



بسم الله الرحمن الرحيم



# Sudan University of Science and Technology

## College of Graduate studies

Physicochemical study of samples of basil from sudan

دراسة فيزيوكيميائية لعينات من الريحان في السودان

**A Thesis submitted in partial full for the  
requirements of the Degree of Master in chemistry**

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Feb 2022



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## إستهلال

قال الله عز وجل :

فَرْوَحٌ وَرِيحَانٌ وَجَنَّتْ نَعِيمٌ ﴿٨٩﴾

سورة الواقعة (89)

وَالْحَبُّ ذُو الْعَصْفِ وَالرَّيْحَانُ ﴿١٢﴾

سورة الرحمن (12)

## Dedication

This work dedicated to the soul of *my father*.  
A special feeling of gratitude to *my mother*.  
A special thanks to *my dear sister AYAH*, my brothers **Freed** and **Ahmed**.

## Acknowledgment

My deepest thanks go to Allah almighty my creator, my source of inspiration, wisdom, knowledge and understanding.

Deep appreciation to my supervisor Professor **DR. Mohammad Abboto** his leadership, encouragement, and valuable advice.

I would like to acknowledge and thank the members of the Medicinal and Aromatic Plant and Traditional Medicine Research Institute [MAPTMRI] for producing samples & information. Special appreciation for **Prof Magda Abakar Osman** and **Abdelgani Muhammad** for their valuable support.

I could not forget my family for constant supporting specially my parents. And I am grateful for my friends who have supported me throughout the process.

## Abstract

This research was aimed to study two samples of fresh basil leaves (*Ocimum basilicum* L and *Ocimum basilicum charmandaschricum*). The leaves of sweet basil have sufficient amount of water, protein, ash fibre and fat.

Result of this study indicated that leaves of *Ocimum basilicum* L was a large value of water content (87.140%) nevertheless, *Ocimum basilicum charmandaschricum* was excellent source of other nutritive composition. The output of proximate analysis of *Ocimum basilicum* L was found as water content (87.140%) > fibre (5.88%)>protein (3.5%)>ash (3%)> fat (0.4%).

The proximate analysis of *Ocimum basilicum charmandaschricum* was shown the percentage of water content (80.463%) >fibre (9.62%) >protein (4.375%)>ash (3.9%)> fat (0.6%).

The constituent of basil leaves can be used as potent nutrient of major minerals such as Na, K, Ca, Mg, Mn and minor elements such as Zn, Cu and Fe in the diet.

The analysis of leaves were determined and the result showed that , the presence if higher amount of K was found in *Ocimum basilicum* L while the greater amounts of other minerals were found in *Ocimum basilicum charmandaschricum*.

The percentage of mineral elements content in *Ocimum basilicum* L were: K (0.112%)>Na (0.053%) > Mg (0.06%) >Ca (0.04%)> Fe (0.009%)> Zn (0.0002%) > Mn (0.00017%)> Cu (0.00012%) whereas, the elements of *Ocimum basilicum charmandaschricum* were:Na (0.107%) >Ca (0.09%)> Mg (0.07%) > K(0.057%) Fe (0.01%)> Zn (0.0003%) > Cu (0.00015%) and Mn not detected .that demonstrate the *Ocimum basilicum charmandaschricum* is a food source of Na while *Ocimum basilicum* L is a good source of K all in all, that gave the basil importance of food .

The chemical composition of the volatile oils of the leaves of fresh basil were obtained by hydro distillation then analyzed by GC-MS, due to that however, the yield of *Ocimum basilicum charmandaschricum* was (2.3%).

The volatile oil were characterized by (GC-MS);the most common compounds of *Ocimum basilicum* L were Eugenol (39.11%) , Caryophyllene(30.39%) ,Naphthalene (24.21%),Caryophyllene oxide (1.85%) and Humlene (1.54%).

Eugenol was the major constituent of the volatile oil of *Ocimum basilicum* L.

Nonetheless, the major constituent of the volatile oil of *Ocimum basilicum charmandaschricum* was Estragole (47.6%) followed by Bicyclo [3.1.1] hept-2-ene,2,6-dimethyle-6- (13.17%), 1,6-octadien-3-ol,3,7 dimethyl- (10.81%), Eucalyptol (7.65%), Tau-cadinol(6.05%) , phenol,2-methoxy-4-(2-propenyl)-acetate (2.84%), Gamma (1.88%) and Cyclohexene 1-ethenyl-2,4-bis (1.02%).

## مستخلص البحث

يهدف البحث إلى دراسة التحليل التقريبي لعينتين من أوراق الريحان الطازجة (الريحان الحلو والريحان القرنفلي)، تحتوي أوراق الريحان الحلو على كمية كافية من الرطوبة والبروتين والرماد والألياف والدهون، وقد أشارت نتائج هذه الدراسة إلى أن أوراق نبات الريحان الحلو كانت ذات قيمة عالية للماء (87.14%) في حين التراكيب الغذائية الأخرى كانت في الريحان القرنفلي. تم إجراء التحليل التقريبي للريحان الحلو وكانت النتائج كالآتي : ماء (87.14%) <ألياف (5.88%) < بروتين (3.5%) < رماد (3.009%) < دهون (0.04%) ، كما أظهر التحليل التقريبي للريحان القرنفلي وكانت النتائج كالآتي : ماء (80.46%) ، ألياف (9.62%) ، بروتين (4.375%) ، رماد (3.9%) ، دهون (0.6%).

يمكن استخدام التركيبة التقريبية لأوراق الريحان كمغذيات قوية للعناصر الرئيسية مثل K ، Mn ، Ca ، Mg والعناصر الثانوية مثل Zn ، Cu و Fe في النظام الغذائي. أظهر تحديد الأوراق وجود كمية أعلى من k تم العثور عليها في الريحان الحلو بينما تم العثور على كميات أكبر من المعادن الأخرى في الريحان القرنفلي وكانت النسب المئوية لمحتوى العناصر المعدنية في الريحان الحلو في البوتاسيوم (0.112%) < الصوديوم (0.053%) < المغنيزيوم (0.06%) < الكالسيوم (0.04%) < الحديد (0.0095%) < الخارصين (0.0002%) < النحاس (0.00012%) ، بينما النسب المئوية للريحان القرنفلي كانت في الصوديوم (0.107%) < الكالسيوم (0.09%) < المغنيزيوم (0.07%) < البوتاسيوم (0.057%) < الحديد (0.01%) < الخارصين (0.0003%) < النحاس (0.00015%) بينما المنجنيز لم يتم تحديده. وهذا يدل على أن الريحان القرنفلي مصدر غذائي للصوديوم بينما يعد الريحان الحلو مصدراً جيداً للبوتاسيوم بشكل عام ، مما أعطى الريحان أهمية غذائية عالية.

تم الحصول على التركيب الكيميائي للزيوت المتطايرة لأوراق الريحان الطازجة بالتقطير المائي ثم تحليل الزيت الطيار بواسطة جهاز GC-MS وكان الناتج (1.9%) لأوراق نبات الريحان الحلو ، و (2.3%) لأوراق نبات الريحان القرنفلي.

تم تحديد الزيوت الطيارة بواسطة كروماتوجرافيا الغاز – مطياف الكتلة (GC-MS) وكانت المركبات الأكثر شيوعاً التي تم الكشف عنها بواسطة قياس الطيف الكتلي للكتلة الغازية للريحان القرنفلي كالتالي : Eugenol (39.11%) ، Caryophyllene (30.39%) ، Naphthalene (24.21) ، Caryophyllene oxide (1.85%) ، Cyclohexene (1.02%) ، والـ



Estragole Humlène(1.54%) . بينما كان المكون الأساسي الريحان الحلوه هو الـ  
Bicyclo [3.1.1] hept-2-ene,2,6-dimethyle-6-(13.17%) ، ويليه (47.06%)  
Tau-Eucalyptol (%7.65) ، 1,6-octadien-3-ol,3,7,-dimethyl-(10.81%)،  
Phenol,2-methoxy-4-(2-propenyl)-acetate (2.84%)،cadinol(6.05%)  
. Cyclohexene,1-ethenyl-1-methyl-2,4-bis (1.02%)، Gamma(1.88%)،

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**List of Abbreviation:**

| Abbreviation | Description                                    |
|--------------|--|
| NIST         | National Institute of Standards and Technology |
| PCA          | Principal Component Analysis.                  |
| OES          | Optical Emission Spectrometry.                 |
| HG           | Hydride Generation.                            |
| CFU          | Colony Forming unit.                           |
| ICP          | Inductively Coupled Plasma.                    |
| PCA          | Principal Component Analysis.                  |

# **CHAPTER ONE**

## *Introduction and Literature Review*

# **1. INTRODUCTION**

## **1.1 Natural products**

Is a chemical substance produced by a living organism; a term used commonly in reference to chemical substances found in Nature that Have distinctive pharmacological effects (Lemin 2005).

Natural products are chemical compounds or substances isolated from living organism (Abiola, et al. 2016).

### **1.1.1 Main Classes of Natural Products**

- Carbohydrates
- Lipids
- Proteins
- Nucleic Acids.

Metabolism is defined as series of enzyme catalyzed biochemical reaction or transformation occurring within the cells of an organism which are mainly required for its growth, development and for proper response to its environment (Lemin 2005).

A primary Metabolites is directly involved in normal Growth, development and reproduction. Example: carbohydrate, protein, fat and oil, alcohol etc. Secondary metabolites are not directly involved in growth, development and reproduction of an organism, but they have an ecological function. Plant secondary metabolite can be found in the leaves, stem, root or the bark of the plant depending on the type of secondary metabolite that is been produced. The most bioactive secondary metabolite are the Alkaloids, Tannins, Flavonoids and Phenolic compounds. Many of these secondary metabolites are indigenous plant use as food, spices and herbs. Secondary metabolites differ from primary metabolite in having a restricted distribution in the plant kingdom. That is, particular secondary metabolite Are found in only one plant species or related group of species, whereas primary metabolites are found throughout the plant kingdom. For many years these compounds were thought to be simply functionless end products of metabolism, or metabolic wastes. studies of these substances Was pioneered by organic chemist of the nineteenth and early twentieth century who were interested in these substances because of their importance as medicinal drugs,



poison ,Flavor and industrial material.Only 5% to15% of plant species have been chemically analyzed so far (Lemin 2005).

## **1.2 Medicinal Plants**

### **1.2.1 Global dimension of traditional medicines**

Traditional medicines are used widely throughout the world. As the name implies, these treatments are a part of the traditions of each country that have been handed down from generation to generation. Acceptance of traditional medicines by a population is largely conditioned by cultural factors. Acknowledging the potential value of traditional medicine for the expansion of health service, the World Health Assembly (WHA) passed a number of resolutions in 1976 to draw attention to the potential reserve constituted by traditional practitioners. In 1977, WHA urged countries to utilize their traditional system of medicine (Abdalla et al., 2012).

### **1.2.2 Sudanese traditional medicine**

Sudanese folk medicine represents a unique blend of indigenous cultures of Islamic, Arabic and African traditions. Consequently, treatments exist for a variety of diseases, both epidemic and endemic.

To face these diseases, people have tapped the environmental resources, e.g. plants, minerals and animal products for the management of their health (Abdalla et al., 2012).

The Sudanese have amassed a large body of curative methods, techniques and recipes. Readers may find further recent and detailed information in the descriptive inventory, which appeared in the Atlas of Medicinal Plants series published by Medicinal and Aromatic Plants Research (MAPRI). This series includes comprehensive surveys of the medicinal plants Erkawit, Nuba Mountains, White Nile, North Kordofan, and Angasana (Abdalla et al., 2012).

Sudanese medicinal plants have been reported to exert antimicrobial activity against viruses, bacteria, and protozoa.As infections with worms or molluscs represent a common affliction in that area, medicinal plants have been considered for treatment of these infections (Abdalla et al., 2012).

## **1.3 Basil**

The basil herb (*Basilici herba*) is one of the most frequently used culinary and pharmacological raw materials, containing a significant amount of biological components with strong curative properties. Basil is a plant of

warm climate and is very demanding as to temperature and insolation, as well as soil fecundity and humidity (Dzida 2010).

Basil has low calorific value and high nutritional values. It contains carotene, vitamins A, B6, C, as well as calcium, potassium, phosphorus, magnesium, iron. The basil herb also contains flavonoids and is an antioxidant (Dzida 2010).

Basil is an annual herb native to Asia and the Middle East and is common in Sudan (Abdalla et al., 2012).

Name: In the English language, it is typically called basil, common basil or sweet basil. In India, specifically in Hindi and Bengali, it called babui tulsi. Other common names of basil are basilica (in French), basilikum or basilienkraut (in German), basilico (in Italian), rehan (in Arabic) and albahaca (in Spanish). In Arabic, it is known as hebak as well as Rihan (Al-Maskri et al., 2012).

There are more than 160 named cultivars in existence today. Popular examples include, *O. basilicum* 'Cinnamon', *O. basilicum* 'Dark Opal' and holy basil (the species *O. tenuiflorum* L., previously known as *O. sanctum* L.). Scents and flavours can range from cinnamon, liquorice and lemon to anise. The plants can be shrubby or herbaceous, and vary in size from 20 cm to 3 m tall, depending on the species (and the literature source used). The leaves can be smooth, shiny, hairy or curly, and they can be green to blue/purple. The flower colour ranges from white to purple to lavender (Al-Maskri et al., 2012).

Habitat: Basil grows in the wild and is also cultivated in northern and central Sudan, especially near streams (Abdalla et al., 2012).

Constituents: Volatile oil containing cineol, pinene, methyl chavicol, d-camphor and ocimene (Abdalla et al., 2012).

The essential oil content of basil is show a similar variability between species and cultivars and is thought to be the result of varying ecological factors, geographic origins, genetic patterns, different chemo types and differences in the nutritional status of plants. The bulk of the essential oil of basil plants is concentrated in the leaves and flowers; there are trace quantities of essential oils in the branches and

stems, but the amounts are not commercially important (Al-Maskri et al., 2012).

### **1.3.1 Family**

Basil (*Ocimum basilicum* L.) is an annual herb belonging to the mint family (Lamiaceae) (Al-Maskri et al., 2012).

### **1.3.2 The Genus**

The genus *Ocimum* contains a range of some (50 to 150) species and varieties that are native to the tropical regions of Asia and Central and South Africa. The uncertainty in the exact number of species within the genus is largely attributed to the enormous variation that is found among the constituent species. The variability is prevalent in the morphology, growth habit, flower colour, leaves, stems and chemical composition (Al-Maskri et al., 2012).

### **1.3.3 History and geographical Distribution**

Basil is grown widely in the following countries: India, Pakistan, Comores Islands, Madagascar, Haiti, Guatemala, Réunion, Thailand, Indonesia, Russia (Georgia, East Caucasus) and South Africa, Egypt, Morocco, France, Israel, Bulgaria, the USA (Arizona, California, New Mexico), Italy, Hungary, Poland, Germany, Greece, Turkey, other Balkan countries and Slovakia (Al-Maskri et al., 2012).

Basil oils were produced in the following countries (the quantities that follow in parentheses are tons): India (15), Bulgaria (7), Egypt (5), Pakistan (4.5), the Comoros (4.5), Israel (2), the former Yugoslavia (1), the USA (1), Madagascar (1), Réunion and Albania (each 0.5), Hungary (0.3) and Argentina (0.2). USA is probably the largest market for basil oil, followed by the European countries of Germany, France, the UK and the Netherlands (Al-Maskri et al., 2012).

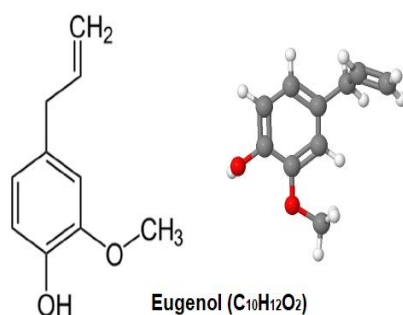
Global statistics for the production of dried basil are also hard to obtain. A large portion of the world production, chiefly in the Mediterranean region, and in India and California, is not sold internationally; most of the basil in these areas is consumed locally. Import statistics also show that the USA is one of the world's biggest users of dried basil. Other important areas for basil importation are the European countries. In the

1990s, the total amount of basil herb imported to Europe was about 830–880 t/year. France is the largest importer at 300–350 t/year, followed by the UK (250 t/year), Germany (200 t/year) and the Netherlands (80t/year). The largest supplier of the Western European countries was Egypt (Al-Maskri et al., 2012).

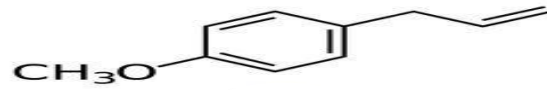
### 1.3.4 Chemistry of Basil

Basil is an impressively aromatic plant and is used as a sweat-smelling herb. Different phenotypic characters, including taste, aroma and many others, are used to de-scribe a varies from 30 to 300 cm and leaf color from green to blue/purple; this de-pends on the type of species (Al-Maskri et al., 2012).

The name of each basil type often represents its particular flavor, with the exception of the sweet basil, whose taste is bright and pungent; anise basil, lemon basil and cinnamon basil offer unique flavors as indicated by their names. The essential oil present in the leaves and other parts of a number of basil species/cultivars is responsible for its distinctive fragrance and aroma. In most species of basil, methyl chavicol, eugenol and linalool are major components. Different species or cultivars have different amounts of each of these chemical constituents, which hence are responsible for the different taste and aroma of each basil cultivar. As an example, the sweet aroma of methyl chavicol has been compared with that of French tarragon and anise, while a floral scent is produced by linalool and eugenol is reminiscent of cloves. The major component present in sweet basils is methyl chavicol while eugenol is present in large amount in spicy basils. Other chemical components responsible for flavour include geranial (a rose flavour), thymol (a thyme flavour), camphor, transmethyl cinnamate (a cinnamon flavour) and citral (lemon) (Al-Maskri et al., 2012).

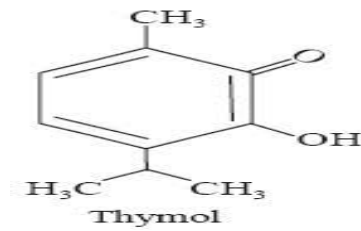


**Figure NO (1.1): Eugenol**

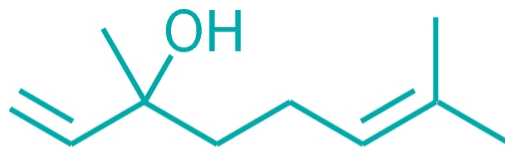


**Methyl Chavicol**

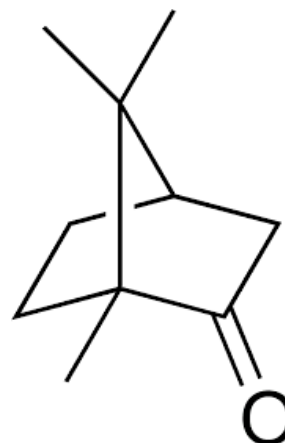
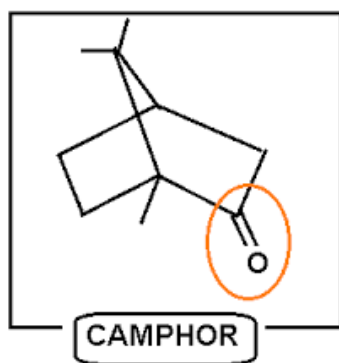
**Figure NO (1.2): Methyl Chavicol**



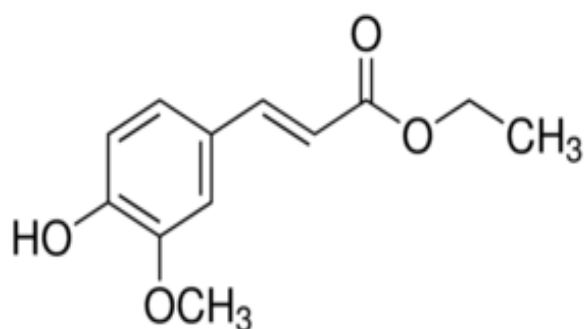
**Figure NO (1.3): Thymol**



**Figure NO (1.4): linalool**



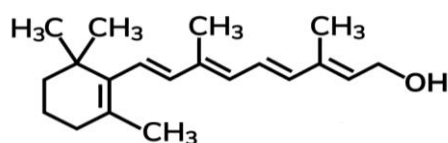
**Figure NO (1.5): Camphor**



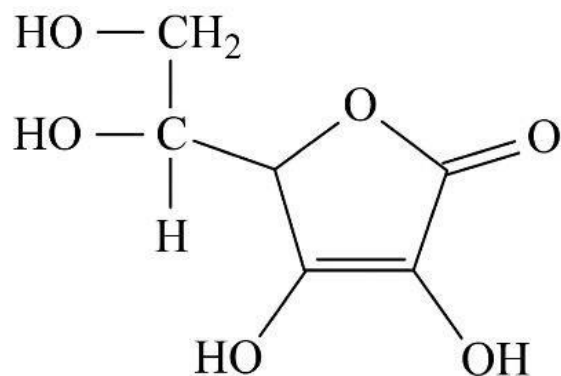
**Figure NO (1.6): Citral**

### 1.3.6 Chemical Composition

In sweet basil, the fat content and calorific value is low while high amount of minerals and vitamin A are present. In 2.5 g of basil leaves (five fresh leaves), there are 96.6 IU vitamin A, 3.85 mg calcium, less than 1 calorie, 11.55 mg potassium, and smaller proportions of vitamin C and other vitamins, protein, fiber and minerals. The GRAS (generally recognized as safe) list of the US Department of Agriculture includes sweet basil leaf to be used in the range of (2–680) ppm and 0.01–50 ppm for the essential oil. The use of exceedingly large quantities of oil is suggested to have a health risk due to the occurrence of carcinogenic compounds. The GRAS-suggested amount of basil essential oil is very minute, and internal use of a large amount of this oil should be avoided (Al-Maskri et al., 2012).



**Figure NO (1.7): Vitamin A**



**Figure NO (1.8): Vitamin C**

### 1.3.7 Uses

The essential oil obtained by distillation is used in perfumery and in food industry as aromatic and Flavoring agent (Dzida and K (2010)). Basil has an extensive list of traditional medical uses. *O. basilicum* has more than 50 medicinal uses, from analgesic to anthelmintic, and is supposed to treat fungal infections, acne, headaches and over 100 such conditions (A., Chhaya et al., 2013). The traditional Chinese medicine system involves the use of *O. basilicum* for treatment of gum ulcers, kidney problems and as a haemostyptic in childbirth. In India, it is used for problems as diverse as earache, menstrual irregularities, arthritis, anorexia and malaria. Rihan (*O. basilicum* in Arabic) is used in treatment of colds, cataract and diarrhea in northern and central Oman. Rihan (the Persian name) is used to treat urinary tract infection, chest and lung problems, ulcers and influenza, and in Iran, basil is employed as a tonic, appetizer and expectorant (Al-Maskri et al., 2012). In Jordan, an infusion of basil is considered to be anthelmintic, anti-emetic and antidiarrhoeal (Al-Maskri et al., 2012). In Guinea, the leaves and stems are used to treat fever, neuralgia, catarrh and renal troubles. In Ethiopia, the leaves are used against malaria, headache and diarrhoea. In homeopathy, the fresh mature leaves are used to treat blood dysentery, inflammation and congestion of the kidney. The roots and the leaves are used to treat bowel complaints in children (Al-Maskri et al., 2012).

Basil has been found to show effectiveness against many fungal, viral, bacterial and protozoal infections. Current studies suggest that basil is helpful in inhibiting the growth of carcinogenic cells and in HIV. Basil leaves are used specifically to treat many fevers and coughs, flu, asthma, influenza, bronchitis, colds, chicken pox and diarrhoea, and they

can lower the cholesterol level in blood and act as anti-stress agents. Basil juice is an effective medicine for inflamed eyes and nightblindness, which is often caused by vitamin A deficiency. There are frequent studies on the antifungal activity of *Ocimum* leaves, essential oils and their components and extracts. Fresh ripe tomato fruits were treated before and after inoculation with *Aspergillus niger* in the presence of *Drosophila busckii* by an ethanolic extract of *O. tenuiflorum*. The fruits did not show signs of rotting for 5 to 7 days after this treatment. The essential oil of *O. canum* was successful against the fungi causing damping-off disease, *Pythium aphanidermatum*, *P. debaryanum* and *Rhizoctonia solani*. *O. canum* gave a 50% reduction in damping-off disease of tomato plants in *P.aphanidermatum*-infected soil and up to 43% reduction in *P.debaryanum*infected soil. Phytotoxicity of this essential oil was not observed and it was superior to common synthetic fungicides such as captan. *O. basilicum* essential oil displayed antifungal properties against an *Aspergillus flavus* strain producing aflatoxin and against *A. parasiticus*. The fungistatic properties of the oil were observed at a dose of 1.5 ml/l and the fungicidal properties at 6.0 ml/l. These doses are much lower than those of industrial synthetic fungicides and fumigants, and effect of the oil treatment is not altered by storage, temperature or increased inocula.<sup>47</sup> Antimicrobial activity of sweet basil has been found against such organisms as *Lactobacillus acidophilus*, *Saccharomyces cerevisiae*, *Mycoderma* sp., *A. niger* and *Bacillus cereus* (Al-Maskri et al., 2012).

### **Pharmacological uses**

Basil oil is known to have strong antioxidant properties. Research has shown the oil contains potent anticancer, antiviral and antimicrobial properties. Antioxidants are an important part of maintaining a healthy and balanced lifestyle, and basil maybe a very important source of these essential compounds. However, despite these reputed properties, it is important to be aware that basil contains estragole, which may be carcinogenic. In Germany, for example, basil is not considered safe for pregnant women or children. There is extensive diversity in the phytochemical constituents of basil; these constituents vary significantly with time, cultivation processes and storage. The nutritional and pharmacological properties of the whole herb in natural form, as it has



been traditionally used, results from the interaction of many different active phytochemicals, and consequently, the overall benefits of basil cannot be completely duplicated using single isolated constituents. There is very little data relating to a standardized dosage available from traditional practitioners, which is problematic for chemists and pharmacists. This raises the issue that there needs to be a greater communication between traditional and orthodox medicine in order to improve our understanding of the interactions and properties of basil (Al-Maskri et al., 2012).

In the last decade or two, an increased methodical interest in the health benefits of plant phytochemicals (in herbs and spices, vegetables and fruits) has gained prominence in the wider study of plant-based nutritional research. Although the study of plant compounds is by no means a new area of research, scientists have only recently started to characterize bioactive compounds in order to explore their effects on human health and disease. In animal and cell culture studies, basil has displayed anti-inflammatory, antidiabetic, antimicrobial, antioxidant and anticancer activity (Al-Maskri et al., 2012).

### **Culinary uses**

Basil has been incorporated into culinary preparations for thousands of years, and it is a very useful gastronomic herb found in a wealth of dishes, sauces and condiments, soups, stews and stuffing, and also in fish, meats and vegetables. It is easily blended with other herbs, including, garlic, oregano, mustard, parsley, pepper, rosemary and thyme. It is also an important constituent in teas, oils, cheeses and liqueurs. Basil is an important component of many alcoholic beverages, including bitters, liquors and spirits. By adding a blend of mixed essential oils of fennel, basil and coriander to a salt solution of whey, Russian researchers found a method to enhance the storage of a carbonated fermented milk beverage. Fresh, frozen or dried basil (1–40 g/l) is also used in spirits, garlic or lemon alcoholic beverages, which may be sweet or dry, according to a German patent. Basil essential oil has significant commercial value. It is utilized in a range of industrial products, including beverages, prepared foods, dental products, fragrances and soaps. *O. gratissimum* and *O. basilicum* essential oils are considered economic materials in their own right (Al-Maskri et al., 2012).

### **Use as a prophylactic agent**

Basil is highly beneficial in healing wounds, cuts and ulcers, and in removing parasites and worms. It supplies numerous antioxidants and offers generous reinforcement against free radical induced damage. Oxygen free radicals are naturally occurring physiological products containing one or more unpaired electrons, and along with reactive oxygen species (ROS), are considered to be harmful to important membrane lipids, proteins, carbohydrates and DNA. This damage has been related to several diseases, for example atherosclerosis, liver cirrhosis, cancer and diabetes, etc. It has been well accepted that dietary antioxidants have great prospects for curing these disease processes. Antioxidants also enhance the activity of superoxide dismutase (SOD) and reduce lipid. Basil antioxidants help in maintaining good health and in preventing the chance occurrence of heart diseases, as well as most of the other degenerative diseases, because oxidative stress is the hallmark of such diseases (Al-Maskri et al., 2012).

### **Anticancer activity**

The anticancer activity of basil has been long established and is mentioned by several investigators. Protection against cancer at the cellular level is provided by the unique array of flavonoids that are found in basil. Water-soluble flavonoids of basil, including vicenin and orientin, have been shown to defend cell structures and chromosomes against radiation and oxygen-based damage in human white blood cell studies. Basil leaf alcoholic extracts have a modulatory impact on carcinogen metabolizing enzymes such as aryl hydrocarbon hydroxylase and glutathione-S-transferase, (GST) and the cytochromes P450 and b5. They are important DE toxicants of mutagens and carcinogens. Basil anticancer activity has also been established against human fibro sarcoma cell cultures, in which alcohol extracts induced cytotoxicity at 50 mg/ml and above. Morphologically, the cancer cells showed condensed nuclei and shrunken cytoplasm and the DNA was found to be fragmented on agarose gel electrophoresis. Basil considerably decreased the occurrence of 3'-methyl-4-dimethylaminoazobenzene-induced hepatomas in rats and benzo( $\alpha$ )pyrene-induced neoplasia of the forestomach of mice. An alcohol extract of basil leaves was shown to have an inhibitory effect on chemically induced skin papillomas in mice. A leaf extract of basil

applied to 7,12-dimethylbenz(a) anthracene (DMBA)-induced papillomas in mice considerably reduced tumour incidence, the average number of papillomas per mouse and the cumulative number of papillomas. Eugenol, a flavonoid present in basil and other plants showed similar activity. Oral treatment with basil fresh leaf paste inhibits the early events of DMBA-induced buccal pouch carcinogenesis. Basil leaf extract suppresses or blocks the events related to chemical carcinogenesis by hindering the metabolic activation of the carcinogen (Al-Maskri et al., 2012).

### **Radio protective activity**

The flavonoids vicenin and orientin from basil leaves exhibited a greater radio protective effect than synthetic radio protectors by protecting human lymphocytes from the lactogenic effects of radiation at low, non-toxic dilutions.<sup>9</sup> Among three plant extracts, viz. *Withania somnifera* (L.) Dunal, *O. tenuiflorum* and *Plumbago rosea* (preferred name *P. indica* L.), tested on experimental mice for bone marrow survival following 2 Gy  $\gamma$ -radiation, *O. tenuiflorum* water extract exhibited maximum radioprotection as measured by an exogenous spleen colony forming unit (CFU-S) assay. (Al-Maskri et al., 2012).

### **Antimicrobial activity**

It is the volatile/essential oils of a hydrophobic nature that account for the biochemical actions of spices and herbs.<sup>7</sup> Basil contains many aromatic essential oil compounds that fluctuate in proportion and quality depending on the cultivar. The important aromatic compounds present include linalool, eugenol, citral, methyl chavicol/estragole, limonene, and methyl cinnamate. These aromatic compounds defend the herb from insects, bacteria and fungi. In similar fashion, they can help in protecting against diseases caused by fungi, bacteria and insects. Basil is also a well-recognized insecticidal, antiviral and antifungal agent. Although it has long been used to treat microbial infections, there is not sufficient data to fully support its efficacy and safety in humans. Basil has potent antimicrobial activity against *P. aeruginosa*, *Bacillus pumilus* and *B. megaterium*, *S. aureus* and *S. albus*, *M. tuberculosis*, *Micrococcus pyogenes* var. *aureus*, *Helminthosporium* spp., *H. oryzae*, *Alternaria tenuis*, *A. solani*, *Curvularia* spp. and *C. penniseli*, *Candida*

*guillermondii*, *Pseudomonas* spp., *S. aureus*, *Fusarium solani*, *Colletotricum capsici*, *Arthrobacter globiformis*, *E. coli* and *Vibrio cholerae*. The high concentrations of linolenic acid in basil oil are considered to be largely responsible for its antimicrobial activity (Al-Maskri et al., 2012).

### **Antioxidant activity**

The unique health benefits of basil are primarily due to its very high antioxidant content, and the antioxidants (e.g. phytochemicals such as phenolics and vitamins) that it contains contribute to disease prevention. The principal subtype of basil phenolics is its flavonoids, which include orientin and vicenin; and the plant also contains eugenol and anthocyanins. The presence of anthocyanins in purple basil is responsible for their deep red–violet pigmentation. All of the cultivars of purple basil contain very high antioxidant activity due to their anthocyanin content (Al-Maskri et al., 2012).

Despite being consumed at relatively low amounts, the high levels of antioxidants and minerals in herbs mean that many of them have significant health benefits. It is not fully understood what quantities of basil should be ingested to achieve its health benefits, There are no standards or recommendations as to the precise amounts to use. Nevertheless, basil is almost completely calorie free and contains high quantities of dietary fiber and minerals. Even though there appears to be no logical evidence for its usefulness to human health, basil tea and oil are readily available in many health food stores. Having said that, basil is a popular food additive and provides a distinctive flavor and aroma. Basil is a great addition to any kitchen; it adds both flavor and personality to many dishes (Al-Maskri et al., 2012).

## 1.4 Literature review

### Previous Studies

Characterization of the essential oil composition of 19 accessions of Basil; all were collected as seeds and grown in at the university of Gezira farm. The essential oil content varied from 0.33 to 0.47% in fresh leaves and from 0.13 to 0.4% in fresh flowers. The essential oil components were separated and /or identified by TLC, GC and GC-MS (Lemin 2005).

Testing of the essential oils of four accessions of basil grown in Sudan, they were selected and tested for *Anopheles* larvae. Malaria is the major health problem in the Sudan and the whole country and *Anopheles* mosquito is the major vector of malaria disease in Sudan. To determine the toxic effects of basil essential oils extracted by steam distillation against *Anopheles* larvae. The active ingredients were separated and identified by TLC, IR and GC-MS. Linalool, geraniol and eugenol are active components of basil essential oil against *Anopheles* larvae (Michael et al., 1999)

This paper discussed the variability in essential oils content and chemical constituents of the aerial plant parts of the basil. The essential oils were hydro distilled from the aerial parts, flowers, leaves, stems, using Clevenger apparatus. The chemical constituents of the essential oils were determined by Gas Chromatograph-Mass Spectrometry (GC-MS). The results revealed that the essential oil content varied with a range of 0.29% to 0.33% for flowers and 0.32% to 0.48% for leaves. As usual, the content of essential oils was higher in leaves than in flowers (Abdalla et al., 2012).

This study reviews primarily the topic of basil essential oils with regards to their chemical composition, their effect on microorganisms, the test methods for antimicrobial activity determination, and their possible future use in food preservation or as the active (antimicrobial), slow release, and component of an active package. The techniques represented in this category include diffusion methods, Dilution methods and Micro-atmosphere method. The contradictory conclusions reached in the early studies on the antimicrobial activity of basil essential oils are not surprising. Poor solubility and high volatility often preclude the application of traditional antimicrobial assays (Lemin 2005).

This study shows that the Acaricidal activity of essential oils extracted from cumin seeds (*Cuminum cyminum*), allspice berries (*Pimenta dioica*) and basil leaves (*Ocimum basilicum*) were tested on 10-day-old *Rhipicephalus (Boophilus) microplus* tick larvae using the LPT. Basil essential oil was not shown to be toxic against *R. microplus* larvae. The most common compounds detected by gas chromatography-mass spectrometry were as follows: cumin: cuminaldehyde (22.03%),  $\gamma$ -terpinene (15.69%) and 2-carene-10-al (12.89%); allspice: methyl eugenol (62.7%) and eugenol (8.3%); basil: linalool (30.61%) and estragole (20.04%) (Lemin 2005).

In this study they characterized the yield of the essential oil and chemical profile of a new basil variety, namely 'Mánes'; then compared qualitative and quantitative composition and extraction yields of seven sweet basil varieties including 'Mánes'; and evaluated an influence of greenhouse conditions of the cultivation in different seasons of one year on the extraction yield and chemical profile of essential oil of seven sweet basil varieties including 'Mánes'. Separation and identification of particular essential oil components in the seven studied basil varieties was carried out by means of the GC-MS method developed for this purpose (for the optimum GC-MS conditions see Section. Chemotypes of the studied varieties were evaluated by quantification of four main basil essential oil constituents (i.e., eucalyptol, linalool, estragole, and eugenol) and comparing the obtained results with data presented in the literature (Abdalla et al., 2012).

The nutrient profile of seeds of selected medicinal plant (sweet basil and psyllium) was determined. The seeds of selected medicinal plants were analyzed using the standard methods (AOAC, 2000). The seeds of sweet basil have sufficient amount of protein and mineral matter as compared to psyllium seeds. The results of this study indicated that seeds of psyllium are being abundant in energy values whereas excellent source of fiber too as compared to sweet basil (Finar 2006).

This study shows that the basil seeds are used not only as pharmaceutical plant but also for culinary purpose. The current study has been undertaken to develop a nutritious, healthy and value added drink.

Proximate, mineral analysis, total polyphenol content and mineral analysis of basil seeds was conducted. Result revealed that basil seeds are not only good source of fiber and protein but they provide appreciable amount of minerals and phenolic compounds. And the paper was concluded that basil seed could be supplemented in different food products for the preparation of value added, healthy and nutritious diets. (Lemin 2005).

This study studied five subspecies in coastal plains of Albania (Toshkëz-Lushnja) is presented in this paper. This study encompassed the five types of basil. The seeds have been brought from Italy, The experiment was set up according to the randomized block scheme, with five variants and four repetitions with variant size of 28m<sup>2</sup> (2.4 m x 11.7 m). The experiment was set up in Toshkez - Lushnja, according to randomized block scheme, as the methodology had provided. The chemical composition of the soil is: humus 0.7%, pH 7.55, nitrogen 0.18%, phosphorus 17.7 %, potassium 12.5 ppm and calcium 9.07 ppm (Balandrin et al., 1985).

The objective of this study was to determine whether reflection from the different colors could influence concentrations of volatile compounds emitted from the fresh leaves. Volatile compounds were isolated by headspace sampling and quantified by gas chromatography. Twenty-six compounds were identified, of which the terpenoids linalool and 1, cineole comprised more than 50% of the total yield. Concentrations of volatile compounds from leaves that developed over green, blue, yellow, white, and red mulches followed the same patterns as they did for air-dried leaves of the same cultivar. However, the concentration of volatile compounds from fresh leaves was about 50-fold higher than those found in the previous study of air-dried leaves (Al-Maskri et al., 2012).

This study shows that the outbreak *S. Senftenberg* isolate attached to a variety of salad leaves (including basil, lettuce rocket and spinach) and that flagella played a major role in bacterial leaf interaction. In contrast, although abundant flagella were seen linking *S. Typhimurium* to basil leaf surface, deletion of the gene for the phase-1 flagellin *fliC* had no measurable effect on the level of leaf association. Leaf contamination can occur during crop growth (for example, through contaminated water, wild or domesticated animals, flies or birds), harvest, distribution, processing and packing or cooking. A better understanding of the mechanism

involved in the attachment of *S. enterica* to salad leaves would be useful in developing interventions to minimize contamination and transmission and in the development of accurate risk assessments (Abdalla et al., 2012).

This study reports on the determination of 11 elements in 33 medicinal plants from Sudan and discusses a possible correlation between their curative effects and their trace elements content. A total of 11 elements (cadmium, lead, mercury, tin, copper, iron, manganese, zinc, chromium, selenium and magnesium) were determined using inductively coupled plasma (ICP)- optical emission spectrometry (ICP-OES), ICP-sector fieldmass spectrometry (ICP-sf-MS) and hydride generation (HG)-ICP-OES techniques. The results of the present study showed no heavy metal accumulation in any of the plants. Cd, Pb, Hg and Sn were found only in trace concentrations significantly below the global limits. This indicates the possibility of a safe use of this medicinal plants (Abdalla et al., 2012).

Analysation the contents of macro and micro elements in the herbs of two basil cultivars ('Kasia' and 'Wala'), depending on the doses of calcium carbonate – 6 and 12 g·dm<sup>-3</sup> substratum (Abiola et al., 2016)

In this study, high performance liquid chromatographic (HPLC) and flow-injection mass spectrometric (FIMS) fingerprinting techniques were used to differentiate organic and conventional sweet basil leaf samples. Principal component analysis (PCA) of the fingerprints indicated that both HPLC and FIMS fingerprints could effectively detect the chemical differences in the organic and conventional sweet basil leaf samples. This study suggested that the organic basil sample contained greater concentrations of almost all the major compounds than its conventional counterpart on a per same botanical weight basis. The FIMS method was able to rapidly differentiate the organic and conventional sweet basil leaf samples (1 min analysis time), whereas the HPLC fingerprints provided more information about the chemical composition of the basil samples with a longer analytical time (Abdalla et al., 2012).

This study shows that the Thermal Treatment on Chemical Structure of B-Lacto globulin and Basil Seed Gum Mixture at Different States by ATR-FTIR Spectroscopy. Attenuated total reflection Fourier transform



infrared spectroscopy was used to compare the structure of  $\beta$ -lactoglobulin, basil seed gum, and  $\beta$ -lactoglobulin-basil seed gum mixtures, at different states (powder, solution, and gel). The effects of heating and different ratios of  $\beta$ -lactoglobulin-basil seed gum were also investigated to determine their impact on chemical structure and understand their interaction. The results showed that gelification process proved a pronounced effect upon  $\beta$ -lactoglobulin secondary structure, leading to the formation of intermolecular hydrogen-bonding  $\beta$  sheet structure. These results confirmed that this structure may be necessary for the formation of a gel network. Basil seed gum had a distinct peak at around  $1603\text{ cm}^{-1}$  that relates to  $-\text{COO}-1$  stretching of carboxylate salts, probably uronic acids, which approved its anionic structure. The Fourier transform infrared spectroscopy findings strongly suggested that these two polymers are thermodynamic incompatible as amide I peak was increased in the  $\beta$ -lactoglobulin-basil seed gum mixed system and carbon–nitrogen (CN) stretching peak was observed at  $2125\text{ cm}^{-1}$ . On the basis of these findings, it was possible to modify the ability of  $\beta$ -lactoglobulin-basil seed gum to form a gel and as a consequence to control the gelling and emulsifying properties (Abdalla et al., 2012).

This study they determine an antioxidant activity of a methanolic extract of *Ocimum basilicum L.* (sweet basil) was examined using different in vitro assay model systems. The DPPH scavenging assay system and the oxidation of the *soy phosphatidylcholin* liposome model system were used to evaluate the antioxidant activity of each fraction. Fraction IV showed the strongest activity followed by fractions V and VI. Phenolic compounds responsible for the antioxidative activity of the fractions were characterized by atmospheric pressure chemical ionization liquid chromatography-mass spectrometry. The major antioxidant compound in fraction IV was confirmed as rosmarinic acid by  $^1\text{H}$  NMR and characteristic fragmentations in the mass spectrum. The results showed that one rosmarinic acid can capture 1.52 radicals, and furthermore, the existence of a synergistic effect between R *tocopherol* and rosmarinic acid was revealed (Abdalla et al., 1982).

### **1.5 Objectives of the research**

This study was under-taken to know the constituent of the leaves and mineral analysis of basil leaves through proximate analysis. Then extraction of essential oil from basil and analysis by GC-MS mass.

# **CHAPTER TWO**

## *Materials and Methods*

## Materials and Methods

### 2.1 Materials

#### 2.1.1 Collection of samples

- i.
  - Latin Name: *Ocimum basilicum L.*
  - Arabic Name: sweet basil
  - Family: Lamiacea.
- ii.
  - Latin Name: *Ocimum basilicum charmandaschricum*
  - Arabic Name: carnation basil
  - Family: Lamiacea.

Plants obtained from the Medicinal and Aromatic Plant and Traditional Medicine Research Institute [MAPTMRI].

#### 2.1.2 Experimental Site

The experiment was laid out at the experiment field of the Medicinal and Aromatic plant and Tradition Medicine Research Institute (MAPTMRI), Shambat, during winter season Nov-2017 to May-2018. Shambat is situated in Khartoum Bahri in Northern part located at North latitude of 15 to 40 and 32 to 32 East longitudes and an altitude of 280 meters above mean sea level. This region falls under agro-climatic zone in Khartoum state.

#### 2.1.3 Harvest stage:

Plant sowing date at 11.1.2020 in winter season, in stage 50% flowering, leaves harvested and weighted using sensitive balance

#### 2.1.4 Plant Materials:

Two spice (species) plants of Basil (*Ocimum basilicum L* & *Ocimum basilicum charmandaschricum*) were collected from Shambat, during winter season Nov–2017 to May–2018. Shambat is situated in Khartoum Bahri in Northern part located at North latitude of 15 to 40 and 32 to 32 East longitudes and an altitude of 280 meters above mean sea level.

## 2.2 Chemicals (All chemicals are of analytical grade)

- Petroleum ether (boiling point (35- 60) C.
- Ethyl alcohol (95%) .
- Hydrochloric acid (4N, 0.1 N, 2%).
- Diethyl ether.
- Sulfuric Acid.
- Boric Acid (4%).
- Sodium Hydroxide (50%).
- Sulfuric Acid Solution (0.225N).
- Sodium Hydroxide Solution (0.313N).
- Potassium Sulfate Solution (10%).

## 2.3 Instruments

- Atomic absorption spectrometer (model: 210 vap, made in USA 2005).
- GC/MS (model QP2010- Ultra, Shimadzu Company, Japan).
- Analytical Balance.
- Hot air oven.
- Desiccator.
- Drying oven..
- Magnetic stirrer.
- Furnace.
- Water Bath.

## 2.4 Methods of analysis

### 2.4.1 Ash Content

2.00g was weighed in a silica crucible and heated in muffle furnace for 6 h at 500 C. The crucible was cooled for 30 min in a desiccator and weighed. It was heated again in the furnace for half an hour, cooled and weighed. The process was repeated till the weight was constant. The ash content was calculated according to the formula below.

$$\text{Ash, g per 100 g} = \frac{(W_3 - W_1) \times 100}{(W_2 - W_1)}$$

Where:

$W_1$  = weight of crucible.

$W_2$  = weight of crucible + sample.

$W_3$  = weight of crucible + ash.

### 2.4.2 Mineral Content

For mineral analysis, 2.00 g each of the samples was dried and placed in a porcelain crucible. Then was placed in a cool muffle furnace and ash at 500 C overnight. The crucible was cooled in a desiccator and the ash was dissolved in 5 mL of 20% HCL, the solution was warmed and the residue was dissolved. And then filtered into a 50ml volumetric flask. The content was made up to mark with deionized water and stored until analyzed for mineral contents using Atomic Absorption spectrophotometer (AAS).

$$\text{mg/kg} = \frac{R \times V}{W}$$

$$\text{g/mg} = \frac{R \times V \times 100}{10^6 \times W}$$

While:

R = reading of device.

V = volume.

W = weight of sample

### 2.4.3 Water Content

Water was determined by oven drying method. The container placed in the drying oven at 100 until constant weight (1–2h) then cooled in a desiccator for 30 min and weighted ( $w_1$ ).

$$\text{Water (g/100g)} = \frac{(W_2 - W_3) \times 100}{(W_2 - W_1)}$$

$$\text{Total solid (\%)} = 100 - \% \text{ water (W/W)}$$

Where:

$W_1$  = weight of container.

$W_2$  = weight of container + sample before drying (g).

$W_2 - W_1$  = weight of sample (g).

$W_3$  = weight of container + sample after drying (g).

$W_2 - W_3$  = loss of weight (g).

#### 2.4.4 Fat Content

2.00 g of dried sample placed in 250 ml extraction tube ( $w_1$ ) then 2ml from alcohol was added, the solution was stirring. 10 mL of the diluted HCL 4N was added and mixed well, the flask was set on the heater and reflexed for 30 min then the tube was placed in water bath at (70–80C) , then was stirring until the sample was completely hydrolyzed (30–40min).

After that 10 mL of alcohol was added and cooled, and then 25 mL of diethyl ether were added in 3 portions, thereafter closed the tube and shacked vigorously for 1min. 25 mL of petroleum ether was added and the solution was shacked again vigorously for 1min. The solution of ether-fat was transferred into a pre- s weighed flask 125 mL by filtering it through a funnel containing a plug of cotton packed firmly in the stem part, then allowed of free passage of ether into the flask.

Before weighing the flask was dried it in drying oven at (100C) then cooled in a desiccator and weighted ( $w_2$ ).Then repeated extraction of the liquid sample remaining in tube twice and used the same solvent. In each time the clear ether solution was transfer through the same funnel into the same flask. The solvents were evaporated completely on a water bath at (70–80C), the fat was dried in an oven at (1000C) until constant weight was obtained .Finally the flask was cooled in a desiccator and weighted ( $w_3$ ).

$$\text{Total Fat (g/100g)} = \frac{(W_3 - W_2) \times 100}{W_1}$$

Where:

$W_1$  = weight of sample.

$W_2$  = weight of flask before fat extraction.

$W_3$  = weight of flask after fat extraction.

#### **2.4.5 Estimation of Crude Protein by Keldahl Method**

The Dissolved sample was placed in a room temperature and mixed well, then added 6.00 g from sample weighted into the digestion tube, after that 6.00 g from catalyst was added then 15 mL from sulfuric acid was added. The digestion tube was placed in the digest initially at low temperature (to prevent frothing) and boiled briskly until the solution became clear and the oxidation was completed then continued the digestion until a clear digest obtained. after the liquid became clear heated the solution to complete breakdown of all organic matter. 750 mL was placed in elementary flask containing 25 mL from 4% boric acid with indicator as receiver on the distillation unit. Then 100 mL of water and 35 mL of sodium hydroxide (50%) were added to the digests and distillation started. After that Titrated the distillation with standardized hydrochloric acid (0.01N) until the color changed to pink then recorded the volume of acid.

$$N(\text{g}\%) = \frac{(\text{ml } 0.1\text{N HCL sample} - \text{mL } 0.1\text{N HCL blank}) \times 0.0014 \times N \text{ HCL} \times 100}{\text{weight of sample}}$$

Protein (g per 100g) = % total nitrogen x appropriate nitrogen conversion factor.

#### **2.4.6 Estimation of Crude Fiber**

2.00 g of fat free material of each sample was transferred to digestion flask 700 mL with added 0.5 g from Asbestos to regulate the boiling process then treated with 200 mL of  $\text{H}_2\text{SO}_4$  0.255N and the mixture was boiled for 30 min. After filtration and washing with hot distilled water, the residue was treated and boiled with 200ml of the solution NaOH 0.313N. The filtrate was washed with hot  $\text{H}_2\text{SO}_4$ , water and alcohol. The residue was ignited and the ash weighed. Loss in weight gave the weight of the crude fiber.



### **2.4.7 Hydro Distillation**

Oil distillation using Hydro distillation for herb Hundred grams of clean dry herb were placed into a flask; in Clevenger apparatus 1000 mL volume scale, then added 500 mL distilled water and the compound was boiled at 100 C the heat decreased to 70 C and concentrated by vacuum distillation for 4 hours, the oil condensed in Clevenger receiver and later the distilled oil was isolated and placed into dark bottle.

### **2.4.8 Detection of Chemical Constituents Isolated by GC/MS**

The qualitative and quantitative analysis of the sample was carried out by using GM/MS technique model (GC/MS-QP2010-Ultra) from Japans' Simadzu Company, with serial number 020525101565SA and capillary column (Rtx-5ms-30m×0.25 mm×0.25µm).

The sample was injected by using split mode, instrument operating in EI mode at 70eV. Helium as the carrier gas passed with flow rate 1.69 ml/min, the temperature program was started from 50 C with rate 7 C/min to 180 C then the rate was changed to 10 C/min reaching 280 C as final temperature degree, the injection port temperature was 300 C, .the ion source temperature was 200 C and the interface temperature was 250 C. The sample was analyzed by using scan mode in the range of m/z 40-500 charges to ratio and the total run time was 28 minutes .Identification of components for the sample was achieved by comparing their retention index and mass fragmentation patents with those available in the library, the National Institute of Standards and Technology (NIST).

# **CHAPTER THREE**

## *Results and Discussion*

### 3. Result and Discussion

The proximate composition of nutritive contents of Basil is depicted in Table(3-1). All the samples have relatively high water content when compared to ash, crude protein, crude fat, and crude fiber. The leaves of *Ocimum basilicum L.* have the highest water content with a value of (87.140%) fresh weight while *Ocimum basilicum charmandaschricum* has the least amount of water (80.463%). Ash content was highest in leaves of *Ocimum basilicum charmandaschricum* (3.904%) while the leaves of *Ocimum basilicum L.* recorded the least percentage (3.009%). There was a wide variation in the amount of crude protein among the extracts of the samples, the maximum amount of crude protein was found in the leaves of *Ocimum basilicum charmandaschricu.* (4.375%) while the minimum was recorded in the leaves of *Ocimum basilicum L.* (3.500%). The crude fat was maximum in *Ocimum basilicum charmandaschricu.* (0.638%) and the least content was found in the leaves of *Ocimum basilicum L.* (0.406%). The maximum amount of crude fiber was found in the leaves of *Ocimum basilicum charmandaschricu.* (9.620) while the minimum amount was found in the leaves of *Ocimum basilicum L.* (5.880%).

The crude protein content (%) of *Ocimum basilicum L.* and *Ocimum basilicum charmandaschricu* were (3.5%, 4.38%) respectively are lower than those (7.00%) reported by (Idiong and Isong 1997), (29.78%) reported by (Akindahunsi and Salawu 2005) and (23.74) (A. B. Abidi et al., 2006).The ash content (in %) of *Ocimum basilicum L.* and *Ocimum basilicum charmandaschricu* were (3.01%, 3.9%) respectively which is lower than (4.84%) reported by (Ahmad et al., 2009) and the values of the leaves(22.84%) reported by (A. B. Abidi et al., 2006) and (15.09%) with (Anita et al., 2006)The moisture content (in %) values for the leaves of *Ocimum basilicum L.* and *Ocimum basilicum charmandaschricu* were( 87.14%, 80.46%) respectively which is higher than the values reported for the leaves of *Xylophia aethiopia* (16.04%) (Abolaji et al., 2007). The values of the crude fat (in %) for the leaves of *Ocimum basilicum L.* and *Ocimum basilicum charmandaschricu* were (0.406%, 0.638%) respectively are lower in amount when compared to those (4.8%) which reported by (Akindahunsi and Salawu 2005) and (3.15%) with (Abolaji et al., 2007).The values of the fiber (in %) for the leaves of *Ocimum*

*basilicum L.* and *Ocimum basilicum charmandaschricu* were (5.88%, 9.62%) respectively the crude protein, Ash, Fat contents (%) of *Ocimum basilicum L.* and *Ocimum basilicum charmandaschricu*. are smaller than the contents reported by (Alkherraz and Mlitan2014) ,due to the Leaves of *Ocimum gratissinum* collected from three different locations in Misurata region (Zaroge, Tamena and Daphnia) in Libya, the percentage of protein, Fat, Ash were (9.10 ,9.8,9.22),(10.80,11.0,11.16), (14.3,14.5 13.9) respectively. But the moisture are higher than in the contents of Z,T,D (10.6,10.4,10.6) and that due to the Leaves were allowed to dry in open air in the shade area for 30 days (Oscar et al., 2004) and (A. U. Eka et al., 1998).

**Table (3.1): Nutritive compositions of the basil plant samples:**

| Samples                                   | Ash % | Moistures% | Protein% | Fiber% | Fat%  |
|---|-------|------------|----------|--------|-------|
| <i>Ocimum basilicum L.</i>                | 3.009 | 87.140     | 3.500    | 5.880  | 0.406 |
| <i>Ocimum basilicum charmandaschricum</i> | 3.904 | 80.463     | 4.375    | 9.620  | Fat%  |

The results of proximate analysis on these two kinds showed that could be good for health by providing most of the essential nutrients for normal body functions when consumed in appropriate combinations. The mineral composition in (mg/100g) of *Ocimum basilicum L.* and *Ocimum basilicum charmandaschricum* are shown in Table (3-2). Different parts of the plants contain minerals like Ca, Mg, Mn, K, Na, Ni, Cu, Zn and Fe in varying concentration. Potassium having the highest concentration in the *Ocimum basilicum L.*, the results indicated a maximum value in in the leaves of *Ocimum basilicum L.* (0.112 %) and a minimum value in the leaves of *Ocimum basilicum charmandaschricum* (0.057%).

Calcium was high in *Ocimum basilicum charmandaschricum* (0.089%) and low in *Ocimum basilicum L.*(0.045%), the high calcium level may be explained by variations in cultivation conditions and location.

This study indicated a higher amount of Sodium content in *Ocimum basilicum charmandaschricum* which is (0.107%), nevertheless, the *Ocimum basilicum L.*with a value (0.053%). Basil were found to contain a large amount of Mg, the results indicated a higher Mg content in *Ocimum basilicum charmandaschricum* (0.074%) while the *Ocimum*

*basilicum L.* with a value (0.0642%). The value of Iron in the leaves of *Ocimum basilicum L.* recorded the lower percentage (0.0094%) than *Ocimum basilicum charmandaschricum* (0.0121%). The value of Copper in the leaves of *Ocimum basilicum charmandaschricum* (0.00015%) is higher than the value of copper in the leaf of *Ocimum basilicum L* (0.00012%). Manganese was present in maximum concentration in the leaves of *Ocimum basilicum L.* and not detected on the leaves of *Ocimum basilicum charmandaschricum*.

Zinc contents of basil were found in small percentages in all the species analyzed. Maximum amount of zinc was present in the leaves of *Ocimum basilicum charmandaschricum* (0.0003%) and the least was observed in the leaf of the *Ocimum basilicum L.* (0.0002%) are low when compared to the mineral analyzed for in *Pilostigma thioningi* (70.1%) (A. Kowalski et al., 2012)

**Table (3.2): Elemental composition of the basil plant samples determined by AAS (mg/l)**

| Elements | Samples                    |   |
|----------|----------------------------|---|
|          | <i>Ocimum basilicum L.</i> | <i>Ocimum basilicum charmandaschricum</i> |
| Ca       | 0.045                      | 0.089                                     |
| Cu       | 0.00012                    | 0.00015                                   |
| Fe       | 0.0094                     | 0.0121                                    |
| Mg       | 0.0642                     | 0.0742                                    |
| Mn       | 0.00017                    | N.D                                       |
| Zn       | 0.0002                     | 0.0003                                    |
| Na       | 0.053                      | 0.107                                     |
| K        | 0.112                      | 0.057                                     |

The composition of the essential oils basil plants were determined by hydro distillation of leaves. The oil content (%) was expressed on a fresh weight basis. Table (3-3) shows the essential contents of the leaf of two type of basil. The value was (1.9%) for *Ocimum basilicum L* and colorless while *Ocimum basilicum charmandaschricum* was (2.3%) and have yellow color. The differences in relative percentage composition of

the essential oils can be explained by the differences in construction of both distillation apparatus and different extraction time.

**Table (3.3): volatile oil content (%) of the fresh leaf of two types of basil**

| Type of Basil                            | volatile Oil % |
|--|----------------|
| <i>Ocimum basilicum L.</i>               | 1.9            |
| <i>Ocimumbasilicum charmandaschricum</i> | 2.3            |

The identity and quantity of particular components of the basil volatile oil were evaluated by the GC-MS analysis method. Table (3.4) shows the analysis of the volatile oil from the leaves of *Ocimum basilicum L* has been detected 29 constituents, the major's content represented 98.12% from 100%.

Moreover, table (3.5) shows the analysis of the volatile oil from the leaves of *Ocimum basilicum charmandaschricum* and it has been detected 9 constituents, the major's content represented 90.48% from 100% in table (3.5).

**Table (3.4): GC-MS analysis (%) composition of volatile oils of *Ocimum basilicum L.***

| ID | Name                             | Ocimum basilicum L |          |        |
|----|----------------------------------|--------------------|----------|--------|
|    |                                  | R. Time            | Area     | Area % |
| 1  | alpha.-pinene                    | 4.138              | 14.2770  | 0.19   |
| 2  | Camphene                         | 4.390              | 26276    | 0.04   |
| 3  | .beta.-pinene                    | 4.797              | 102903   | 0.14   |
| 4  | beta-Myrcene                     | 4.858              | 336110   | 0.45   |
| 5  | Eucalyptol                       | 5.843              | 573951   | 7.65   |
| 6  | 1,3,7-octatriene                 | 6.150              | 639686   | 0.85   |
| 7  | 1,6-octadien-3-ol,3,7,-dimethyl- | 7.198              | 8114174  | 10.81  |
| 8  | (+)-2- Bornanone                 | 8.086              | 479001   | 0.64   |
| 9  | L-.alpha.-Terpinol               | 9.084              | 386282   | 0.51   |
| 10 | Estragole                        | 9.262              | 35314712 | 47.06  |
| 11 | Acetic Acid                      | 10.838             | 319103   | 0.43   |

Continue

|    |   |        |          |        |
|----|---|--------|----------|--------|
| 12 | gamma- Elemene                                    | 11.793 | 213634   | 0.28   |
| 13 | 2-Oxabicyclo                                      | 11.909 | 216534   | 0.29   |
| 14 | Phenol,2-methoxy-4-(2-propenyl)-<br>,acetate      | 12.441 | 2134771  | 2.84   |
| 15 | Cyclohexene,1-ethenyl-1-methyl-<br>2,4-bis        | 12.834 | 764345   | 1.02   |
| 16 | Methyl eugenol                                    | 13.242 | 612641   | 0.82   |
| 17 | Bicyclo   | 13.364 | 322263   | 0.43   |
| 18 | Bicyclo [3.1.1] hept-2-ene,2,6-<br>dimethyle-6-   | 13.615 | 9886339  | 13.17  |
| 19 | Humlene   | 13.987 | 287244   | 0.38   |
| 20 | 1-H-<br>cyclopenta[1,3]cyclopropa[1,2]be<br>nzene | 14.156 | 195363   | 0.26   |
| 21 | 1,6cyclodecadiene,1-methyl-5-<br>methylene        | 14.487 | 640524   | 0.85   |
| 22 | Cis-.beta.-farnese                                | 14.504 | 457583   | 0.61   |
| 23 | 1,5-cyclodecadiene,1,5-dimethyl                   | 14.754 | 595129   | 0.79   |
| 24 | Azulene,1,2,3,5,6,7,8,8a,-<br>octahydro-          | 14.902 | 201765   | 0.27   |
| 25 | 1,4-din   |        |          |        |
| 26 | Gamma-muurolene-                                  | 15.054 | 1412236  | 1.88   |
| 27 | Cedrene   | 15.205 | 292686   | 0.39   |
| 28 | Cubenol   | 16.798 | 504472   | 0.67   |
| 29 | Tau.-Cadinol                                      | 17.22  | 4540689  | 6.05   |
| 30 | Phytol  | 23.190 | 170642   | 0.23   |
|    |   | -      | 75049228 | 100.00 |

**Table (3.5): GC-MS analysis (%) composition of volatile oils of *Ocimum basilicum charmandaschricum***

| I<br>D | Name  | <i>Ocimum basilicum<br/>charmandaschricum</i> |          |        |
|--------|---|---|----------|--------|
|        |   | R. Time                                       | Area     | Area % |
| 1      | Eugenol                                     | 12.442  | 14.2770  | 39.11  |
| 2      | Cyclohexane                                 | 12.720  | 26276    | 1.02   |
| 3      | Naphthalene                                 | 12.879  | 102903   | 24.21  |
| 4      | Caryophyllene                               | 13.408  | 336110   | 30.39  |
| 5      | Humlene                                     | 13.996  | 573951   | 1.54   |
| 6      | Naphthalene<br>2,3,4a,5,6hexahydro-1,4a     | 14.587  | 639686   | 0.61   |
| 7      | IR3Z,9s-4,11,11-Trimethyl-8-<br>methylenebi | 14.736  | 570019   | 0.66   |
| 8      | Caryophyllene oxide                         | 16.280  | 479001   | 1.85   |
| 9      | -1Naphthalenol,decahydro-<br>1,4a-dimethyle | 17.493  | 386282   | 0.61   |
|        |   | -   | 86676355 | 100.00 |

Eugenol was the major constituent of the essential oil of *Ocimum basilicum L* (39.11%) followed by Caryophyllene, Naphthalene, Caryophyllene oxide, and Humlene [30.39, 24.21, 1.85, 1.54] % respectively, they are represented in Table (3.6), Fig (3.9). While the major constituent of the essential oil of *Ocimum basilicum charmandaschricum* was Estragole (47.6%) followed by Bicyclo [3.1.1] hept-2-ene,2,6-dimethyle-6-,1,6-octadien-3-ol,3,7,-dimethyl-,Eucalyptol, Tau-cadinol, Phenol,2-methoxy-4-(2-propenyl)-,acetate, Gamma, and Cyclohexene,1-ethenyl-1-methyl-2,4-bis[13.17, 10.81, 7.65, 6.05, 2.84, 1.88, 1.02]% respectively and they represented in Table (3.7), Fig (3.10). additionally, the constituent% of the major contents of *Ocimum basilicum L* & *Ocimum basilicum charmandaschricum* demonstrated in Fig (3.11).

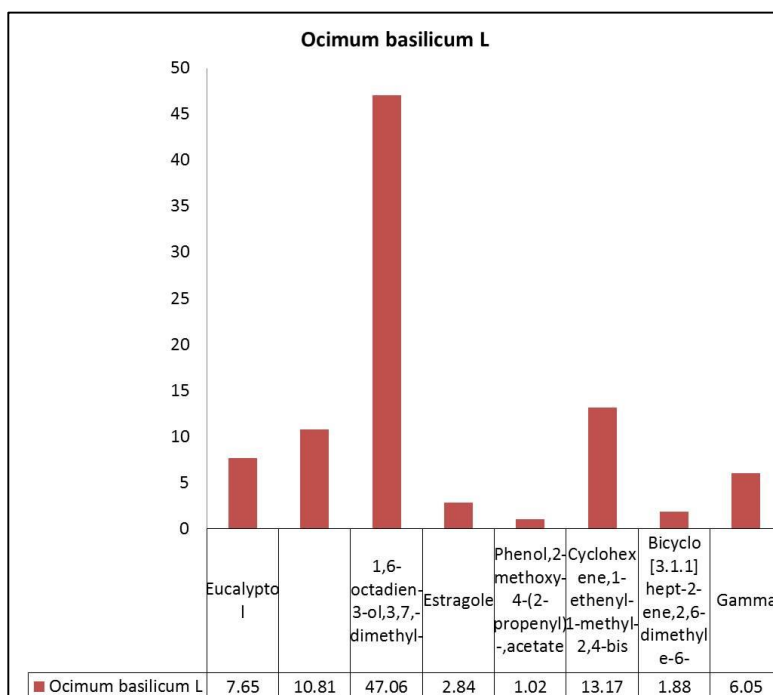
In other studies Eugenol was also found as major constituents of the basil oil with varied percentages (Abduelrahman et al., 2009), (Dagnaw et al., 2013). On the contrary, the constituents of essential oil hydro distilled from the aerial parts of *Ocimum basilicum* from Northern Ethiopia was examined by GC-MS (Michael et al., 1999). The major constituents



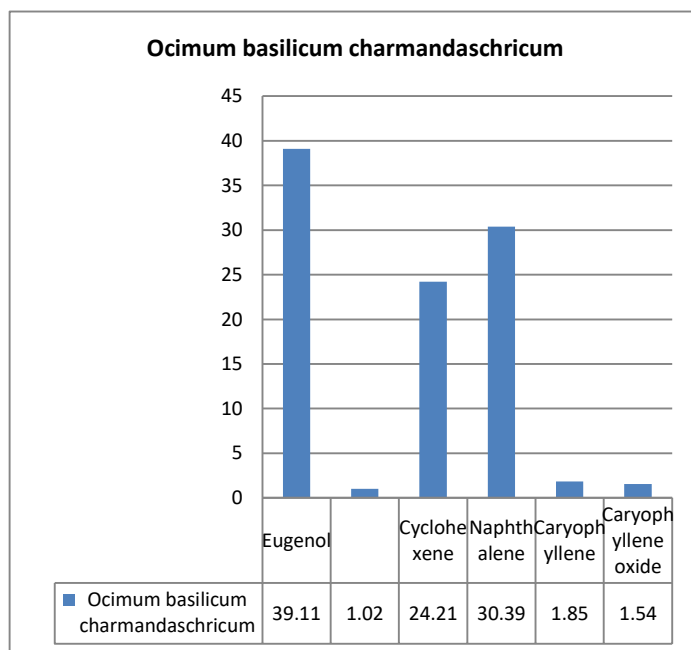
identified were copaene (25.5%), p-menth-2-en-1-ol (7.7%), eugenylacetate (4.8%). These results confirm the classification of *O. basilicum* from Sudan as linalool and eugenol chemo type reported by (Dagnaw et al., 2013).

**Table (3.6): The major constituent of content of two basil samples %**

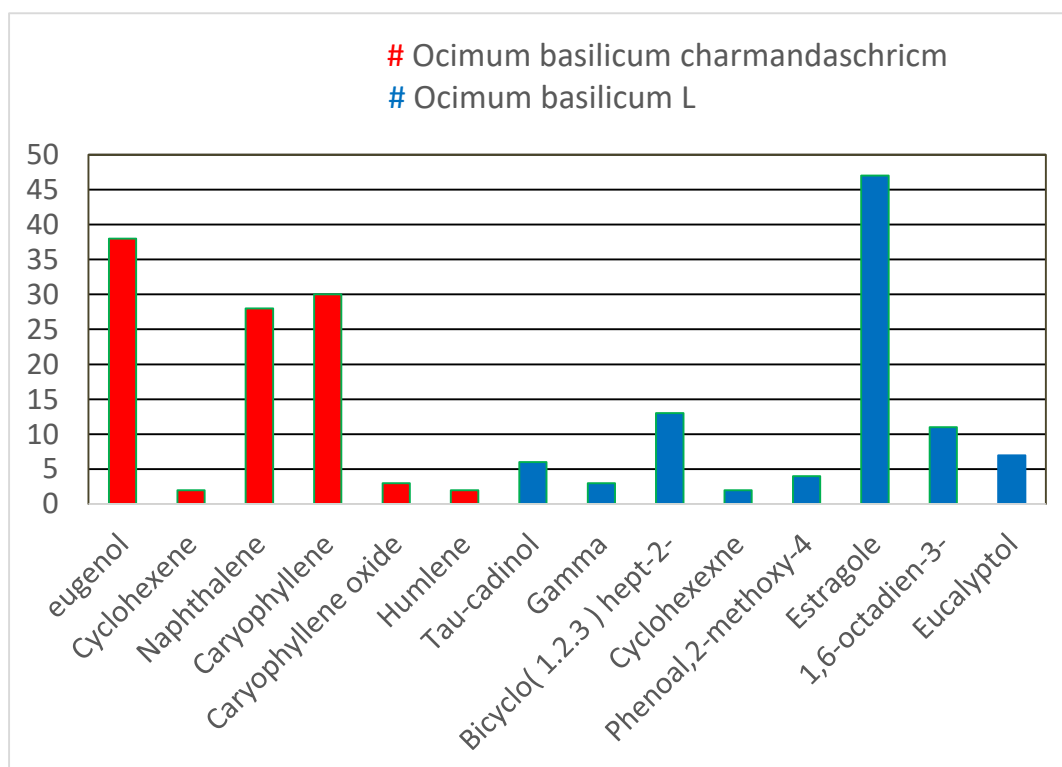
| ID | <i>Ocimum basilicum L</i>                   |              | <i>Ocimum basilicum charmandaschricum</i> |              |
|----|---|--------------|---|--------------|
|    | Name  | Constituent% | Name                                      | Constituent% |
| 1  | Eucalyptol                                  | 7.65         | Eugenol                                   | 39.11        |
| 2  | 1,6-octadien-3-ol,3,7,-dimethyl-            | 10.81        | Cyclohexene                               | 1.02         |
| 3  | Estragole                                   | 47.06        | Naphthalene                               | 24.21        |
| 4  | Phenol,2-methoxy-4-(2-propenyl)-,acetate    | 2.84         | Caryophyllene                             | 30.39        |
| 5  | Cyclohexene,1-ethenyl-1-methyl-2,4-bis      | 1.02         | Caryophyllene oxide                       | 1.85         |
| 6  | Bicyclo [3.1.1] hept-2-ene,2,6-dimethyle-6- | 13.17        | Humulene                                  | 1.54         |
| 7  | Gamma                                       | 1.88         | -   | -            |
| 8  | Tau-cadinol                                 | 6.05         | -   | -            |
|    |   | 90.48        |   | 98.12        |



**Fig no (3.9):** The chart demonstrates the constituent% of the major content of *Ocimum basilicum L*



**Fig (3.10):** The chart demonstrates the constituent% of the major content of *Ocimum basilicum charmandaschricum*.



**Fig (3.11): The chart demonstrates the constituent% of the major contents of Ocimum basilicum L & Ocimum basilicum charmandaschricum.**

### **3.3 Conclusion**

Nutrients and dietary supplement are major contributors of food which help to enhance the structure and function of the body. Therefore, this study demonstrated that the leaves of the basil plants are good sources of nutrition elements and would be helpful in developing our body that can control and cure different diseases. The investigation will further enhance the existing knowledge about the nutritional values as well as the benefits of dietary consumption of these basil plants.

It was concluded that basil leave could be supplemented in different food products for the preparation of value added, and healthy.

### **3.4 Recommendations**

Through this study, I discovered that there are 21 types of Basil, so I recommend the researchers to conduct proximate analysis for all types of basil. Additionally, mixing two types of Basil and analyzing them to know the approximate analysis of them and then determine the attribution of important nutrients to maintain health and further to promote the quality of life.

I also recommend more research on the nutritional analysis of the basil seed, as well as further studies may need to be carried for other parts of basil plant.

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