. J. Agric. 43: 16-54.

Robertsa, L. 2014. Cadmium and phosphorous fertilizers: The issues and the science, International Plant Nutrition Institute, Norcross, GA.

Rathore, S.V.S., D.K. Dera and U. Chand. 1985. Studies of nitrogen nutrition through foliar spray of urea on the performance of African marigold. Udyarika. 5: 37-40.

Sell, C.S. 2003. A fragrant introduction to terpenoid chemistry**.** The Royal Society of Chemistry, Thomas, Graham House, Scientific Park Miltron Roasd Cambidge, UK.

Sanghamitra, M., V.V. Bhaskar, A.V.D.D. Rao, and P. Subbaramamma. 2015. Effect of different sources and levels of potassium on yield and carotenoids content of African Marigold (*Tagetes erecta* Linn.) cv."Maxima Yellow". Plant Archives, 15(2): 633-636.

Sharafzadeh, S. M., A.H. Esmaeili, and A.H. Mohammadi. 2011. Interaction effects of nitrogen, phosphorus and potassium on growth, essence, and total phenolic content of sweet basil. Adv. Environ. Biol. 5(6): 1285-1289.

Silva, S.M., J.M.Q. Luz, P.A.M. Nogueira, A.F. Blank, T.S. Sampaio, J.A.O. Pinto, and A.W. Junior, 2017. Organo-mineral fertilization effects on biomass and essential oil of Lavender (*Lavandula dentata L.*). Ind. Crops Prod. 103: 133-140.

Shabala, S., I.I. Pottosin. 2010**.** Potassium and potassium-permeable channels in plant salt tolerance. Signal. Commun. Plants. 87-110.

Thimann, K.V., T. Sachs, and K.N. Mathur. 1971. The mechanism of apical dominance in coleus plant. Plant Physiol. 24(1): 68-72.

Tisdale, S.L., W.L. Nelson, and J.W. Beaton. 1985. Soil and fertilizer potassium. In: Soil Fertility and Fertilizers 4th ed., S.L. Tisdale, W.L. Nelson, and J.D. Beaton (eds.), MacMillan Publ. Co., New York. 249-291

Touati, B., H. Chograni, I. Hassen, M. Boussaid, L. Toumi, and N.B. BrahiMm. 2011. Chemical composition of the leaf and flower essential oils of Tunisian *Lavandula dentata* L. (Lamiaceae). Chem. Biodivers. 8(8): 1560-1569.

Van Ginkel, B. 1972. Botanical and carrying capacity change through fertilization of Cymbopogon themed veld type. Safr. Dept. Agric, Tech. Serv. Common. 98: 1-11.

Woronuk, G., Z. Demissie, M. Rheault, and S. Mahmoud. 2011. Biosynthesis and therapeutic properties of *Lavandula* essential oil constituents. Planta medica. 77(1): 7-15.

Westervelt, P.M. 2003. Challenges in the greenhouse production of *Rosmarinus officinalis* L (Doctoral dissertation, Virginia Tech).

Wellwood, C.R.L., Cole R.A. 2004. Relevance of carnosic acid concentrations to the selection of rosemary, *Rosmarinus officinalis L.*, accessions for optimization of antioxidant yield. J. Agric. Food Chem. 52(20): 6101-6107

Yasemin, S., A. Ozkaya, N. Koksal, and B. Gok. 2017. The effects of nitrogen on growth and physiological features of lavender. Full Text Proceedings Book Adana, Turkey, published May 10, .