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SİİRT UNIVERSITY
INSTITUTE OF SCIENCE

DETERMINATION OF HEAVY METALS, ANTIOXIDANT AND TOTAL PHENOLIC OF IRAQ KURDISH REGION AND SIIRT PROVINCE THYME

MASTER DEGREE THESIS

Karwan Mohammed Amin Mahmood KHOSHNAW
(153101009)

Chemistry Department

Thesis adviser: Assoc. Prof. Dr. İbrahim TEĞİN

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THESIS ACCEPTANCE AND APPROVAL

Karwan Mohammed Amin Mahmood KHOSHNAW prepared a thesis study titled "**Determination of Heavy Metals, Antioxidant and Total Phenolic of Iraq Kurdish Region and Siirt Province Thyme**" was accepted as a MASTER OF DEGREE by Siirt University Institute of Science and Technology, Department of Chemistry on …./…/2017 unanimously by the following jury.

Juri Members	**Signature**
President Prof. Dr. Ömer Yavuz	……..
Supervisor Doç. Dr. İbrahim TEĞİN	……..
Member Yrd. Doç Dr. Uyan Yüksel	……..
……..	

I confirm the above results.

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Karwan Mohammed Amin Mahmood KHOSHNAW

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Thank you from all my heart for your Love,
Love which was a source of everything
And brought me everything!!!

PREFACE

Heavy metals are naturally occurring elements, and are present in varying concentrations in all ecosystems. There is a huge number of heavy metals. They are found in elemental form and in a difference of other chemical compounds. Those that are volatile and those that become attached to fine particles can be widely transported on very bigger scales. Each form or compound has various charasteristics which also affect what happens to it in food web, and how toxic it is. Human activities have drastically changed the biochemical cycles and balance of some heavy metals (Nriagu, 1995; CACAR, 1996). The main anthropogenic derivations of heavy metals are different industrial processes, mining, foundries, smelters, combustion of fossil fuel and gasoline, and waste incinerators. Environmental exposure to high concentrations of heavy metals has been linked with example. several cancers and kidney damage. There are considerably more measurements data on mercury, cadmium and lead in Europe than for other metals. Medicinal plants growing in nature can accumulate heavy metals to a certain extent depending on their individual properties and the concentration of heavy metals in soil, air and water (Rai et al, 2001; Früchtenicht, 1982)

Traditional experienced of drugs herbs has always guided the search for new cures. In spite of the advent of modern high throughput medicine discovery and screening techniques, traditional knowledge systems have given clues to the discovery of valuable drugs (Buenz et al, 2004). Traditional drugs herbs are often cheaper, locally accessible and easily consumable, raw or as simple drugs preparations. today, traditional medical practices form an integral part of complementary or alternative drugs. also their efficacy and mechanism of action have not been tested scientifically in most cases, these simple medicinal preparations often mediate beneficial responses cause their active chemical constituents (Park and Pezzutto, 2002).

Many medicinal herbs contain large amounts of antioxidants such as polyphenols, which have an important role in preventing a variety of stress-related diseases and aging which are closely related to the active oxygen and lipid peroxidation (Noguchi and Niki, 1999). Antioxidants have been utilise for the avoidance and treatment of free radical-related disorders (Middleton et al, 2000). However, there have been concerns about synthetic antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) due to their possible activity as carcinogen (Barlow, 1990). So there has been a renewed scientific interest to find naturally occurring antioxidants for utilise in foods to replace synthetic antioxidants, which are being restricted cause their carcinogenicity (Velioglu et al, 1998). Opuntia dillenii belongs to the family Cactaceae is a succulent shrub native to America and West Indies (Trenary, 1997).

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LIST OF ABBREVIATIONS

Abbreviation	Explanation
WHO	: World Health organization
RSD	: Relative standard deviation
App	: Appendix
RON	: Reactive oxygen species
RNS	: Reactive nitrogen species
RNA	: Ribonucleic acid
DNA	: Deoxyribonucleic acid
DPPH	: Radical scavenging activity
FRAP	: Ferric reducing antioxidant power
PCA	: Principal component analysis
ICP-MS	: Inductively coupled plasma mass spectrometry
BHA	: Butylated hydroxyanisole
BHT	: Butylated hydroxytoluene
GC	: Gas chromatography
HPLC	: High performance liquid chromatography
UHPLC	: Ultra-high performance liquid chromatography
UV	: Ultraviolet radiation
TLC	: Thin layer chromatography
LC	: Liquid chromatography

ÖZET

YÜKSEK LİSANS TEZİ

IRAK KÜRT BÖLGESİ VE SİİRT İLİ KEKİKLERİN ANTİOKSİDAN, TOPLAM FENOLİK MİKTARI VE AĞIR METALLERİN BELİRLENMESİ

Öğrencinin Adı SOYADI

Siirt Üniversitesi Fen Bilimleri Enstitüsü
Kimya Anabilim Dalı

Danışman : Doç. Dr. İbrahim TEĞİN

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Bu çalışmada, kekik bitkisinde bulunan elementler (Li, Be, B, Na, Mg, P, K, Ca, Ti, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Se, Sr, Mo, Cd, Sn, Sb, Ba, La, Ce, Pt, Tl ve Pb) belirlenmiş ve sınıflandırmak için çeşitli kemometrik teknikler uygulanmıştır. Örnekler Irak Kürt Bölgesi'nden (Kandil, Asos, Balambo, Karokh) ve Siirt ilinden (Eruh, Şirvan, Kezer, Tillo, Akdoğmuş Köyü, Kurtalan Üç Yolu, Kurtalan Zokayt, Baykan) toplanmıştır. Mikrodalga yöntemi ile çözülen numunelerdeki metal miktarını saptamak için ICP-MS (Termo Bilimsel ICAP Q ICP-MS) cihazı kullanılmıştır. Temel bileşen analizi (PCA) ve küme analizi (CA) sınıflandırma teknikleri kullanılarqak sınıflandırma yapılmıştır.

Bunun yanında; Irak Kürt Bölgesinin dört farklı bölgesinden ve Siirt'tin sekiz farklı alanından toplanan kekik örneklerinin metanol ektraktındaki toplam fenolik madde miktarı, toplam flavonoid miktarı, antioksidan aktivitesi ve fenolik madde içeriğinin incelenmiştir.

Toplam fenolik madde miktarı Folin-Ciocalteu metoduyla, toplam flavonoid miktarı ise alüminyum klorür kalorimetrik yöntemiyle belirlenmiştir. Toplam fenolik ve toplam flavonoid madde içeriğinin maksimum değeri sırasıyla 226,83±0,21 µg gallik asit/mL ve 2,59±0,02 mg rutin/mL ekstrakt olarak bulunmuştur. Antioksidan aktivitesi FRAP ve DPPH yöntemleri kullanılarak belirlenmiştir. En yüksek antioksidan aktivitesi (IC50) sırasıyla 1,32 mg/mL ve 2,05 mg/mL bulunmuştur. Metal şelatlama aktivitesi Rival ve ark., 20014 yöntemi ile belirlenmiş olup maksimum aktivite % 22,92 olarak tespit edilmiştir. Bununla birlikte fenolik bileşiklerin kimyasal bileşeni de LC-MS/MS cihazı ile belirlenmiştir. Taranan 37 bileşenden 29'u kalitatif ve kantitatif olarak tespit edilmiştir. Bunlardan baskın olanları; Hesperidin (62,37±0,02 mg g-1), Kinik asit (1,53±0,01 mg g-1), Ferulik asit (2,23±0,01 mg g-1), Rosmarinik asit (19,90±0,01 mg
g-1), Sinnamik asit (0,81±0,01 mg g-1), Malik asit (7,62±0,09 mg g-1), Hesperetin (1,67±0,09 mg g-1).

Çalışmanın sonuçları bu örneklerin potansiyel biyolojik aktiviteye sahip olduğunu ve önemli doğal antioksidan kaynağı olarak tanıtılabileceğini göstermiştir.

Anahtar Kelimeler: Kekik, ağır metal, ICP-MS, PCA, CA

ABSTRACT

MS THESIS

DETERMINATION OF HEAVY METALS, ANTIOXIDANT AND TOTAL PHENOLİC OF IRAQ KURDISH REGION AND SİİRT PROVINCE THYME

Karwan Mohammed Amin Mahmood KHOSHNAW

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Despite an increased awareness of metal pollution, there are no studies about the metal content of thyme plant cultivated in Iraq and Siirt area. In this study value of thirty metals (Li, Be, B, Na, Mg, P, K, Ca, Ti, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Se, Sr, Mo, Cd, Sn, Sb, Ba, La, Ce, Pt, Tl and Pb) in thyme plant were determined and the results were subjected to various chemometric techniques for classification purposes. Samples were collected from Iraqi Kurdish Region (Qandil, Asos, Balambo, Karokh) and Siirt, Turkey (Eruh, Şirvan, Kezer, Tillo, Akdoğmuş Village, Kurtalan Üç Yol, Kurtalan Zokayt, Baykan). The ICP-MS (Thermo Scientific ICAP Q ICP-MS) instrument was used to detect the amount of metal in the samples dissolved by the microwave method. The analytical accuracy of the method was controlled by use of a standard reference material NASS-6. Principal component analysis (PCA) and cluster analysis (CA) as classification techniques results support each other.

Besides; Total amount of phenolics, total flavonoid content, antioxidant activity and phenolic content of methanol extracts from four different regions of Iraqi Kurdish region and eight different areas of Siirt were examined.

Total phenolic contents were assessed by Folin Ciocalteau's method and the total flavonoid content was determined by the aluminium chloride colorimetric method. The maximum value of total phenolic and total flavonoid content was found 226.83±0.21 µg gallic acid/mL and 2.59±0.02 mg rutin/mL of extract respectively. The antioxidant activity was determined by using DPPH and FRAP methods. The highest antioxidant activity (IC_{50}) was found 1.32 mg/mL and 2.05 mg/mL respectively. Metal chelating activity was estimated by method of Rival et al, 2001[4] and the highest metal chelating activity percentage of extract was 22.92 %. And also, chemical composition of the phenolic compounds were determined with LC-MS/MS instrument. As a result, 29 phenolic compounds were quantitatively determined, among the 37 phenolic compounds analyzed. The predominant of them were Hesperidin (62.37±0.02 mg g^{-1}), Quinic acid (1.53±0.01 mg g^{-1}), tr-Ferulic acid (2.23±0.01 mg g^{-1}), Rosmarinic acid (19.90±0.01 mg g^{-1}), Cinnamic acid (0.81±0.01 mg g^{-1}), Malic acid (7.62±0.09 mg g^{-1}), Hesperetin (1.67±0.09 mg g^{-1}).

The results of the study confirm that these samples have potential biological activities and can be introduced as important sources of natural antioxidants.

Keywords: Thyme, Heavy Metal, ICP-MS, PCA, CA

1. INTRODUCTION

1.1. Thyme

Plants are one of the indispensable basic sources of life since mankind's existence. Since ancient times, people have identified plants used them for various purposes and tried to promote them (Baytop, 1999). People use plants not only for nutritional purposes but also for the treatment of various diseases. Many diseases have been tried to be treated with extracts, especially from herbs. At the beginning of the 20th century, underground and overland resources were rapidly polluted as a result of the rapid industrialization with the beginning of the industrial revolution. In particular, the harmful gases which released into the atmosphere have played a major role in the ozone layer. The depletion of the ozone layer has brought the harmful effects of sunlight. Ultraviolet radiation (UV) is thought to produce free-radical species in skin, leading to premature aging and cancer (Norins, 1962). Other than this, many factors such as oxygen metabolism in living cell, environmental pollutants, pesticides and contaminant waters inevitably lead to the formation of oxygen-derived free radicals too (Koca and Karadeniz, 2003). Free radicals are necessary for life; but they can react with biomolecules easily because they are highly reactive and can form toxic compounds which can damage the cells. It has been established that oxidative stress is among the major causative factors in induction of many chronic and degenerative diseases including atherosclerosis, ischemic heart disease, ageing, diabetes mellitus, cancer, immunosuppression, neurodegenerative diseases and others (Young and Woodside, 2001).

The scope of medicinal plants is extremely differing and it has been evaluated that around 70000 kind plant species have been utilized at any rate once amid the historical backdrop of customary medication (Marimuthu et al, 2008). According to the world health organization (WHO), about three-quarters of the world population rely upon traditional remedies (herbs/plants) for their health care (Who, 1991). With eleven distinctive bioclimatic locales and around 7500 kind plants species. Thyme is also one of these plants.

Thyme is one of the most significant genera as regards numbers of species within the Lamiaceae family ,Thymus belongs to the tribe Mentheae, subfamily Nepetoideae and includes 350-400 species worldwide (Pedersen, 2008: Evans, 1989: Morales, 1997), and thyme is widely distributed in temperate zones (Demsew, 1993). Various Thymus

species are among the most popular plants throughout the world. They are commonly used as herbal teas, flavoring agents, and aromatic and medicinal plantsit is used of diseases including gastroenteric and bronchopulmonary disorders, anthelmintic, carminative, sedative ,diaphoreti as antispasmolytic, antibacterial, antifungal, secrotolytic, expectorant, antiseptic, antlelmintic and antitusive Thymus have shown activity against viruses, bacteria and fungi (Pederse, 2000).

These species have been used as carminative, antitussive, and expectorant agents. They are also reported to possess several and potent biological and pharmacological properties such as anti-inflammatory, antispasmodic, among others (Imelouane, 2009). The fresh or dried leaves of both species are utilise locally as condiments in the preparation of chilli powder, stew, bread and tea. Thyme has many uses: in chicken broth or stuffing; in clam chowder and marinades for meats or fish; in sauces; with onions, carrots or peas; in egg dishes with other sweet herbs; even in a baked apple dessert. The flavor can be captured in oils or butter. The caraway-scented form (or chemotype) of thymus herb has a historical association with roast beef (baron of beef). Lemon thyme, T. xcitriodorus, is recommended for fish, for tea, and for salad dressings, or anywhere milder thyme is desired (Asfaw et al, 2000 and Demsew, 1994). The leaves are opposite, grayish-green, entire, linear or elliptic, up to 15 mm long, tomentose beneath. The flowers are small, pale-purple or white, arranged in terminal inflorescences that may be dense or loose. Flowers appear since the beginning of summer until the end of autumn. The fruit is an ovoid smooth achene. The two species, T. schimperi Ronniger and T. serrulatus Hochst. Example Benth are endemic to the Ethiopian highlands growing on edges of roads, in open grassland, on bare rocks and on slopes, between 2200-4000 m altitudes. Both species are perennial herbs, woody at the base and 5-40 cm high. The inflorescence is commonly crowded into globose and oblong heads with pink corollas (Asfaw, 2000 and Demsew, 2000).

(Davis, 1982) is showed the genus thymus in Turkey with (39 species 60 taxa), 20 of which endemic. Acoording to (Gumus, 2010) members of this genus are called “kekik”or turkish name (kekik) in Turkish and most widely used as spices and in accustomly folk medicine to treat infectious diseases and disorders

(Jamil, 2009) showed *Thymus spp*. in Kurdistan-Iraq grow in dry stony places on overgrazed mountain slopes, in open coppiced oak forest, on rocky limestone slopes and red marl banks, on the sandstone slabs in the foot hills, also in sandy places in valley beds in the steppe, with an altitude of (500-900 m). The distribution of the genus thyme

and its species can be found at several regions in Sulaimani province such as Qaradakh, Qaladza, Ranyah, Sharbazher, Hawraman and Dokan, and in a water bed on the planes of Arbil. Thyme comes under different names in Iraqi Kurdistan, such as Jatira, Hazbe and Hezbela, perhaps it has different names in Bahdinan region where it borders with Turkey. It is therefore necessary to carry out a comprehensive scientific survey in all the regions for its distribution and species identification. *Thymus syriciacus* is found in stone rich and rock slab areas as well as in water beds among the pebbles. The harvesting period is from middle of May to middle of July, and in full flowering (white petals) stage in June. Since the species is not perennial.

In this study, it is aimed Determination of Heavy Metals, Antioxidant and Total Phenolic of Iraqi Kurdish Region and Siirt Province Thyme.

1.2. Antioxidant

Antioxidants are molecules that are capable of protecting the human body (Atoui et al, 2005). They inhibit oxidative rancidity in oil and fat-based foods, particularly meat, dairy products and fried foods. Several studies investigated their role in modulating events associated with cardiovascular disease and cancer that may also be important, antioxidants in reducing oxidative stress-induced tissue injury. Antioxidant activity inhibiting iron-induced superoxide anion formation and lipid peroxidation in microsomal and mitochondrial systems, consumers prefer natural additives that produce the same effects. The major trend therefore in the food industry has been a shift from utilise of synthetic antioxidants to natural ingredients in food products (Pokorny et al, 2006 and Pourmorad et al, 2006). Among the numerous naturally occurring antioxidants; ascorbic acid, Rosmarinic acid, carotenoids and phenolic compounds are more effective (Duh et al, 1999). They are known to inhibit lipid peroxidation (by inactivating lipoxygenase), to scavenge free radicals and active oxygen species by propagating a reaction cycle and to chelate heavy metal ions (Sudarajan et al, 2006)**.** Today, there are many various substances that have discovered antioxidant properties. While the body itself produce some of these substances as a defense system against the free radicals, occasionally people take some of them from diet, especially from herbs. Since the side effects of synthetic antioxidants have begun to show up in recent years, there is a growing attension in searching for antioxidants naturally present in herbs (Gu L et al, 2009). Due to due to their (plan's) antimicrobial activity, and their medicinal

properties, the attension in the traditional methods known as "alternative medicine" has increased and in recent years use of herbal medicine has become more predominant, and the previous few decades have seen a fastly expanding request around the world (Edrir, 2007).

1.3. Flavonoids

Flavonoids are as one set of the polyphenolic compounds among secondary metabolites in different organs of herbs that possess a wide range of biological activities (Parr and Bolwell, 2000: Noori, 2002: Noori et al, 2009). Their distribution in plants, synthesis and mode of action have been extensively studied (Shirley, 1996). They are the most numerous of the phenolics and are found throughout the plant kingdom, about 6500 kind flavonoid compounds have been detected (Harborne, 1994 and Baxter, 1999). Flavonoids are present in high concentrations in the epidermis of leaves and the skin of fruits and have significant and various roles as secondary metabolites. They are widely distributed in foods and beverages of plant origin, such as fruits, vegetables, tea, cocoa, and wine (Schreier, 2005). Different category of flavonoids and their conjugates have many functions during the interactions of plant with the environment, both in biotic and abiotic stress situations (Paiva, 1995 and Shirley, 1996). Moreover flavonoids possess a wide range of biological activities, medicinal and pharmacological effects (Parr and Bolwell, 2000, Noori, 2002, Noori et al, 2009). Some flavonoids such as quercetin, kaempferol, myricetin, apigenin, and leuteolin also have antioxidative activity in many in vitro studies (Dwyer, 1995). A large different of colours such as orange, scarlet, crimson, mauve, violet, blue and purple that we encounter in variety part of plants, especially flowers and fruits, are caused by anthocyanins (=anthocyanidin glycosides). Chalcones and some flavones and flavonols also absorb light in the visible region and are associated with bright yellow or cream coloured flowers. Other flavones account for the whiteness in most white flowers, without which they would perhaps appear translucent. Even some of the brown and black pigments found in plants are either due to oxidative products of flavonoids or related phenolic compounds (Farman, 1990). They are beneficial for the plant itself as physiological active compounds, as stress protecting agents, as attractants or as feeding deterrents, and, in general, by their significant role in plant resistance (Treutter, 2006). Also these compounds serve essential functions in plant reproduction by recruiting pollinators and seed disperses.

They are also responsible for the beautiful display of fall colour in many plant species, which has recently been suggested to protect leaf cells from photo-oxidative damage, thereby enhancing the efficiency of nutrient retrieval during senescence (Field et al, 2001).

1.4. Elements

1.4.1. Heavy metals

Heavy metals are individual metals and metal compounds that can impact human health. Eight common heavy metals such as: arsenic, barium, cadmium, chromium, lead, mercury, selenium, and silver. These are all naturally occurring substances which are often present in the environment at low levels. In larger amounts, they can be dangerous. Generally, humans are exposed to these metals by ingestion (drinking or eating) or inhalation (breathing). Working in or living near an industrial site which utilizes these metals and their compounds increases ones risk of exposure, as does living near a site where these metals have been improperly disposed. Subsistence lifestyles can also impose higher risks of exposure and health impacts because of hunting and gathering activities. Heavy metals in medicinal plants can be affected by environmental situations and geochemical characteristics of soil or location in which the plant is grow (Maksimovic et al, 2007 and Blagojević et al, 200) Heavy metals has some beneficial uses and also harmful effects on both humans and animalss (EPA, 2015).

1.4.2. Trace elements

Trace elements has an important biological activity role in human, animal and plant health. Trace metals content in medicinal plant research conducting is of great interest because of their effect on the biological active compounds in medicinal plants and may cause serious effects on human health. (Djukic-Cosic et al, 2007). Trace elements regulate metabolism as enzymatic cofactors that transport substrates across cell membranes they can guide to serious complications when deficient or in excess (Boullata,2013). Some trace elements are given such as (manganese, iron, cobalt, boron, molybdenum, chlorine, nickel). But only at low concentrations. They catalyze many biochemical reactions occurring in the organisms involved in the formation of red blood cells, hormones, and vitamins, as well as take part in the processes of photosynthesis

and the creation of pigments, respiration, oxidation, and reduction. Are part of the bone and tissues of living organisms, and participate in the functioning of neural systems. But in too high doses these metals can be toxic (Sattar, 2989: Caldas E.D, 2004; Basgel, 2006: Kara D, 2012). Iron (Fe) is also a trace element found in the heme proteins hemoglobin and myoglobin (Fraga, 2009 and Grayson et al, 2012). A relatively small amount of Ni is required to aid in the absorption of Fe in the body. However, high Ni concentrations can interfere with magnesium (Mg), and calcium (Ca) utilization and metabolism (Lawrence et al, 1989).

1.4.3. Macro elements

Elements contents of the medicinal plants and their ratios should be checked in accordance with health safety measures and it is imperative to screen for their quality control (QC) (Somer, 1983 and Arceusz et al, 2010). In recent years, several authors across the world have reported in many studies, on the importance of elemental constituents of the herbal drugs which enhanced the awareness of trace elements in the plants (Wong et al, 1993; Sharma et al, 2009; Basgel and Erdemoglu, 2006). Macro elements are usually participate in various metabolism. Other elements such as Ca and Mg are essential macro-elements required for bone structure development and necessary for carbohydrate and protein metabolism (Adebowale et al, 2007 and Bourne, 1985) Calcium and magnesium are also required in fairly large amounts to maintain body electrolytes and tissue homeostasis (Kazi et al, 2011).

1.5. Spectroscopy

Spectroscopy was originally the study of the interaction between radiation and matter as a activity of wavelength (λ). In reality, historically spectroscop belong to the utilise of visible light dispersed according to its wavelength, example, by a prism. Later the concept was expanded greatly to include any measurement of a quantity as function of either wavelength or frequency. Thus it also can belong to a respond to an alternating field or varying frequency (ν). Spectroscopy often used in physical and analytical chemistry for the identification of substances through the spectrum emitted from or absorbed by them. Common sorts (Absorption, Fluorescence, X- ray, Flame, Visible, Ultraviolet, Infrared, Near Infrared). (Baianu, 2002: Haupts et al, 1999: Robello and Diaspro, 1999).

1.6. Chromatography

Chromatography is usually introduced as a technique for separating and/or distinguish the components in a mixture. The basic principle is that components in a mixture have various tendencies to adsorb onto a surface or dissolve in a solvent. It is a powerful method in industry, where it is used on a large scale to separate and purify the intermediates and products in different syntheses. The theory There are several different sorts of chromatography currently in utilise – ie paper chromatography; thin layer chromatography (TLC); gas chromatography (GC); liquid chromatography (LC); high performance liquid chromatography (HPLC); ion exchange chromatography; and gel permeation or gel filtration chromatography. Basic principles All chromatographic methods require one static part (the stationary phase) and one moving part (the mobile phase). The techniques rely on one of the following phenomena: adsorption; partition; ion exchange; or molecular exclusion (Hostettmann, 2005).

2. LITERATURE REVIEW

2.1. Heavy metals

An imperative source of natural product drugs and the origin of modern pharmacology and drug enhancement is the herbal medicines. Thus, according to (Goldman, 2001) a significant research field is continued to be the determination of the composition of the elements of medicinal plants. This is because the necessity to observe the concentration of elements. While the consumption of some are too low, causing nutritional issues, the consumption of the other too high, causing poisonousness (Goldhaber, 2003). Human health is possibly impacted negatively by heavy elements for instance lead (Pb), Mercury (Hg), or Cadmium (Cd) (Mamani et al, 2005). Plants are a vital connection in the transmission of trace elements from soil to humans. The content of these elements is highly crucial for plants. However, the geochemical property of soil the capability of plants to selectively collect some of these elements affects the content and the condition of these elements in the plants. Plants easily integrate with these elements, which can be melted in waters and happen in ionic shapes, via the roots (Miroslowki et al, 1995). Plant protection agents, rainfall, atmospheric dusts and fertilizers are some other sources of such elements for plants, which are adsorbed via the leaf blades (Lozak et al, 2002). Information on the total concentrations of the element in biological samples like leaves offers information on the daily consumption of essential elements. Because of sampling factors, like season, time of sampling, age of the leaf, sampling height and weather conditions, the concentration of elements in leafy materials indicates huge differences (Reddy, 1997).

Analyses exposed that natural sources for trace elements are medicinal plants. The essential elements for sustaining the life processes in animals and plants are micro-nutrients like cobalt (Co), copper (Cu), manganese (Mn), molybdenum (Mo) and selenium (Se). The necessitated quantities of macronutrients like (calcium) Ca, (magnesium) Mg, (potassium) K, (nitrogen) N and (phosphorus) P are much higher compared necessitated quantities of trace elements. A poisonousness is resulted from the high consumption some essential trace elements. Recently, Heavy metals have become one of hazardous substances and measured as severe chemical health dangers for animals and human (Abu-Arab and Abou Donia, 2000, and Johnsson, 2003).

Mohammad et al, (2009) studied the level of various heavy metals such as (iron) Fe, (copper) Cu, (nickel) Ni, (cadmium) Cd, (cobalt) Co, (lead) Pb and (chromium) Cr

in the wild thyme plant (Thymus serpyllum L.). These plants are grown at diverse weather condition in Jordan. From different natural climatic zones three samples were gathered. The outcomes revealed that the highest average concentration of copper (Cu) was (10.40 mg/kg) however it is within the range of allowable boundary for medicinal plants. In addition, in *T.serpyllum*, the mean levels of lead were 1.45, 0.05, and 0.79, 1.26 mg/kg which were under the recommended level of world health organization. The content of iron (Fe) demonstrated to be apronounced differences between the various plant samples. This might be attributed to the location of cultivation. Furthermore, level of iron (Fe) differed between 15.31 and 205.80 mg/kg. The concentration of Cadmium (Cd) was under the guidelines poisonous concentrations in samples. The result indicated that contents of heavy metal in T.serpyllum were largely impacted by various natural weather circumstances. In addition, the present examination displayed that *T.serpyllum* species cultivated in in Jordan are categorized as low contents of heavy metal and can be used for edible and pharmaceutical purposes safely without any dangerous impacts on human health.

Elements of trace minerals are constitutive plant composition with biological activity as critical or poisonous metabolic agents. Heavy metals' poisonous impacts on plants are highly complicated and can influence pharmacologically active composites in medicinal plants. Therefore, it extremely decline the safety, quality and efficiency of natural plant products (Djukic-Cosic etal, 2007).Thus, since the industrial contamination of agricultural land and forests becomes a severe ecological problem in several locations of the world, the identification of heavy metals become the most important quality control analyses of the raw material of medicinal plants, particularly poisonous ones (Jones and Case,1990; Maksimović et al, 1999; Al-Jundi, 2000; Ajasa et al, 2004; Erdemoglu and Basgel, 2006; Al-Alawi and Mandiwana, 2007).

Ibironke et al, (2013) investigated some significant herbal plants from the South-west of Nigeria. The study utilized atomic absorption spectrophotometer for analyzing the content of heavy metals such as K(potassium), Na(sodium), Ca(calcium), Mn (manganese), Mg(magnesium), Cu(copper), Fe(iron), Zn(zinc), Pb(lead) and P (phosphorus), and macro-nutrient status. The results demonstrated that the most predominant heavy metals were K (162– 524 mg/mL), Ca (102 – 472 mg/mL), Mg (48.10 – 136.00 mg/mL) and Na (3.51– 10.10 mg/L) respectively. The outcomes revealed that the concentration of Cu and Zn were (0.65 – 1.48 mg/mL) and (2.40 –

6.77 mg/mL) respectively in the herbal plants which are greatly lesser compared to the stated range of the elements in agricultural products.

Premkumar et al, (2008) utilized Thyme extracted from Thymus *vulgar L.* plant like the inhibiter of volatile corrosion for minor steel in sodium chloride (NaCl) atmosphere with relative humidity of 100 %. The rate of corrosion and efficiency of inhibition of the thyme powder was evaluated by weight loss studies. In addition, thyme saturated craft paper was assessed by weight loss and potential dynamic polarization examinations. Significant inhibitive impacts was revealed by both powder and impregnated craft paper. With escalation in the concentration from 250 to 1000 mg for the powder and 250 to 1000 mg sq^{-1} ft^{-1} for impregnated craft paper, the efficiency of inhibition. It is because of adsorption of inhibitor molecule upon the surface of the metal. The thyme's adsorption behavior follows isotherm adsorption of Temkin. The inhibition efficiency declines at higher concentration. This could be because of soluble metal inhibitor complex formation in (sodium chloride) NaCl atmosphere. The highest inhibition efficiency of 80.49 and 78.64%, were found of the thyme powder of concentration 1000 mg and impregnated craft paper of concentration 1000 mg sq^{-1} ft^{-1} respectively.

Trace elements have great role in the human nutrition and health. Trace elements are physiological significant principally in metabolism of the body although the mineral elements compose a tiny percentage of total composition of most plant materials and of total body weight. It has to be mentioned that trace elements do not participate to the food energy value (Schwart, 1975). The importance of the trace elements varied between the roles as essential components biologically and imbalance produced once the excess of one impedes with the functions of another. Different trace elements are co-enzymes in particular biochemical responses in the body as well (Mensah et al, 2008).
The ancient Egyptian, Assyrian, Chinese and Indian Records indicated that plant utilization of for medicinal purposes prolongs back to earliest documented history Trease, (1989). Nowadays, it is acknowledged that trace elements have a vital role in nutrition depending on trace elements chemical arrangements or position in the body tissues or blood fluid, the function of trace elements differ. For increasing the percentage of chemical reactions in the cells some elements form crucial part of enzymes. As the elements are effective even once present in merely low level in the body cells. These enzymes are utilized over and over again. Plants encompass different quantity of mineral element (Ansari et al, 2009).

2.2. Antioxidant

Antioxidants are identified as substances, when existed at low level in comparison to oxidisable substrate, meaningfully prevents or delays oxidation (Halliwell and Gutteridge, 2000). Carvacrol and thymol have been found to be two important antioxidants composition of thyme volatile oil (Yanishlieva *et al*, 1999). According to (Jamil, 2009), in comparison to carvacrol, thymol is a better antioxidant in lipid systems because of the steric hindrance impact in phenolic group.

Antioxidants are either natural or synthetic compounds. While synthetic antioxidants are composites with phenolic structures of different amounts of alkyl substitution, natural antioxidants are nitrogen compounds, carotenoids, phenolic compounds and ascorbic acid (Hall, C A Cuppet, 1997). From the beginning of this century to the present day, synthetic antioxidants like butylated hydroxyl toluene (BHT) and butylated hydroxyl anisole (BHA) have been utilized as antioxidants. Nonetheless, due to their carcinogenicity, constraints upon the utilizations of such composites are being imposed. As a result the interest towards the application of natural antioxidant substances are increased (Mahdavi, D.L.; Salunkhe, D.K, 1995). Because the capability of antioxidant compounds in increasing the shelf life of food products, especially lipids and lipid-containing foods via hindering the lipid peroxidation process, which is the main cause for food product deterioration throughout processing. Consequently, recently the exploration for natural antioxidants, particularly removed from of plant source, has outstandingly escalated (Halliwell and Gutteridge, 2003).

Li HB et al, (2008) expressed that high capability for donating hydrogen from phenolic hydroxy groups are demonstrated by the naturally happening antioxidants of medicinal plants like flavonoids and polyphenols. Thus, establishing constant free radicals and prevent the initiation or propagation of further lipids oxidation (Fecka et al, 2007; Gülçin et al, 2004; Havsteen, 2002). Thus, the preservative behavior of aromatic plants resulted in the escalation of the interest for discovering secondary metabolites with antioxidant properties for application in foods for the replacement of synthetic antioxidants (Amrutha and Bhaskar, 2010). Moreover, human health is positively affected by the secondary metabolites with phenolic hydroxyl groups. These substances have a significant role in neutralizing free radicals that might result in immune system disorders and the expression of gene (Halliwell, 1995; Pourmorad et al, 2006; Safaei-Ghomi et al, 2009; Sharma and Bhat, 2009). A Probable mechanism of action for

antioxidant composites is the scavenging of reactive oxygen species (ROS) (Havsteen, 2002). As antioxidants are acknowledged to have some degree of protective and therapeutic impact upon several human degenerative diseases, ROS might be the contributory factor involved in these disorders. When the inner enzymatic procedures fail or are insufficiently effective, tiny molecular weight antioxidants are regarded as a probable preventative agents, which could diminish oxidative harm in the body (Halliwell, 1995).

According to several investigations, plant origin antioxidants have the ability to impede oxidation in food effectively and decrease the danger of age-dependent illnesses. Flavonoids which is prevalent in vegetables, fruits, medicinal plants as well as teas, have acquired the highest attention. In addition, because flavonoids are extremely active antioxidants, less toxic in comparison to synthetic antioxidants (butylated hydroxyanisole (BHA) or butylated hydroxytoluene (BHT)), it has been examined widely (Burda and Oleszek, 2001; Hollman et al, 1996).

In several Portuguese traditional meat dishes, especially in rabbit meat, fish food, and tomato salads, aromatic plants like Origanum vulgare, Thymbra capitata and Calamintha baetica are utilized. Various model systems were utilized for identifying the antimicrobial and antioxidant activities of the essential oils isolated from some of these plants (Miguel et al, 2003 ; Miguel et al, 2003 ; Miguel et al, 2005; Faleiro et al, 2005).

ChewYL et al, (2005) stated that for providing protection against oxidative degradation of food, antioxidants have been broadly employed as food additives. Moreover, numerous synthetic antioxidant composites have mutagenic and poisonous impacts. In contrast, food deterioration by spoilage fungi results in substantial problem because of the potential of fungi for mycotoxins production. The growth of resistant strains caused by the indiscriminate application of synthetic antifungals. In this case, greater concentrations of synthetic antifungals is required, which in turn escalates the amount of poisonous remains in food products. Furthermore, various phytochemicals arrays, which are valuable for new drug development, are produced by plants. These compositions are generally secondary metabolites continuously manufactured by the plant for protective purposes.

Mohammad et al, (2013) declared that in comparison to maceration approach, Soxhlet extraction approach is more efficient for extracting methanol crude. By rising the temperature, the yield of extraction was escalated. The DPPH was applied for measuring the antioxidant activity of various crude extracts from both extraction

approaches. The outcomes indicated that activity was highest in butanol crude extracts and the lowest in hexane crude when utilizing Soxhlet extraction approach. Nevertheless, the activity was lowest in chloroform and the highest in ethyl acetate when applying maceration approach. The results of the investigation revealed that the best approach for evaluating the antioxidant activity is maceration method.

While numerous examinations have declared antioxidant effect of different Thymus species, a small number of studies pointed out the anticholinesterase activity of thymus species. (Sigurdsson and Gudbjarnason, 2007), for example, reported that the ethanolic extract of *T.praecox* does not have AChE inhibitory impacts. In the study of (Mata et al, 2007), the inhibitory impacts of *T.serpyllum*'s H_2O and EtOH extracts and essential oil were examined against AChE. The examination discovered insignificant inhibition. Furthermore, (Rhee et al, 2003) applied the micro plate reader and fluorometric flow assays for identifying the inhabitation effects of methanol extract of *E.purpurea* roots. The study discovered that the methanol extract of *E.purpurea* roots is inactive in anti-AChE assay.

Singlet oxygen (O_2), superoxide ion ($O^{2-}\cdot$), hydroxyl ion (OH) and hydrogen peroxide (H_2O_2) are included in the Reactive oxygen species (ROS). Under normal metabolic activities, these are extremely poisonous and reactive molecules produced in cells. Nevertheless, their generation escalated in response to a different of aspects such as contaminants, tobacco smoke, alcohol, ionizing radiations, synthetic pesticides and solvent (B Halliwelland.J. M. Gutteridge, 1989). The oxidative damage to enzymes, proteins, lipids, and DNA are caused by ROS. Additionally, ROS are connected to pathogenesis of oxidative illnesses (B.Halliwell, 1997). For avoiding surplus ROS induced cellular injury, living cells have an excellent scavenging mechanism. Nevertheless, these mechanisms become ineffective with ageing and under the effects of outside stresses, therefore, nutritional complementations of synthetic antioxidants is necessitated. Recently, there has been an escalated interest in the utilization of natural materials as antioxidants and food preservatives because of toxicological concerns related to the utilization of synthetic materials in food and escalating awareness regarding natural foods (W Peschel et al, 2006).

Reactive nitrogen species (RNS) and reactive oxygen species (ROS) might cause Oxidative modifications of DNA, lipid, proteins, and tiny cellular molecules. They have an important role in a broad variety of illnesses and age associated degenerative disorders (Borek, 1993). Commonly, it is acknowledged that oxidative impairments like

aging, Parkinson's disease, cardiovascular diseases, Huntington's disease, atherosclerosis, heart disease, stroke, cancer, arteriosclerosis, diabetes, Alzheimer's disease and acquired immunodeficiency syndrome resulted from various degenerative disorders (Lee and Kim, 2003).

Safaa et al and Nafez et al, (2009) evaluated blood constituents, performance, carcass characteristics and gastrointestinal tract in broiler chickens fed various concentrations of thyme. Five dietary treatment groups with various concentrations of thyme was experimented in broiler chickens from two days of age. Body weight and body weight gain was increased, and feed conversion ratio were significantly improved by Dietary thyme. In addition, thyme supplementation considerably augmented the serum levels of glucose, total protein, and globulin. There was considerable decrease in the serum levels of cholesterol and triglyceride. Thyme feeding decreased the lengths of gastrointestinal tract segments. In addition, dressing and breast percentage of female broilers are enhanced by similar concentration of thyme. Furthermore, the leg and wing proportion were also escalated significantly. Identical trends were monitored in male broilers. In conclusion, the results recommended that in broiler rations, thyme can be utilized as a natural growth promoter.

El Mokhtar et al, (2011) employed free radical scavenging for evaluating the antioxidant activity of essential oil and extracts. The1,1-diphenyl-2picrylhydrazyl (DPPH) the activity of scavenging augmented in the sequence ethyl acetate > diethyl ether > essential oil. Between the total phenolic and flavonoid levels and antioxidant activity potential of the extracts, a relationship was monitored. Lastly, essential oils and extracts of T. capitatus can be utilized like a natural preservative ingredient in food and pharmaceutical industries because they indicate antioxidant activity.

In two diverse examinations, the variation between T.vulgaris and T. algeriensis could be illustrated by various mechanisms related with corresponding assays. Thus, every plants had various compositions with particular capabilities to contribute in those mechanisms. An evidence of traditional utilizations of these plants is the Antioxidant activity exhibited by the oils examined. For helping in the protection against food deterioration, Antioxidants are employed as food additives. The global market for industrial antioxidants had an over-all amount of approximately 0.88 million tons in 2007. The antioxidant potential observation ought to be addressed to the phenolic oil ingredients and stated chemo defending impacts against oxidative stress-mediated

disorders, primarily because of its metal chelating characteristics and free radical scavenging (Hazzit et al, 2009).

For surveying the antioxidant activity of a particular compositions or plant extracts, stable free radical DPPH° approach is a simple, fast and sensitive way (Ebrahimzadeh et al, 2008, Takeuchi et al, 2004; Bazylco and Strzelecka, 2007). Because of their hydrogen donation capability, as well as their structural requirement regarded to be vital for efficient radical scavenging, it has been stated that this activity might be caused by:

a) The existence of a 3, 4-dihydroxy, for instance, an o-dihydroxy group in the ring B, owning electron donating characteristics and being a radical target.

b) For the flavonoids antioxidant activity, the 3-OH moiety of the C ring is beneficial as well.

c) The conjugation of C2-C3 double bond with a 4-keto group is accountable for the delocalization of electron from the B ring. This further improves the capacity of radical-scavenging.

d) The combination of both 3-OH and 5-OH groups with the function of 4-carbonyl and C2-C3 double bond.

e) The substituents of hydroxyl in a catechol structures on the A-ring. This is for compensating the lack of the o-dihydroxy structure in the B-ring. Additionally become a greater determinate of antiradical activity of flavonoids (Amić.et al.2003)

Anoosh Eghdami, Mojtaba Eizadi, Fatemeh Sadeghi (2013) indicated that by employing ferric reducing antioxidant power (FRAP), 1, 1-diphenyl-2-picryl hydrazyl (DPPH) radical scavenging activity and reducing power methods, the antioxidant activities can be determined. In comparison to alcoholic extract, Hydro alcoholic extract of Thymus vulgaris had the greater flavonoid and overall phenolic contents. In this examination, it was observed that there was a strong positive correlation of R2= 0.92 between antioxidant activity and total phenolic content. The investigation demonstrated that high antioxidant activity, flavonoid and phenolic content were exhibited by Thymus vulgaris. Therefore, it can be utilised as a potential natural antioxidant.

Via the phosphor-molybdenum reduction assay and DPPH assay, the antioxidant potential was identified. The outcomes indicate that the utilized Thymus vulgaris for the current investigation belongs to thymol chemo-type. High volumes of thymol and p-cymene in Volatile oil gained via steam distillation. The above stated terpenes are the merely volatile composites identified by GC for the sample obtained by non-polar

solvent extraction. In both samples the antioxidant activity exhibited marginally greater for volatile oil acquired via steam distillation. The examination verifies that the activity of scavenging is not completely because of volatile composites, however, it also caused by other liposoluble substances (Grigore, 2010).

In foods, the Antimicrobial agents are becoming progressively significant. On fresh vegetables and fruits, the growth of fungi is accountable for spoilage of food and abundant plant diseases, which cause substantial economic losses. The spoilage of numerous foods are caused by Aspergillus, Fusarium and Penicillium (Rojas, 2005 and Agrios, 1997). Numerous species of Penicillium, Aspergillus and Alternaria can synthesize mycotoxins despite producing diseases in plants. As these components can be carcinogenic, lethal, mutagenic, immunosuppressant, teratogenic, or might mimic estrogens, they are hazardous to animal and human health (da Cruz, 2013). Conversely, according to (Dorman, 2004 and Hsieh, 2001). Antioxidants have broadly been applied as food additives. The demand for reducing the utilization of synthetic antioxidants like butylated hydroxyl anisole (BHA) and butylated hydroxyl toluene (BHT) in the food manufacturing has prompted the necessity for developing alternative active composites, which are safe to the environment and consumers (Shi J et al, 2009 and Reische, 1998).

2.3. Phenolic compounds

Mainly the Structure of phenolic composites consists of an aromatic ring. It is varied between simple phenolic molecules to extremely polymerized composites. (Bravo, 1998). Regardless of this variety in the structure, the group of composites is regularly denoted as polyphenols. The conjugation with poly- and mono-saccharides is the most naturally happening phenolic composites, connected to one or more of the phenolic groups. In addition, it might happen as functional derivatives like methyl esters and esters. The key dietary phenolic composites are flavonoids, tannins and phenolic acids (King and Young, 1999). As indicated in figure (2.1) the hydroxyl benzoic and hydroxyl cinnamic acids are the two subgroups of phenolic acid. Gallicp-hydroxy benzoic, protocatechuic, syringic and vanillic acids, which commonly have the C6–C1 structure, are included in hydroxy benzoic acids. in contrast, Hydroxy cinnamic have a three-carbon side chain (C6–C3) structure, with caffeic, Flavonoids, p-coumaric and ferulic, are composed the biggest class of phenolic plant, comprise fifty percent of the eight thousands naturally happening phenolic composites (Harborneet al, 1999). The

Flavonoids, which consists of fifteen carbon atoms organized in a C6–C3–C6 structure, have low molecular weight. Principally it consists of two aromatic rings A and B, connected via a three-carbon bridge, frequently in a heterocyclic ring, C configuration, as shown in figure (2.2). Whereas aromatic ring B is originated from phenylalanine throughout the shikimate path, the ring A is originated from the acetate/malonate path (Bohm, 1998). Differences in replacement patterns for ring C cause the main flavonoid classes for instance, flavonols, flavanols, flavones, isoflavones, flavanones, anthocyanidins and flavanonols, as demonstrated in figure (2.3) (Hollman and Katan, 1999). The most structurally diverse and broadly happening flavonoid classes are flavonols and flavones (Harborne et al, 1999). Within every single class of flavonoids, replacements to rings B and A result in the various compositions (Pietta, 2000). Alkylation, oxygenation, sulfation, acylation and glycosylation are examples of these substitutions (Bohm, 1998; Hollman and Katan, 1999).

Thousands of molecules that have a polyphenol structure are identified in higher plants. Because of the reactivity of the phenol moiety, phenolics behave as antioxidants. Ubiquitous are the origin of polyphenols, as stated above. However, literatures indicates that phenolic compounds in spices have been the interest of researchers. Various quantities of flavonoids and phenolic acids have been detected in spices relying on plant part analyzed (leaves, flowers), growing conditions, extraction conditions spices are utilized for aromatization of oil either for expanding the commercial benefits or oil enrichment. Thyme, oregano are the most regularly utilized spices for oil aromatization. According to Literature, these spices comprise various quantities of phenolic composites (Jayasingne et al, 2003; Javanmardi et al, 2002; Grayer et al, 1996). In accordance to (Javanmardi et al, 2002, Exarchou et al, 2002) and (Pizzale et al, 2002) Rosmarinic acid is a dominant antioxidant and was recognized as a major phenolic composites in oregano. Luteolin, caffeic acid, dihydroxicampherol, apigenin, dihydroxiquercitine and eriodictyol are other phenolic compositions in oregano (Škerget et al, 2005: Pizzale et al, 2002, Kulevanova et al, 2001). In addition, glycuronids of apigenin, eriodyctiol, luteolin, rosmarinic acid, quercitine and luteolin glycosides are the major phenolic compounds in thyme (Justesen, 2000; Guillen and Manzanos, 1998).

Zanda Kruma et al, (2008) stated that spices are among different plentiful origins of phenolic compositions, for example thyme determined phenolic compounds in oregano. Rosmarinic acid, caffeic acid, eriodyctiol, apigenin, luteolin and naringenin were the most popular phenolic compounds in spices. Rosmarinic acid was the major

phenolic composition in spices. On the other hand other composition presented below 4% from the overall phenolics. Caffeic acid was only detected in thyme. The flavonons (eriodictyol, naringenin) and flavones (apigenin, luteolin) were detected in spices. Furthermore, Luteolin and apigenin were identified in thyme and oregano. Whereas naringenin was detected in thyme, Eridyctiol was identified in all spices, the maximum level were identified in oregano. In comparison to flavons (apigenin and luteolin), Flavonons (eriodyctiol and naringenin) were detected in spices in higher level. Thyme contained the maximum phenolic compounds.

Ioannis K et al, (2004) applied ANOVA and Linear Discriminant Analysis for differentiating Greek thyme honeys in accordance to geographical origin, grounded upon conventional physicochemical factors and phenolic compound content. Throughout the picking time in 2011 thirty five thyme, honey samples were gathered from various zones in Greece which recognized to yield decent quality thyme honey. High pressure liquid chromatography was applied for analyzing phenolic compounds such as chrysin, quercetin, kaempferol, myricetin, and syringic acid. The detection of pH, and total acidity, moisture, ash, electrical conductivity and lactonic/free acidity proportion and color parameters were included in the Conventional quality parameter analysis. Thyme honey samples were reasonably classified (91.2% correct prediction) in accordance to geographical origin by using five phenolic compounds and conventional quality parameters.

For the determination of both geographical and floral source of honey, volatile compounds, Phenolic content, free amino acids, trace metal content and physicochemical parameters like total acidity, electrical conductivity (EC), pH and water activity have been employed (Acquarone et al, 2007; Al et al, 2009; Senyuva et al, 2009). Anti-carcinogenic, anti-atherogenic, anti-inflammatory, antithrombotic, antibacterial immune-modulating and analgesic activity are demonstrated by phenolics, flavonoids and phenolic acids (Vinson et al, 1998; Nasuti et al, 2006; Viuda-Martos et al, 2008).

Ali and Neda, (2001) reported that phenolic acids and flavonoids composed one of the most prevalent classes of phenolics plant. Because of the importance flavonoid in human health and plants, it might be beneficial to acquire a better comprehension of the concentration of biological activities and flavonoid which might demonstrate potentials of flavonoid as healing factors, as well as for the prediction and monitoring the medicinal herbs' quality. Moreover, humans consumed herbs and plants which

encompass thousands of various flavonoid and phenolic acid compositions. Because of phenolics anti-oxidative and possible anti-carcinogenic activities, the impacts of their dietary is presently of great interest. Phenolic acids and Flavonoids work as free radical scavengers, declining agents and quenchers of singlet oxygen creation. Furthermore, they have significant roles in controlling cancer as well as other human illnesses.

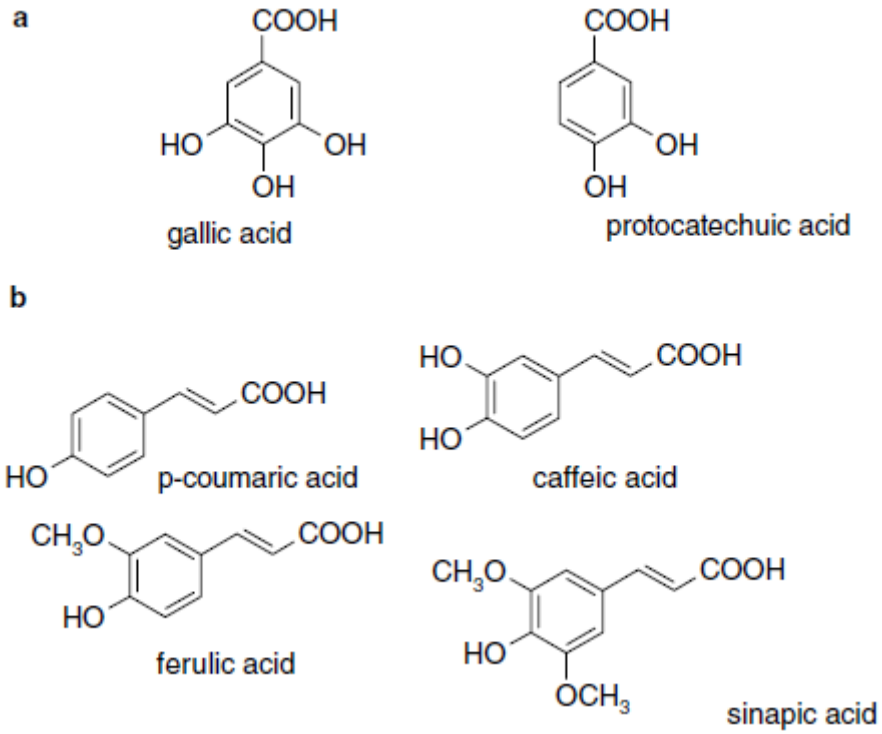

Figure 2.1. Examples of (a) hydroxyl benzoic acid (b) hydroxinnamicacid

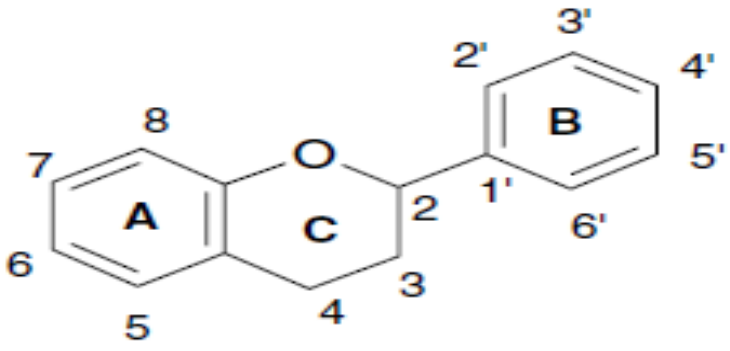

Figure 2.2. Examples of generic structure of aflavonid molecules

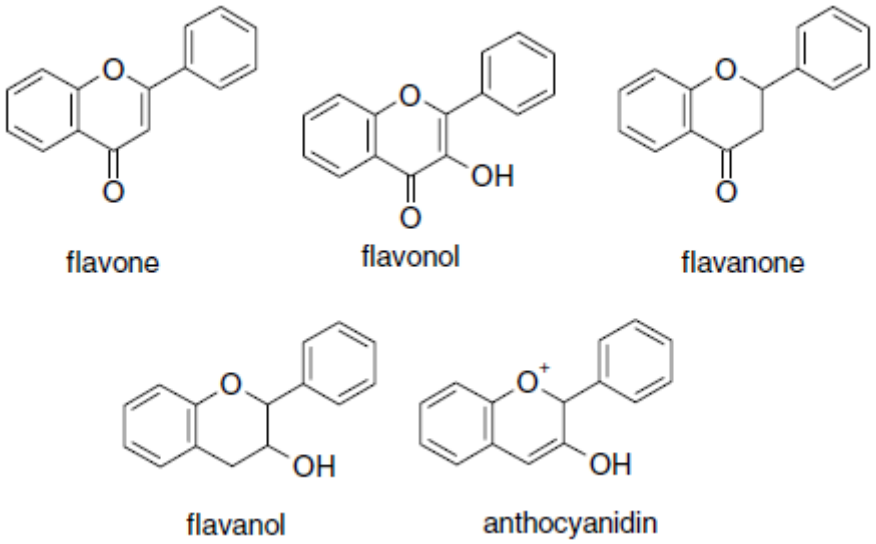

Figure 2.3. Examples of generic structure of major class flavonid

3. MATERIAL METHODS

3.1. Materiels

Thyme samples collected from 12 different localities from Siirt Province and Kurdistan regional of Iraq.

3.2. Methods

3.2.1. Preparation of plant specimens

The collected plants were dried in the laboratory environment, in a place where the air flow was present and at the room temperature in the shade area, and then powdered with a mixer. Powdered plant samples were placed in glass jars and stored at room temperature.

3.2.2. Heavy metals

An ICP-MS instrument (Thermo Sciencetific İCAP Q ICP-MS) was used for the determination of metal.

3.2.3. Digestion procedure

A Berghof Speedwawe MWS-3 model microwave digestion system was used for acid digestion of samples. The microwave acid digestion was carried out as follows: 2.0 g portion of dried sample was weighed and transferred into a pressure- resistant polytetrafluoroethylene (PTFE) vessel (volume 100 mL), and the mixture of acids (HNO_3 + H_2O_2, 2.5: 7.5 mL) was added. Microwave digestion system under the conditions described in Table 3.1. The power applied in program was 1450 W. The reaction mixture was subjected to an evaporation module in order to remove the acids after the final digestion. Then the residue was dissolved in Milli-Q water and filtered, and the filtrate was diluted to a fixed volume.

Table 3.1. Operating conditions for digestion by microwave oven.

	1	**1**	**2**	**3**	**4**
T (ºC)	100	100	160	180	100
Ta (min) [a]	10	10	10	10	10
Time (min) [b]	5	5	3	3	3

[a] Waiting time at desired temperature.
[b]The time between the two sequential temperature

3.2.4. LC-MS/MS device and chromatographic conditions

37 LC-MS / MS system used for qualitative and quantitative analysis of phytochemicals; Shimadzu Neexera model UHPLC device and Shimadzu LCMS 8040 model triple quadrupole mass spectrometer device. The liquid chromatography system used consists of the LC-30 AD model gradient pump, the DGU-20A3R model degasser, the CTO-10ASvp model column oven and the SIL-30AC model autosampler. Chromatographic separation was performed on Inertsil ODS-4 model C18 (100 mm × 2.1 mm, 2 μm) column. During the analysis the column furnace is set at 35 ºC. In the elution gradient, ultrapure water for mobile phase A and acetonitrile for mobile phase B were used. In addition, 10 mM ammonium formate and 0.1% formic acid were added to the water phase to facilitate better chromatographic separation and ionization. After several attempts to achieve optimal separation of analytes, the most suitable UHPLC gradient profile was obtained with a gradient profile of 5-20% B (0-10 min), 20% B (10-22 min), 20-50% B (22-36 min) % B (36-40), 5% B (40-50 min). The moving phase flow rate was 0.25 mL / min and the injection volume was 4 μL

The triple quadruple mass spectrometer is equipped with an ESI (electrospray ionization) source operating in both a negative and a positive mode. The LC-ESI-MS / MS data were collected and processed by the registered software Lab Solutions (Shimadzu, Kyoto, Japan). The quantitative analysis of the analytes was carried out in the device in multiple reaction monitoring (MRM) mode and the parent ions were combined with one or two product ions (the other was used for qualitative purposes for quantitative purposes). Other parameters that are optimized in the mass spectrometer are: interface temperature; 350 ºC, DL temperature; 250 ºC, heat block temperature; 400 ºC, nebulizer gas (N_2) flow; 3 L / min and drying gas (N_2) temperature; 15 L / min (Yılmaz, 2015).

Table 3.2 Analytical parameters of the LC-MS / MS analysis method ([a]RT: Retention time, [b]Master ion (*m/z):* Molecular ions of standard compounds (m/z rate), [c]R^2: Determination coefficient, dRSD: Relative standard deviation, eLOD/LOQ (µg/L): Detection limit/ Assignment limit, f U (%): 95% Relative standard uncertainty at confidence level (k=2)) (Yılmaz, 2015)

No	Analytes	RT[a]	Main Ion (m/z)[b]	Fragmenation ions	Ion mode	Calibration Equation	R^{2c}	RSD%[d] Same day	different days	Linearity December (µg/L)	LOD/LOQ (µg/L)[e]	Regain (%) Same day	Different days	U[f]
1	Kumarin	17.40	147.05	91.0-103.2	Poz	y=33.64×-89700	0.994	0.01306	0.01239	1000-20000	208.4/228.4	0.99947	1.00081	0.0237
2	Hesperidin	12.67	610.90	303.1-465.1	Poz	y=1340.27×-43769	0.998	0.00945	0.01126	25-1000	3.4/4.2	1.01733	1.01263	0.0262
3	p-Kumaric acid	11.53	162.95	119.3-93.3	Neg	y=3199.20×+13002	0.992	0.01820	0.01727	25-1000	7.3/9.1	1.00617	1.01224	0.0516
4	o-kumaric acid	15.45	162.95	119.4-93.3	Neg	y=1219.34×-10915	0.999	0.02730	0.02566	25-1000	24.4/31.1	0.98344	0.99061	0.0513
5	Gallic acid	3.00	168.85	125.2-79.2	Neg	y=226.76×+38152	0.998	0.01601	0.01443	250-10000	95.5/106.9	1.00004	1.00454	0.0282
6	Caffeic acid	8.80	178.95	135.2-134.3	Neg	y=3963.32×+178156	0.998	0.01454	0.01469	25-1000	18.4/22.4	1.00917	0.98826	0.0354
7	Vanilic acid	8.57	166.90	152.3-108.3	Neg	y=35.84×-12097	0.999	0.00528	0.00619	1000-20000	122.2/139.7	1.00093	1.04095	0.0508
8	Salicylic acid	11.16	136.95	93.3-65.3	Neg	y=5286.26×+309192	0.989	0.01016	0.01242	25-1000	5.0/6.5	1.00989	0.99013	0.0329
9	Kynic acid	1.13	190.95	85.3-93.3	Neg	y=41.06×+10671	0.996	0.00259	0.00274	250-10000	75.8/79.4	1.00288	0.98778	0.0082
10	4-OH-Benzoicacid	7.39	136.95	93.3-65.3	Neg	y=409.03×+112079	0.998	0.01284	0.01538	250-10000	33.2/38.1	0.99662	1.00058	0.0289
11	Ferulic acid	12.62	192.95	178.3	Neg	y=80.45×-31782	0.997	0.00708	0.00619	250-10000	36.6/42.0	0.99987	1.00289	0.0494
12	Chlorogenicacid	7.13	353.15	191.2	Neg	y=781.36×-18697	0.998	0.00058	0.00076	25-1000	6.2/8.1	1.00806	0.99965	0.0069
13	Rozmarinic acid	14.54	359.00	161.2-197.2	Neg	y=909.67×-201692	0.994	0.02014	0.01751	100-5000	6.6/8.8	0.99206	1.03431	0.0713
14	Protocatechic acid	4.93	152.95	108.3	Neg	y=297.75×+30590	0.995	0.01236	0.01296	100-5000	28.2/31.4	0.99404	1.01070	0.0411
15	Cinnamic acid	25.61	147.00	103.15-77.3	Neg	y=9.06×-12403	0.996	0.00648	0.00816	5000-20000	821.8/859.7	1.00051	0.99927	0.0143
16	Sinapinic acid	12.66	222.95	208.3-149.2	Neg	y=141.96×-73294	0.992	0.01446	0.01517	250-10000	78.7/86.1	1.00164	0.99962	0.0281
17	Fumaric acid	1.48	115.00	71.4	Neg	y=64.99×-11592	0.997	0.00536	0.00460	100-5000	28.1/34.5	0.99748	0.99867	0.0124
18	vanillin	10.87	151.00	136.3-92.2	Neg	y=446.10×+70934	0.998	0.00696	0.00793	250-10000	44.3/53.1	0.99679	0.99611	0.0280

Table 3. 3. Analytical parameters of LC-MS / MS analysis method (Continued) (Yılmaz, 2015)

No	Analytes	RT[a]	Main ion(m/z)[b]	Fragmenation Ions	Ion Mode	Calibration Equation	R^{2c}	RSD%[d]		Lineerity December (µg/L)	LOD/LOQ (µg/L)[e]	regain (%)		U[f]
								Same Day	Different Days			Same Day	Differenet Days	
19	pyrocatechol	6.48	109.00	108.35-91,3	Neg	y=30.61×+14735	0.996	0.01313	0.01339	1000-20000	261.1/278.4	0.99987	0.99936	0.0235
20	Malic acid	1.23	133.00	115.2-71,3	Neg	y=316.95×-42041	0.999	0.00477	0.00527	250-10000	55.3/67.5	1.01266	0.99836	0.0113
21	Syringic acid	9.02	196.95	182.2-167,3	Neg	y=42.33×-52547	0.996	0.01049	0.01345	1000-20000	212.5/233.3	0.99922	0.99977	0.0238
22	Hesperetin	31.76	300.95	164.2-136,2	Neg	y=876.67×+48916	0.997	0.03209	0.02605	25-1000	5.6/6.9	0.98850	0.99435	0.0562
23	Naringenin	30.68	270.95	151.2-119,3	Neg	y=4315.1×+178410	0.995	0.02054	0.02019	25-1000	5.4/6.4	0.99883	1.01002	0.0521
24	Routine	12.61	609.05	300.1-271,1	Neg	y=561.91×-16879	0.997	0.00473	0.00624	25-1000	5.5/6.5	1.00994	0.98017	0.0159
25	quercetin	28.17	300.90	151.2-179,2	Neg	y=1198.48×+480562	0.990	0.01589	0.01360	100-5000	23.3/28.9	0.98470	1.00103	0.0543
26	Kersitr	16.41	447.15	301.1-255,1	Neg	y=339.39×+38910	0.999	0.01528	0.02320	100-5000	22.0/25.2	0.99726	1.00620	2.0079
27	Apigenin	31.43	268.95	117.3-151,2	Neg	y=4548.36×+295252	0.990	0.02304	0.02204	25-1000	5.4/6.3	1.01444	1.01331	0.0650
28	Chrysin	36.65	252.95	143.3-119,4	Neg	y=2032.13×+95593	0.993	0.00490	0.00630	25-1000	5.4/6.2	1.00338	1.00437	2.0083
29	Likiritigenin	25.62	254.95	119.3-135,1	Neg	y=2384.96×+59141	0.996	0.01849	0.01738	25-1000	5.5/6.6	1.00333	0.99957	0.0341
30	İzokersitrin	13.42	463.00	300.1-271,1	Neg	y=803.23×+4981	0.999	0.00682	0.00515	25-1000	5.4/6.3	1.00594	1.00722	0.0133
31	Apigetrin	16.59	431.00	268.2-239,2	Neg	y=1775.55×+91121	0.993	0.01797	0.01607	25-1000	5.4/6.1	1.01394	1.00419	0.0597
32	Roifolin	16.11	577.05	269.2-211,1	Neg	y=237.15×+11887	0.999	0.00747	0.01528	100-5000	23.1/27.9	1.01046	1.01739	0.0941
33	Nikotiflorin	14.68	593.05	285.1-255,2	Neg	y=498.38×+79274	0.991	0.00737	0.00875	100-5000	22.4/25.5	1.02558	1.00970	0.0276
34	Fisetin	19.30	284.95	135.2-121,3	Neg	y=547.46×+274791	0.991	0.00557	0.00820	250-10000	54.4/61.4	0.99877	1.00031	0.0148
35	Luteolin	28.27	284.75	133.2-151,2	Neg	y=3272.65×+150557	0.997	0.00575	0.00696	25-1000	5.4/6.5	1.00772	0.99524	0.0174
36	myricetin	18.72	317.00	179.2-151,3	Neg	y=583.55×+205727	0.999	0.00652	0.00711	250-10000	53.2/57.2	0.99982	1.00042	0.0126
37	Kamferol	31.88	284.75	255.1-117,3	Neg	y=26.29×+87558	0.992	0.01436	0.01070	1000-20000	206.6/214.3	0.99971	0.99851	0.0209

3.2.5. Antioxidant

3.2.5.1. Extract preparation

4 g of the pulverized plant sample was placed in a beaker and 40 ml of 80% methanol was added. The mixture was homogenized for 2 minutes and ultrasonicated for 5 minutes, then left overnight on a shaker. The extract is then filtered through filter paper. After the extract was dried at 38 °C in stock tubes, stock concentrations were established by adding 80% methanol as the concentration of solid part remaining in the bottom of the tube was 10 mg / ml.

3.2.5.2. Total phenolic analysis

Total phenolic contents of the thyme were determined according to Folin-Ciocalteu reactivity and gallic acid standard (Slinkard and Singleton, 1977). Extract solution was taken into a 0.1 ml tube and 1 ml of Folin-Ciocalteu was added and the flask was thoroughly shaken. After 3 minutes, 1 ml of a 6% Na_2CO_3 solution was added and the mixture was allowed to stand for 1 hour with intermittent agitation.Absorbance was measured at 760 nm. The same procedure was repeated in galic acid sols.

3.2.5.3. Total flavonoid content analysis

Total flavonoid content was determined using the aluminum chloride colorimetric method (Zhishen et al, 1999, and Zou et al, 2004). 0.5 ml of the extract solution was mixed with 2 ml of distilled water and 150 µl of 5% sodium nitrate. After 6 minutes, 150 µl of 10% aluminum chloride and 2 ml of 1 M sodium hydroxide were added and left at room temperature for 15 minutes. The absorbance of the samples was measured at 510 nm. Routine is used as standard.

Table 3.4. Total Flavonoid Content Analysis

Rutin Standard (mg/ml)	1 mg/ml of Rutin (ul)	% 80 Aqueous Methanol	Total Volume
1	1000	0	1000
0.8	800	200	1000
0.6	600	400	1000
0.4	400	600	1000
0.2	200	800	1000
0.1	100	900	1000
0.05	50	950	1000
Blank	0	1000	1000

3.2.5.4. DPPH analysis

DPPH analysis (Villaño et al, 2007). 4 ml of 0.01 mM DPPH solution (prepared with 80% methanol) was added to 1 ml extract and incubated for 15 minutes in the dark and then absorbed on a spectrophotometer at 517 nm wavelength.

Control: 1 ml of solvent and 4 ml of DPPH

DPPH activities (% incubation) = $\frac{(A_C - A_1)}{A_C} x100$

(A_C: Control absorbance, A_1: Sample absorbance)

3.2.5.5. DPPH analysis

FRAP analysis was performed according to (Benzie and Strain, 1996) 3 mL of FRAP reactant was added onto 100 µL of sample diluted appropriately. The incubation was carried out in the dark at room temperature for 6 minutes and then absorbed at 593 nm)

FRAP solution: 10 ml Acetate Buffer + 10 ml Iron (III) Chloride-Hexahydrate + 1 ml TPTZ solution.

Solutions:

Acetate Buffer: (300 mmol / L; PH: 3.6) solution preparation; 3.1 g of $NaOOC_2H_3$ in 16 ml of Acetic Acid / L distilled water.

HCl: (40 mmol / L) solution preparation; 400 µl HCl (32%) / 100 ml purified water.

Preparation of TPTZ: (10 mmol) solution; 31.2 mg TPTZ / 10 ml HCl 40 mmol.

$FeCl_3$ * $6H_2O$: solution preparation; 54.1 mg $FeCl_3$ * $6H_2O$ / 10 ml pure water, as shown in table 3.5

Table 3.5. FRAP Analysis

Frap tandard (ul)	Dilution	Stock Solution	Purified water
500 umol/L	1:10	100	900
625 umol/L	1:8	100	700
1000 umol/L	1:5	200	800
1667 umol/L	1:3	300	600
2500 umol/L	1:2	500	500

3.2.5.6. Iron chelating analysis

The metal chelating activity of the samples is described by (Rival et al, 2001) and (Duh et al, 2001) 1 ml of the samples diluted in the appropriate dilution was mixed with 3.7 ml of 95% ethanol. Each sample was incubated with 0.1 ml 2 mM $FeCl_2$ for 60 minutes. After incubation, 0.2 ml of 5 mM ferrous sulfate was added and incubated at room temperature for 10 minutes. The absorbance of the complex formed by ferrous ions and ferrozine was measured at 562 nm. The control was prepared under the same conditions as ethanol without the sample material. All experiments were carried out in 3 replicates and the averages of the results were taken.

4. RESULTS AND DISCUSSION

4.1. Metal analysis

Table 4.1, 4.2 and 4.3 showed the mean of element analysis of thyme samples and table 4.4, 4.5 and 4.6 showed the standard deviation of element analysis of thyme samples.

Table 4.1. The mean of element analysis of thyme samples (N=3)

No	Name	Li [ng g^{-1}]	Be [ng g-1]	B [µg g-1]	Na [µg g-1]	Mg [mg g-1]	P [mg g-1]	K [mg g-1]	Ca [mg g-1]	Ti [µg g-1]	V [ng g-1]
M1	Qandil	172.43	n.d.	60.89	157.19	3.72	2.29	28.23	1.22	27.48	0.50
M2	Asos	564.29	51.26	80.36	634.91	4.71	1.85	31.70	4.09	92.40	2.02
M3	Balambo	1024.99	n.d.	85.19	422.99	5.43	1.10	29.87	1.96	43.14	0.71
M4	Karokh	283.56	n.d.	70.35	638.04	5.00	1.42	29.93	1.93	41.59	0.62
M5	Eruh	972.32	26.99	59.37	239.90	9.91	1.91	29.31	2.51	60.19	3.86
M6	Şirvan yolu Kezer	384.72	n.d.	53.47	246.58	3.71	1.97	25.62	2.11	46.04	0.87
M7	Kampus	1105.52	n.d.	62.11	255.19	5.74	1.32	33.30	3.71	53.57	1.93
M8	Tillo Akdoğmuş	394.94	n.d.	69.04	265.51	5.18	1.61	26.85	2.10	50.55	2.22
M9	Koyu Kurtalan	526.62	n.d.	35.32	205.30	2.71	1.45	20.74	2.48	52.94	2.41
M10	Üçyolu Kurtalan	972.18	53.97	51.81	207.78	6.55	1.06	19.07	2.17	49.80	1.87
M11	Zokayt	1217.86	55.31	38.70	249.68	5.86	2.24	22.44	2.34	54.98	2.90
M12	Baykan	283.52	56.66	79.49	297.08	4.87	2.48	44.86	2.79	62.36	0.97

Table 4.2. The mean of element analysis of thyme samples (N=3)

No	Name	Cr [µg g-1]	Mn [µg g-1]	Fe [µg g-1]	Co [ng g-1]	Ni [µg g-1]	Cu [µg g-1]	Zn [µg g-1]	As [ng g-1]	Se [µg g-1]	Sr [µg g-1]
M1	Qandil	1.12	82.03	160.26	228.10	20.57	243.82	68.61	142.34	0.83	38.44
M2	Asos	2.64	136.32	436.89	942.32	38.14	258.13	103.26	427.62	4.37	90.83
M3	Balambo	1.25	86.49	193.52	525.71	17.86	143.29	111.72	185.74	0.69	102.55
M4	Karokh	2.05	60.23	231.17	1027.22	11.61	58.19	43.83	434.90	1.46	66.87
M5	Eruh	4.84	130.49	698.03	1254.79	46.84	1100.39	291.91	523.39	6.90	50.47
M6	Şirvan yolu Kezer	17.43	83.44	341.63	411.07	12.51	1216.98	394.24	188.81	2.18	56.03
M7	Kampus	29.93	271.06	596.36	785.07	223.46	8428.85	4477.43	782.55	1.76	1353.64
M8	Tillo Akdoğmuş	6.74	63.29	529.83	607.44	75.52	7052.18	2361.62	628.19	3.29	63.63
M9	Koyu Kurtalan	6.46	97.11	415.41	649.76	69.70	3679.32	1478.08	458.94	3.87	171.71
M10	Üçyolu Kurtalan	4.01	89.94	416.38	644.21	6.85	37.12	58.13	271.88	1.88	109.76
M11	Zokayt	4.18	155.49	563.82	941.96	13.64	44.04	26.59	317.62	4.93	124.55
M12	Baykan	4.59	183.39	309.24	680.86	24.30	32.99	50.79	273.17	2.63	92.29

Table 4.3. The mean of element analysis of thyme samples (N=3)

No	Name	Mo [µg g-1]	Cd [ng g-1]	Sn [µg g-1]	Sb [ng g-1]	Ba [µg g-1]	La [µg g-1]	Ce [µg g-1]	Pt [ng g-1]	Tl [ng g-1]	Pb [µg g-1]
M1	Qandil	2.27	27.89	11.36	187.33	18.70	0.26	0.53	12.14	12.86	2.96
M2	Asos	1.92	80.91	3.47	259.28	51.26	1.42	1.94	4.62	16.14	5.64
M3	Balambo	1.94	58.75	12.15	220.09	18.85	0.32	0.75	3.57	10.19	12.17
M4	Karokh	1.45	34.02	3.43	192.23	13.22	0.17	0.33	3.79	7.83	1.96
M5	Eruh	1.88	2134.38	12.52	275.85	33.68	2.76	3.39	2.86	22.33	54.10
M6	Şirvan yolu Kezer	1.76	67.21	7.22	387.98	55.81	0.58	0.79	3.06	10.22	23.50
M7	Kampus	2.55	433.84	129.21	1146.00	81.70	0.73	1.34	6.95	19.72	292.89
M8	Tillo Akdoğmuş	2.42	345.03	109.61	708.07	66.84	0.88	1.77	3.38	19.09	161.97
M9	Koyu Kurtalan	1.30	205.32	26.84	475.44	44.69	1.05	2.00	1.92	10.68	103.42
M10	Üçyolu Kurtalan	1.85	147.22	7.27	216.28	85.46	0.67	1.28	2.04	21.45	3.61
M11	Zokayt	1.86	223.89	5.94	260.90	164.27	1.15	2.23	2.24	33.04	6.69
M12	Baykan	1.58	80.26	7.91	222.60	43.44	1.18	2.28	2.03	9.46	2.82

Table 4.4. The standard deviation of element analysis of thyme samples (N=3)

No	Name	Li [ng g^{-1}]	Be [ng g-1]	B [µg g-1]	Na [µg g-1]	Mg [mg g-1]	P [mg g-1]	K [mg g-1]	Ca [mg g-1]	Ti [µg g-1]	V [ng g-1]
M1	Qandil	37.33	2.34	3.50	1.40	0.04	0.05	0.27	0.01	0.58	0.02
M2	Asos	235.08	4.05	6.13	1.66	0.06	0.09	0.24	0.02	0.72	0.05
M3	Balambo	270.16	n.d.	7.66	6.37	0.05	0.05	0.09	0.04	1.12	0.03
M4	Karokh	0.00	n.d.	5.42	6.81	0.05	0.07	0.37	0.03	0.54	0.01
M5	Eruh	405.13	2.34	6.05	0.42	0.09	0.09	0.30	0.04	1.40	0.07
M6	Şirvan yolu Kezer	133.27	2.34	2.06	4.10	0.09	0.12	0.26	0.04	0.44	0.01
M7	Kampus	85.04	n.d.	5.09	6.43	0.04	0.01	0.53	0.05	1.43	0.03
M8	Tillo Akdoğmuş	105.24	n.d.	6.96	5.56	0.02	0.03	0.33	0.02	1.59	0.07
M9	Koyu Kurtalan	120.66	n.d.	4.46	2.04	0.04	0.07	0.13	0.02	0.48	0.09
M10	Üçyolu Kurtalan	280.64	4.05	2.82	5.58	0.02	0.11	0.13	0.01	0.77	0.05
M11	Zokayt	95.88	4.05	4.73	2.32	0.04	0.09	0.15	0.02	1.05	0.09
M12	Baykan	49.11	7.01	4.10	2.33	0.07	0.08	0.73	0.06	0.91	0.02

Table 4.5. The standard deviation of element analysis of thyme samples (N=3)

No	Name	Cr [µg g-1]	Mn [µg g-1]	Fe [µg g-1]	Co [ng g-1]	Ni [µg g-1]	Cu [µg g-1]	Zn [µg g-1]	As [ng g-1]	Se [µg g-1]	Sr [µg g-1]
M1	Qandil	0.02	0.65	3.19	5.43	0.36	4.28	0.50	10.69	0.50	1.11
M2	Asos	0.02	2.84	3.44	27.04	0.65	3.27	0.93	75.59	2.82	1.26
M3	Balambo	0.04	0.50	5.28	10.19	0.21	1.59	0.59	74.27	1.08	2.05
M4	Karokh	0.05	0.15	0.45	28.30	0.15	0.79	0.95	88.95	0.55	0.62
M5	Eruh	0.14	1.60	12.11	29.35	0.85	7.00	3.46	36.52	2.54	0.99
M6	Şirvan yolu Kezer	0.25	1.70	4.80	9.86	0.12	5.86	5.52	19.24	0.60	1.69
M7	Kampus	0.25	1.66	7.29	10.37	2.79	15.92	45.29	0.00	0.66	15.42
M8	Tillo Akdoğmuş	0.03	0.82	6.09	7.41	0.53	103.17	49.33	141.05	2.82	1.08
M9	Koyu Kurtalan	0.06	1.81	6.78	17.59	0.14	34.53	6.27	101.31	0.71	1.73
M10	Üçyolu Kurtalan	0.02	1.38	6.27	16.35	0.14	0.73	1.05	30.38	0.96	0.39
M11	Zokayt	0.11	1.99	5.47	14.74	0.13	0.97	0.53	108.37	2.80	1.40
M12	Baykan	0.03	1.27	2.50	4.24	0.35	0.05	0.38	45.42	2.12	0.92

Table 4.6. The standard deviation of element analysis of thyme samples (N=3)

No	Name	Mo [µg g-1]	Cd [ng g-1]	Sn [µg g-1]	Sb [ng g-1]	Ba [µg g-1]	La [µg g-1]	Ce [µg g-1]	Pt [ng g-1]	Tl [ng g-1]	Pb [µg g-1]
M1	Qandil	0.19	5.23	0.27	9.79	0.26	0.01	0.01	1.46	1.41	0.06
M2	Asos	0.18	1.44	0.05	23.92	0.53	0.01	0.01	1.09	1.05	0.06
M3	Balambo	0.12	7.04	0.14	23.66	0.26	0.00	0.02	0.26	0.72	0.19
M4	Karokh	0.08	12.14	0.02	17.87	0.59	0.00	0.01	0.38	0.28	0.02
M5	Eruh	0.14	86.44	0.12	23.22	0.57	0.02	0.05	0.35	0.79	1.36
M6	Şirvan yolu Kezer	0.16	8.29	0.17	21.09	0.64	0.02	0.01	0.20	0.50	0.23
M7	Kampus	0.16	12.48	2.01	77.70	2.83	0.02	0.04	0.07	1.51	1.22
M8	Tillo	0.12	8.12	1.88	40.20	0.36	0.02	0.00	0.32	1.05	3.39
M9	Akdoğmuş Koyu	0.12	5.11	0.50	22.09	1.26	0.01	0.02	0.53	0.30	0.91
M10	Kurtalan Üçyolu	0.14	21.31	0.08	13.18	2.29	0.03	0.04	0.28	1.15	0.10
M11	Kurtalan Zokayt	0.11	16.83	0.27	22.94	5.03	0.01	0.06	0.25	3.36	0.01
M12	Baykan	0.12	7.58	0.09	9.03	0.36	0.00	0.02	0.30	1.20	0.01

Means and standard deviation of heavy metal concentration (part per million) in thymus plants are demonstrated in Table (4.1, 4. 2, 4. 3, 4. 4, 4. 5 and 4. 6) The outcomes exposed obvious differences in the amount of heavy metals produced from thyme plant grown at various geographical areas in Kurdistan region/ Iraq and Turkey.

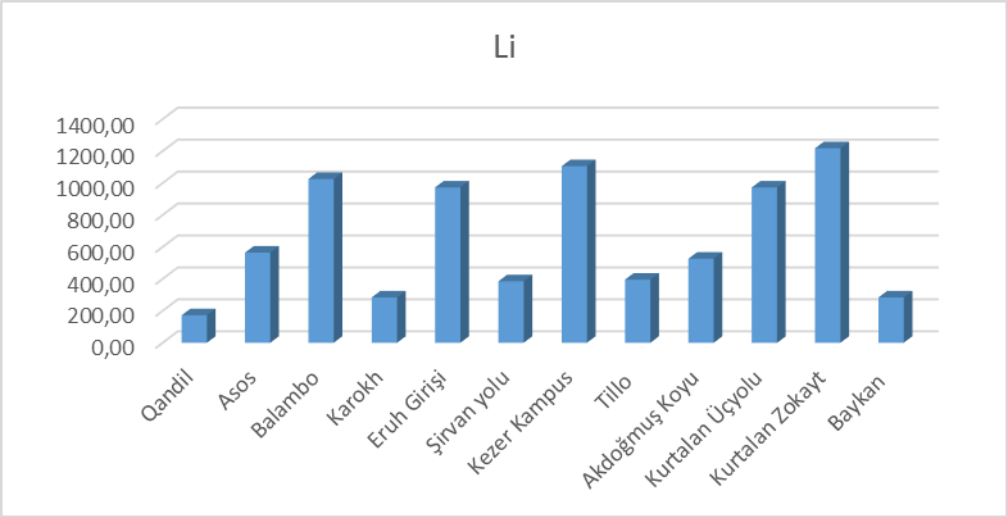

Figure 4.1. The concentration of Li in different locations. (ng g^{-1})

Figure 4.1 displays the concentration of Li element in different location. It is revealed that the highest amount of Li (1217.86±95.88 ng g^{-1}) was found in the Kurtalan (Zokayt-Şelale) following by Kezer Kampus (1105.52±85.04 ng g^{-1}) while the lowest metal content was 172.43±37.33 ng g^{-1} in the Qandil region.

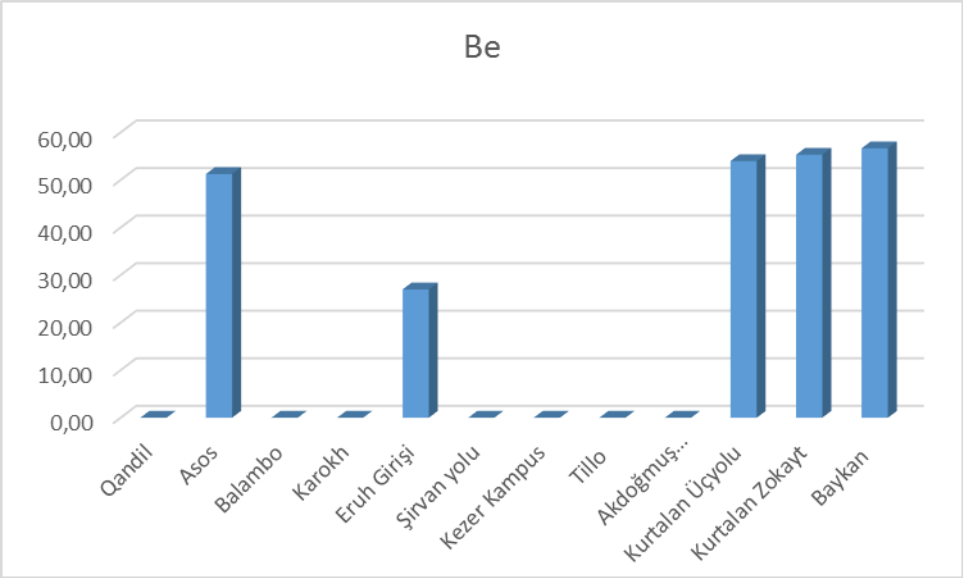

Figure 4.2. The concentration of Be in different locations.(ng g^{-1})

In this figure the amount of beryllium in twelve different location are demonstrated. The outcomes that the amount of Be was the largest (56.66±7.01 ng g^{-1}) in sanples collected from Baykan while the smallest quantity (26.99±2.34 ng g^{-1}) of Be was recorded in sample gathered in Eruh. Additionally, the Be was not detected in some samples such as in Qandil, Balambo, Eruh, Karokh, kezer Kampus and Koyu Kurtalan.

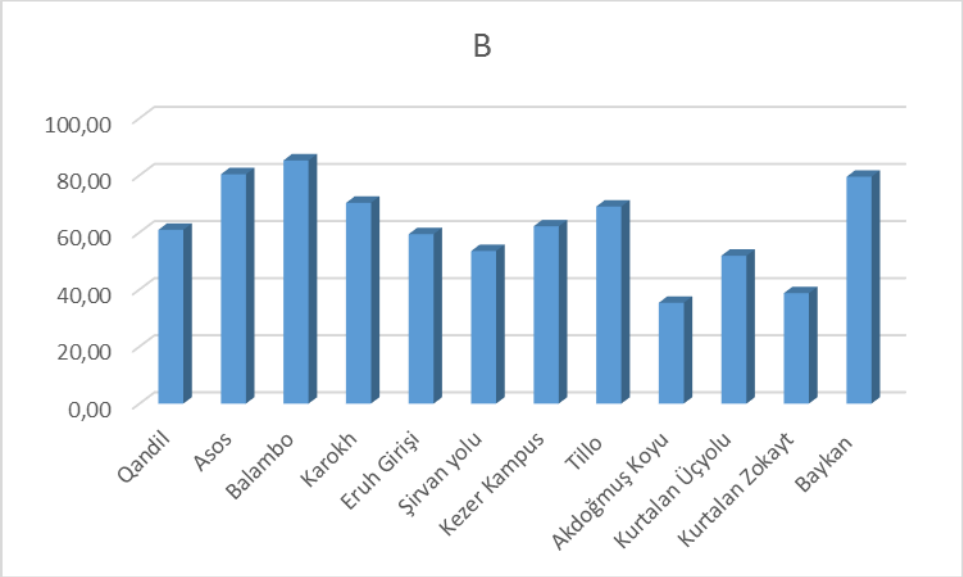

Figure 4.3. The concentration of B in different locations.(µg g-1)

The concentrations of boron in various sights are displayed in figure 4.3. it can be seen that the lowest level of B was 35.32±4.46 µg g^{-1} in Akdoğmuş Koyu. However, samples of Balambo contain the highest amount (85.19±7.66 µg g^{-1}). Morover, the concentration of

Boron in Qandil, Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu and Kurtalan (Zokayt-Şelale) were 60.89, 80.36, 70.35, 59.37, 53.47, 62.11, 69.04, 51.81, 38.70 and 79.49 respectively. (Pellerano et al, 2012) found that the average level of B is 46.23 µg g^{-1}.

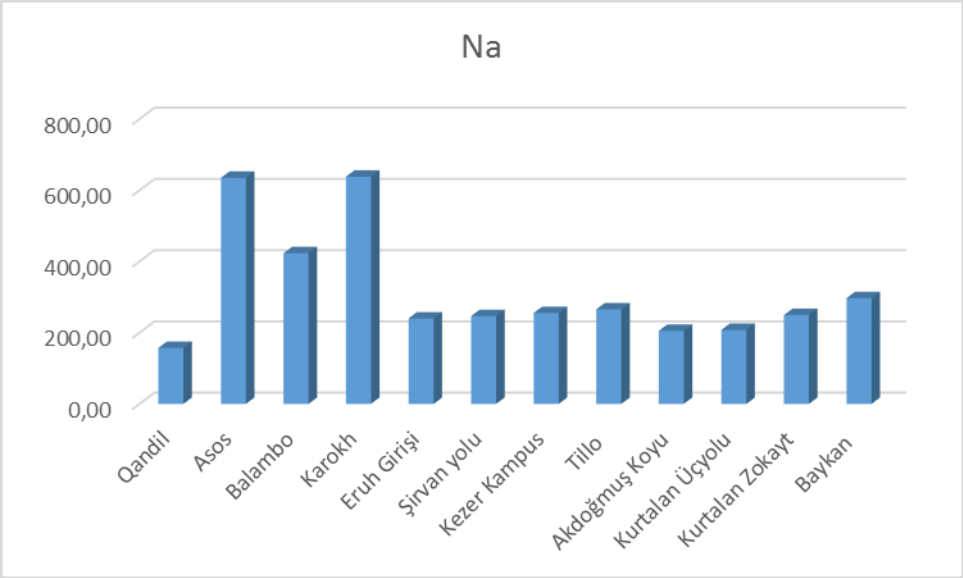

Figure 4.4 The concentration of Na in different locations.(µg g^{-1})

Figure 4.4 describes the level of sodium in deffrent positions in Kurdistan region of Iraq and Turkey. The outcomes uncovered that the mean values of Na were 157.6, 634.99, 442.99, 638.04, 239.90, 246.58, 255.19, 265.51, 205.30, 20778, 249.68 and 297.08 µg g^{-1} for Qandil, Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan and Baykan respectively.

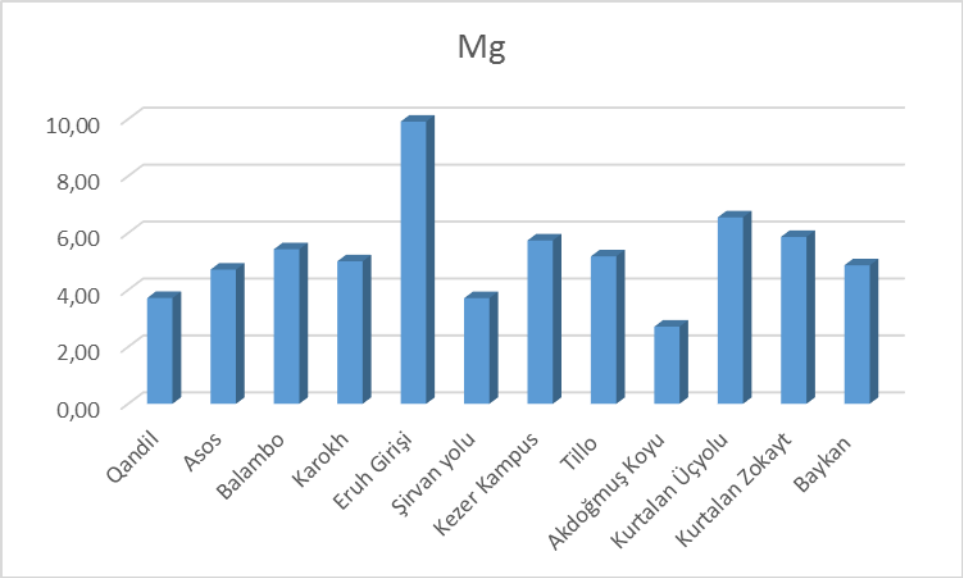

Figure 4.5. The concentration of Mg in different locations.(mg g^{-1})

The level of Mg was found to be varied according their location of cultivation. It can be seen in this figure that the highest Mg concentration (9.91±0.05 mg g^{-1}) was in the Eruh Girişi area. However, the minimum amount of magnesium (2.71±0.04 mg g^{-1}) was recorded in the Akdoğmuş Koyu region. These are clearly illustrated in the figure (4.5).

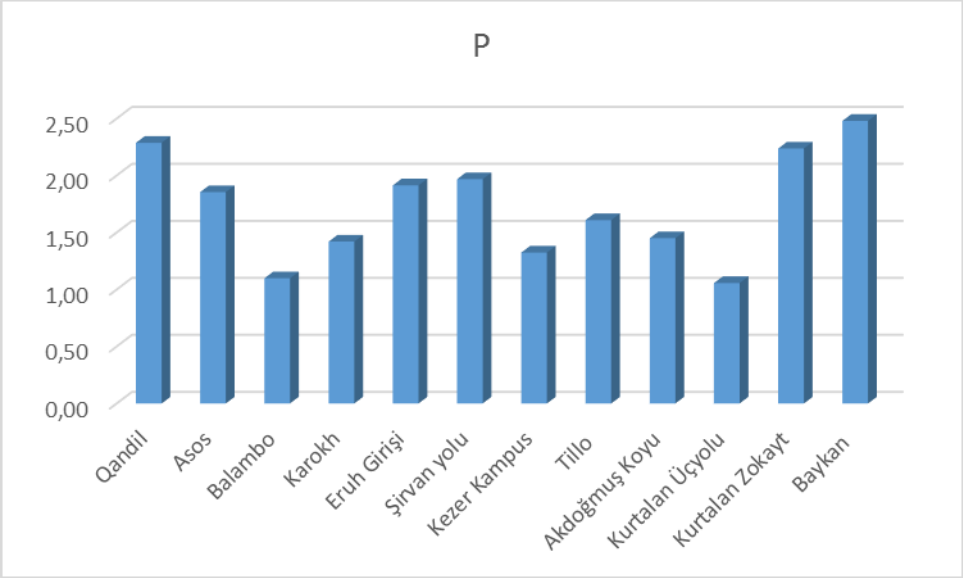

Figure 4.6 The concentration of P in different locations (mg g-1)

Figure 4.6 indicates the concentration of phosphate in thyme at different region of Siirt provenice and Kurdistan regional of Iraq lowest results recorded at Kampus which 1.06±0.05 mg g^{-1} and highest at 2.48±0.08 mg g^{-1} in Şirvan yolu. The amount of P in Qandil, Asos,

Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan (Zokayt-Şelale) and Baykan were 2.29, 1.85, 1.10, 1.42, 1.91, 1.97, 1.32, 1.61, 1.45, 1.06, 2.24 and 2.48 mg g-1 respectively.

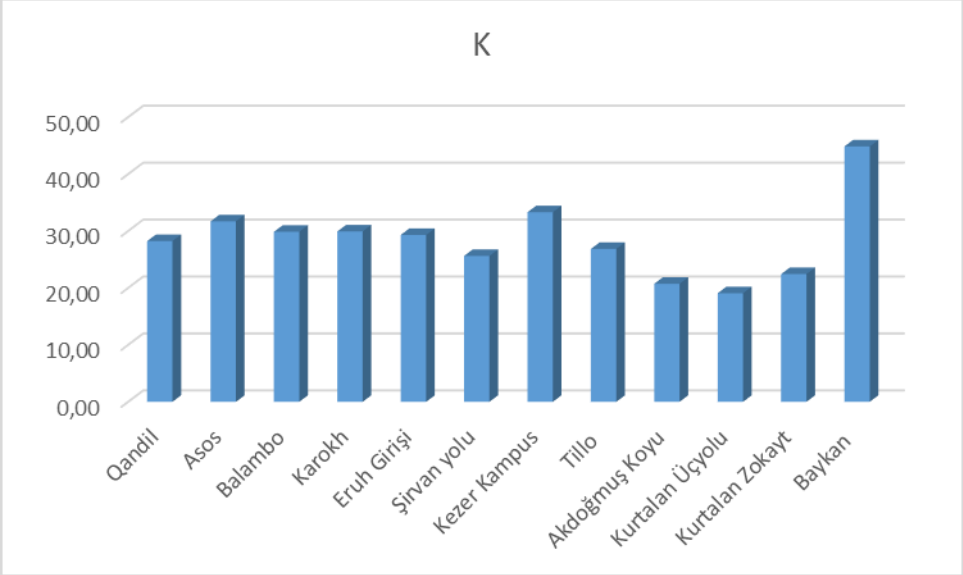

Figure 4.7. The concentration of K in different locations (mg g^{-1})

Figure 4.7 explains the level of potassium in thymus in cultivated in different locations. It is unveiled that the level of K is fluctuated according to the different location. Additionally, the highest level of K (44.86±0.73 mg g^{-1}) was recorded at Baykan. Nonetheless, the concentration of Potassium was the lowest (19.07±0.13 mg g^{-1}) in thymus collected from Kurtalan Üçyolu

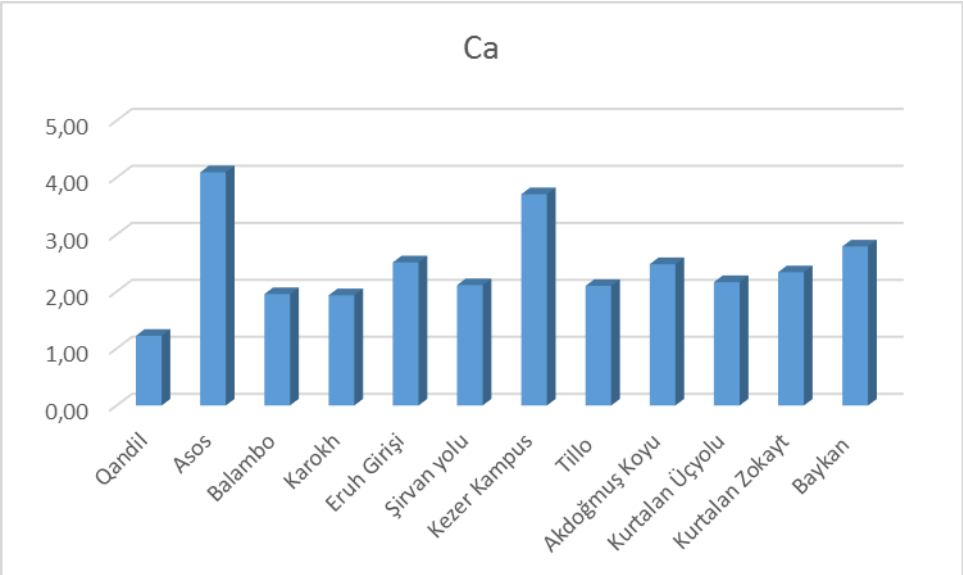

Figure 4.8. The concentration of Ca in different locations (mg g^{-1})

The figure 4.8 explains the calcium content in thumus plant collected in different location. This figure revealed that there are variations between the Ca content in different regions. It is also clear that the thymus plant in the Asos region had the highest Ca content 4.09 mg g^{-1} in comparison to the other investigated regions. On the other hand, the lowest Ca content 1.22 mg g^{-1} was recorded in samples taken from Qandil mountain.

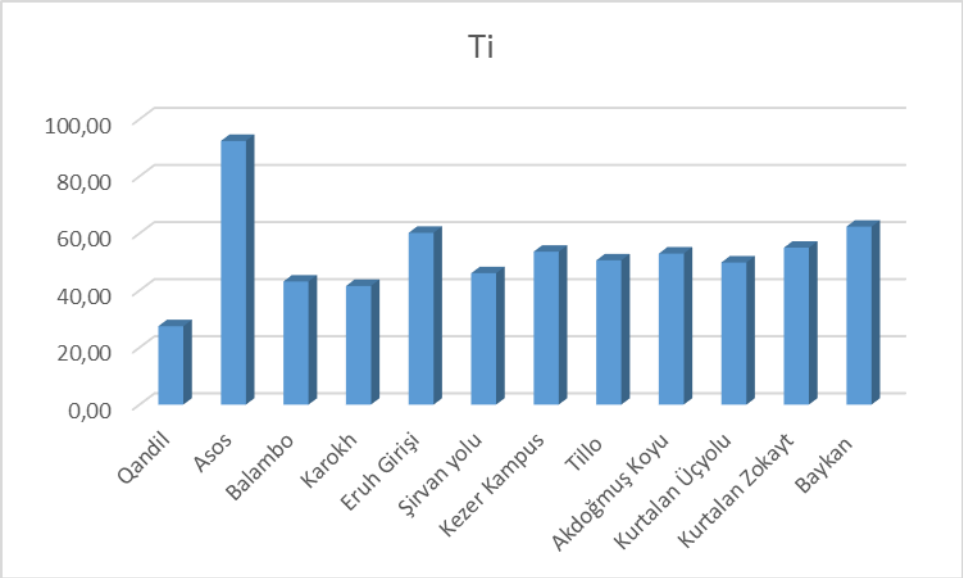

Figure 4.9. The concentration of Ti in different locations (ng g^{-1})

The figure above explains the level of titanium in thymus plants in various locations. It is discovered that the highest and the lowest level of Ti in these plant were 92.39±0.78 µg g^{-1} and 27.47±0.58 µg g^{-1} in Asos and Qandil respectively. Balambo, Karokh, Eruh, Şirvan yolu, Kezer Kampus, Tillo, kdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan (Zokayt-Şelale) and Baykan (Girişi) were 43.14, 41.59, 60.19, 46.04, 53.57, 50.55, 52.94, 49.80, 54.98 and 62.36 respectively.

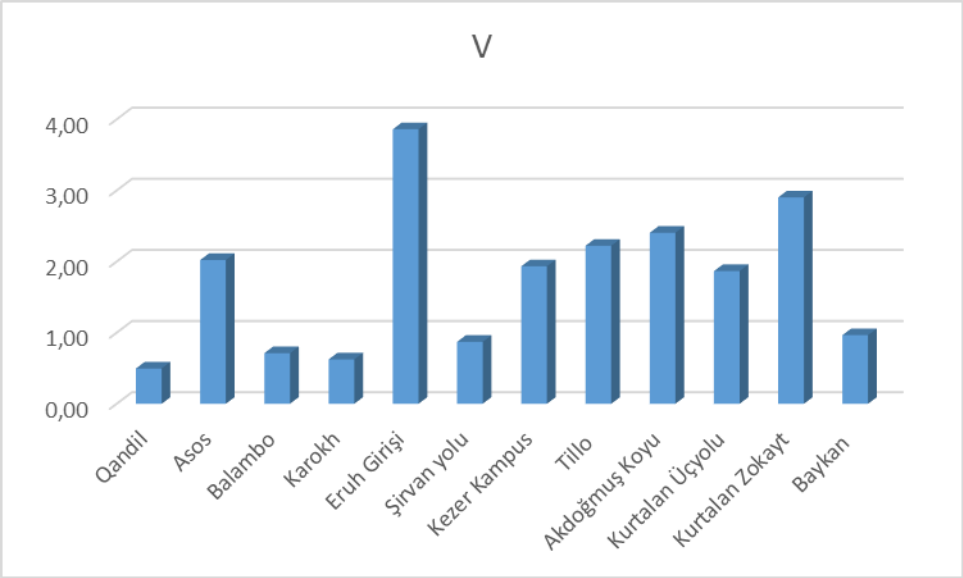

Figure 4.10. The concentration of vanadium in different locations (ng g^{-1})

This figure exhibits the concentration of vandium in different samples of thymus plants taken from various locations. Figure 4.10 shows that the level of vanadium is the highest (3.86±0.007 ng g^{-1}) in Eruh Girişi sample and the lowest (0.50±0.02 ng g^{-1}) in the Qandil sample. Furthermore, the amount of V content in the thymus taken from Asos, Balambo, Karokh, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan Zokayt-Şelale and Baykan (Girişi) were 2.02, 0.71, 0.62, 0.87, 1.93, 2.22, 2.41, 1.87, 2.90 and 0.97 accordingly.

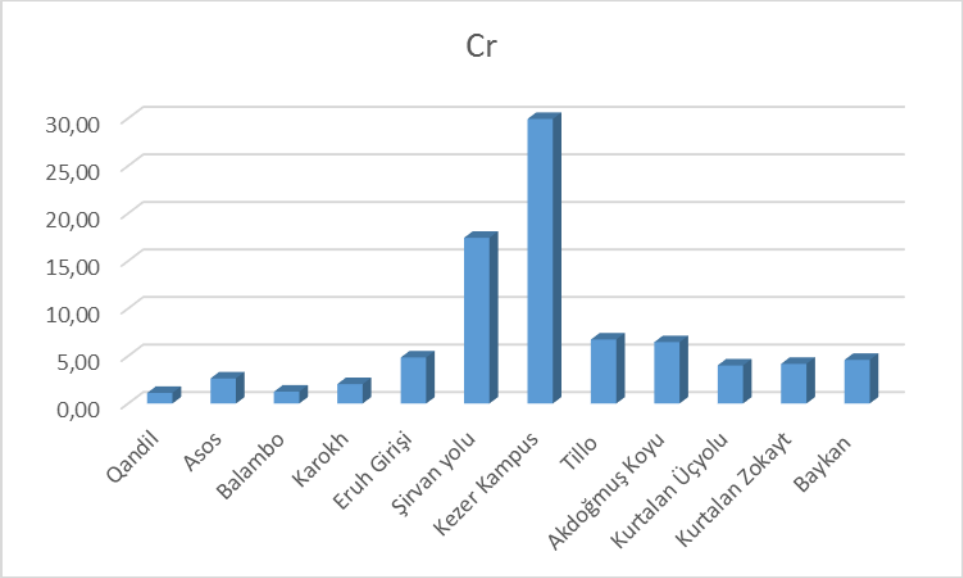

Figure 4.11 The concentration of Cr in different locations (µg g-1)

Additionally, a huge variations was observed in the concentration of Cr, where the highest amount of Cr was (29.93±0.25 µg g^{-1}) in Kezer Kampus and the lowest was (1.12±0.02 µg g^{-1}) in the Qandil region as shown in figure (4.11). Moreovere, the concentrations of crom were 2.64, 1.25, 2.05, 4.84, 17.43, 6.74, 6.46, 4.01, 4.18 and 4.59 in Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan (Zokayt-Şelale) and Baykan (Girişi) respectively. In addition, (Abu-Darwish Mohammad S et al, 2009) found that average level of Cr in thyme were less than 0.004 µg g^{-1}. In addition, (Pellerano et al, 2012) found that the level of Chromium is around 0.47 µg g^{-1}.

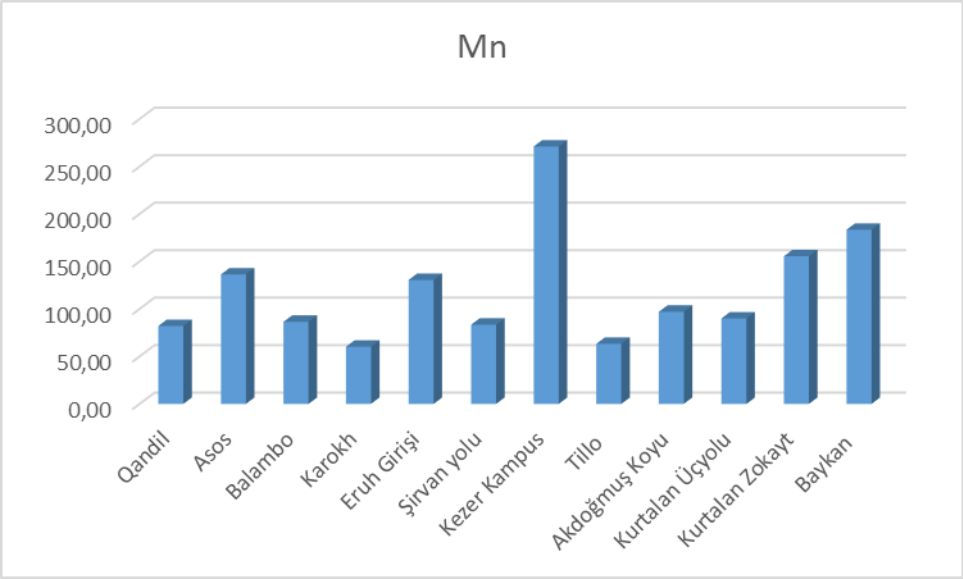

Figure 4.12. The concentration of Mn in different locations (µg g^{-1})

As it is seen in figure 4.12, the level of manganese was varied according their location of cultivationas. It can be seen from the figure that the highest Mn was 271.36±1.66 µg g^{-1} in the Zokayt area. However, the minimum amount of Mn was 60.23±0.15 recorded in the Karokh region. Furtheremore, the concentrations of Mn in Qandil Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan (Zokayt-Şelale) and Baykan (Girişi) were 82.03, 136.32, 86.49, 60.23, 130.49, 83.44, 271.06, 63.29, 97.11, 89.94, 155.49 and 183.39 respectively.

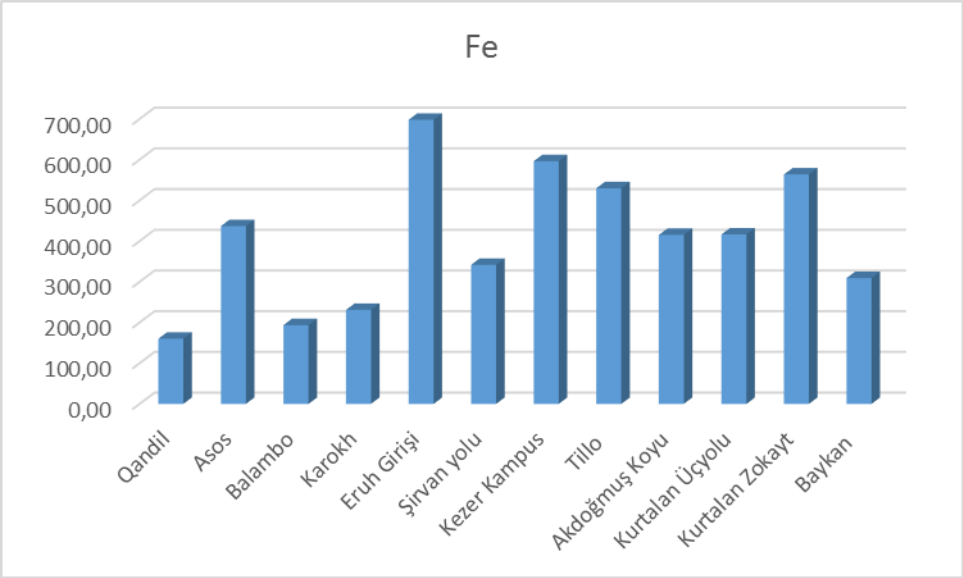

Figure 4.13. The concentration of Fe in different locations.(µg g^{-1})

Figure 4.13 indicates broad difference in level of Fe between the environmental regions of Turkey and Kurdistan region/ Iraq. The concentrations varied between 160.26 to 698.03 µg g^{-1}. Where the highest concentration of iron was (698.02±12.11) µg g^{-1}in the sample of thymus taken from Eruh Girişi whle the lowest concentration of this metal was (160.26±3.19) µg g^{-1} in samples of Qandil region. Abu-Darwish, Mohammad S et al (2009) stated that the level of Fe in the thyme in different regions of Jordan is 15.2 to 205.08 ppm. According to (Pellerano et al, 2012) stated that the level of Fe is 6.44 µg g^{-1}.

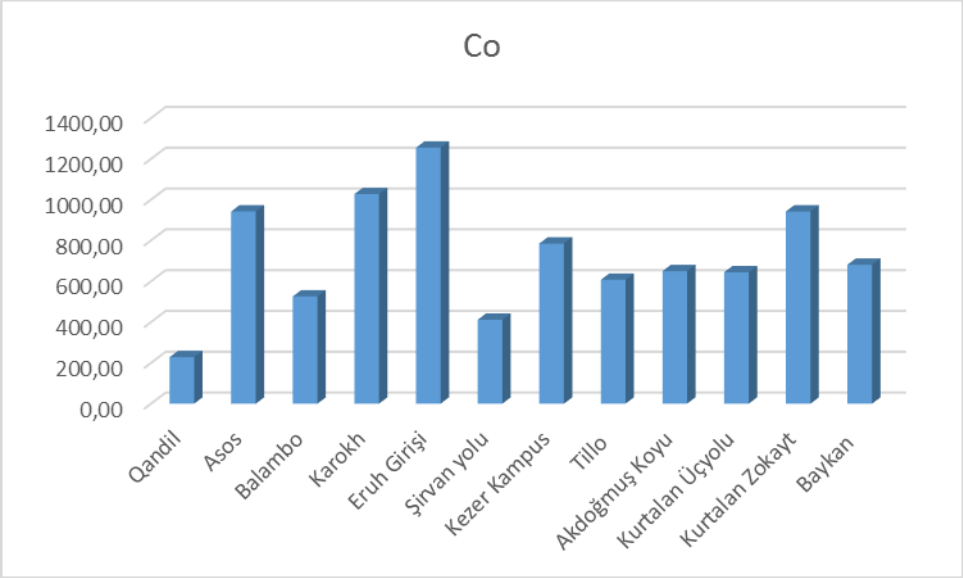

Figure 4.14. The concentration of Co in different locations (ng g^{-1})

It can be seen from the figure number 4.14 that the outcomes of cobalt (Co) level in Qandil, Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan (Zokayt-Şelale) and Baykan (Girişi) were 228.10, 924.32, 525.71, 1027.22, 1254.79, 411,07, 685.07, 607.44, 649.76, 644.21, 941.46 and 680.86 ng g^{-1} respectively.

This variation was higher in comparison to several average Co in some medicinal herbs eaten in Turkey. According to (Syamasri et al, 1994) the cobalt dissemination in plants is completely species reliant. Some factors such as temperature, salinity, pH of the intermediate, and existence of other metals impacts the process of Co gathering in medicinal plant. According to (Pellerano et al, 2012), reported that the concentrationof Co is 12.8 and Abu-Darwish, Mohammad S., et al (2009) reported that the level of Co in several location in jordan is rangend from (0.4 to 5.0) ng g^{-1}.

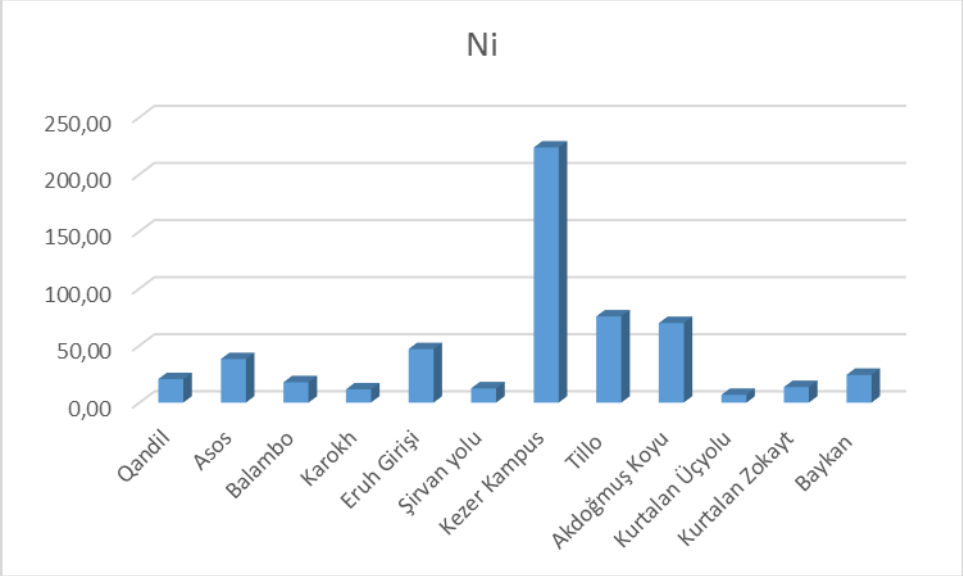

Figure 4.15. The concentration of Ni in different locations (µg g^{-1})

In this figure the concentration of nickel in thymus plant collected in several areas. It is appeared that the highest concentration of nikle was 223.46±10.37 µg g^{-1} in the sample of thymus that collected in the Kezer Kampus while the lowest level 6.85±0.14 µg g^{-1} of Nickle was found in thymus which are taken from Kurtalan Üçyolu.Nickel is required for normal animals development and reproduction. It seems toplay numerious role in the modulation of the immune system and in brain development. However, (Abu-Darwish, Mohammad S et al,

2009) confirmed that Ni level in thymus in Jordan is less than (0.07 µg g^{-1}). (Pellerano et al, 2012).

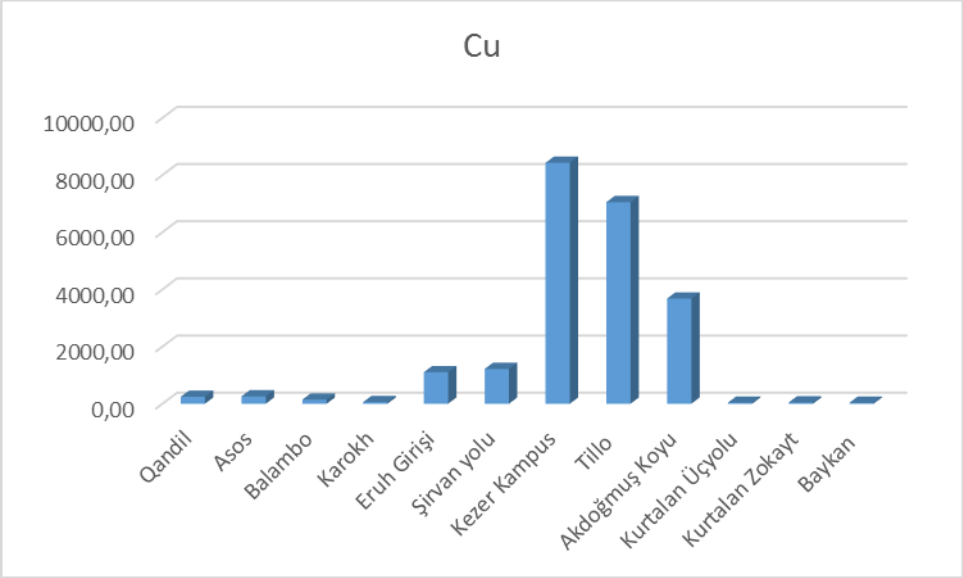

Figure 4.16. The concentration of Cu in different locations (µg g^{-1})

The level of Copper (Cu) in different location is demonstrated in figure 4.16. The concentration of copper (Cu) were significantly higher (1216.98±5.86 µg g^{-1}) in the Şirvan yolu area in comparison to the other regions. This significant concentration might be to the heavy traffic of automobile on the main route of transportation to the factory runs through the region. However, the lowest level of Cu was recorded in the Baykan. (Abu-Darwish, Mohammad S et al, 2009) found that thymus in Jordan has the content of Cu ranged from 8.62 to 10.40 ppm.

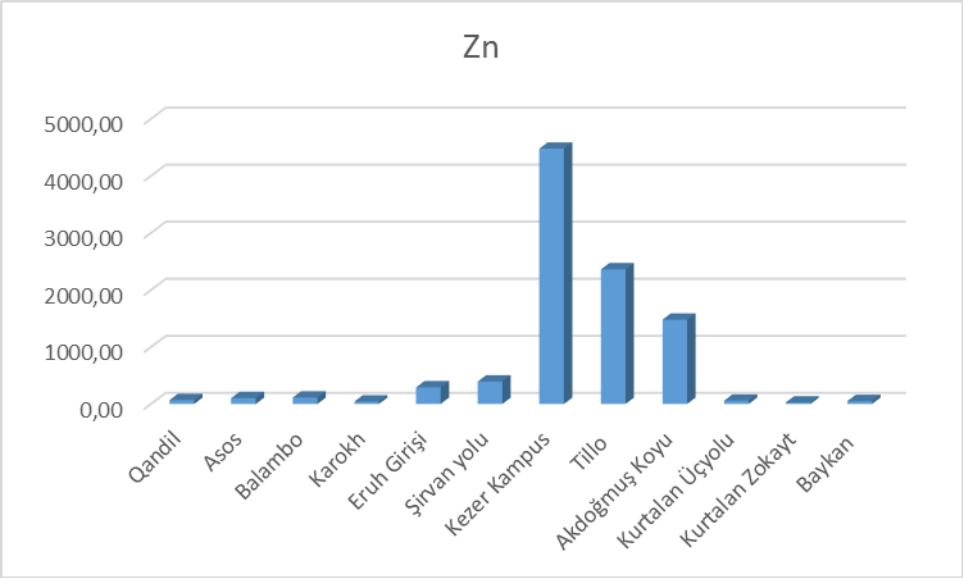

Figure 4.17. The concentration of Zn in different locations (µg g^{-1})

The amount of zinc content in thymus plants collected from different areas are displayed in this figure. The variation in the concentration of Zn was obviously shown in the chart (4.17). It can be monitored that the lowest level of Zinc was 26.58±0.53 µg g^{-1} in the sample collected in thyme grown in the Kurtalan (Zokayt-Şelale) while the maximum concentration of zinc was 4477.43±45.29 µg g^{-1} in thyme plant cultivated at Kezer Kumpus. (Pellerano et al, 2012), stated that the concentration of Zn is 0.85 µg g^{-1}.

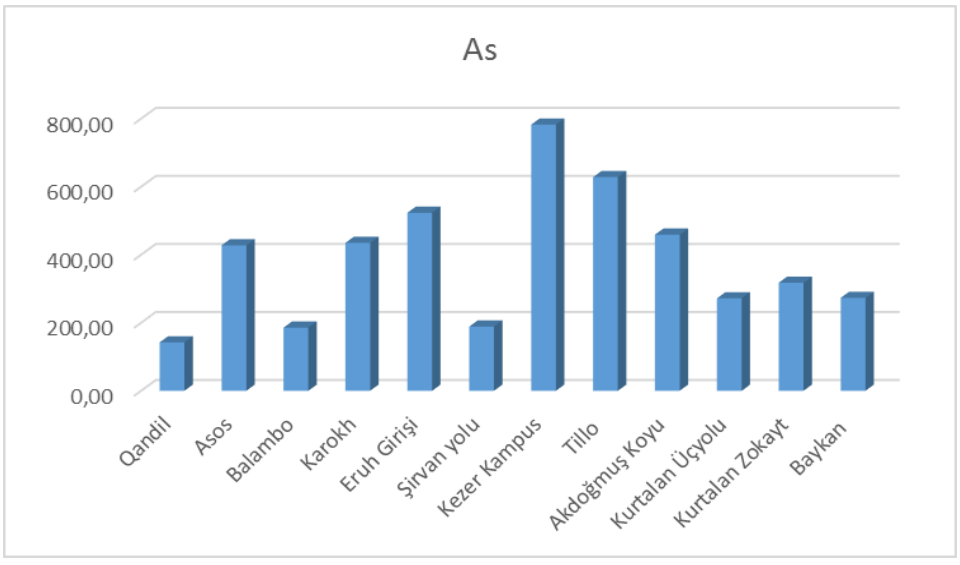

Figure 4.18. The concentration of As in different locations (ng g^{-1})

Figure 4.18 illustrates the level of arsenic in various thyme samples collected from different locations. In addition, the outcomes of the study revealed that the level of arsenic

showed a broad difference between the environmental regions of Turkey and Kurdistan region/ Iraq. The concentration varied between 782.55 ng g^{-1} Kampus to 142.34 ng g^{-1} Qandil. Furthermore, the concentration of As in Qandil, Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan (Zokayt-Şelale) and Baykan (Girişi) were 142.34, 427.62, 185.74, 434.90, 523.39, 188.81, 782.55, 628.19, 458.94, 271.88, 317.62 and 273.17 ng g^{-1} respectively.

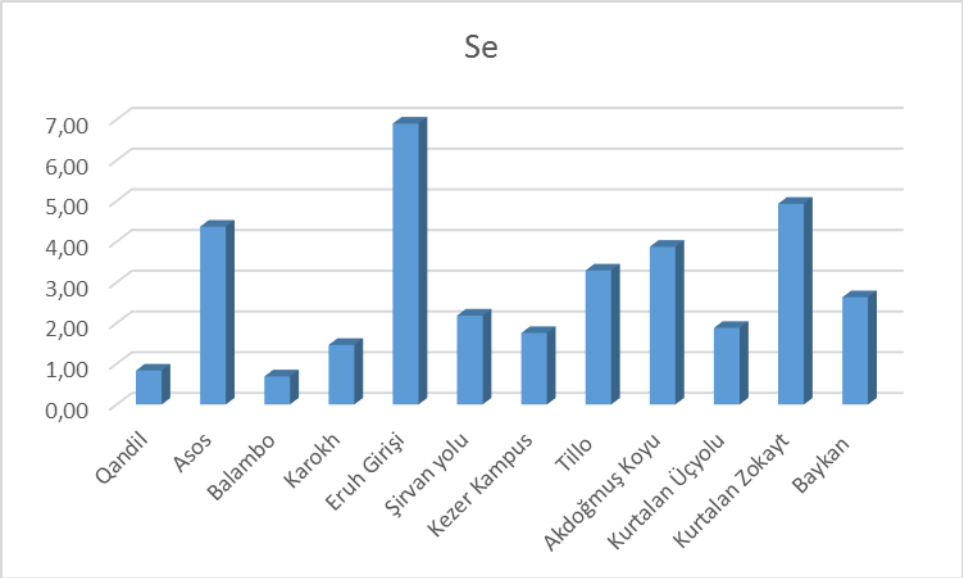

Figure 4.19. The concentration of Se in different locations (µg g^{-1})

This figure illustrates the amount of Se in different samples. The results of the study revealed that the level of selenium demonstrated a broad variation between the environmental regions of Turkey and Kurdistan region/ Iraq. The concentration varied between 2.82 µg g^{-1} Asos and Tillo to 0.5 µg g^{-1} Qndil. The concentration of Se by µg g^{-1} were 0.83, 4.37, 0.69, 1.46, 6.90, 2.18, 1.76, 3.29, 3.87, 1.88, 4.93 and 2.63 in thyme plant caltivated in Qandil, Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan (Zokayt-Şelale) and Baykan respectively.

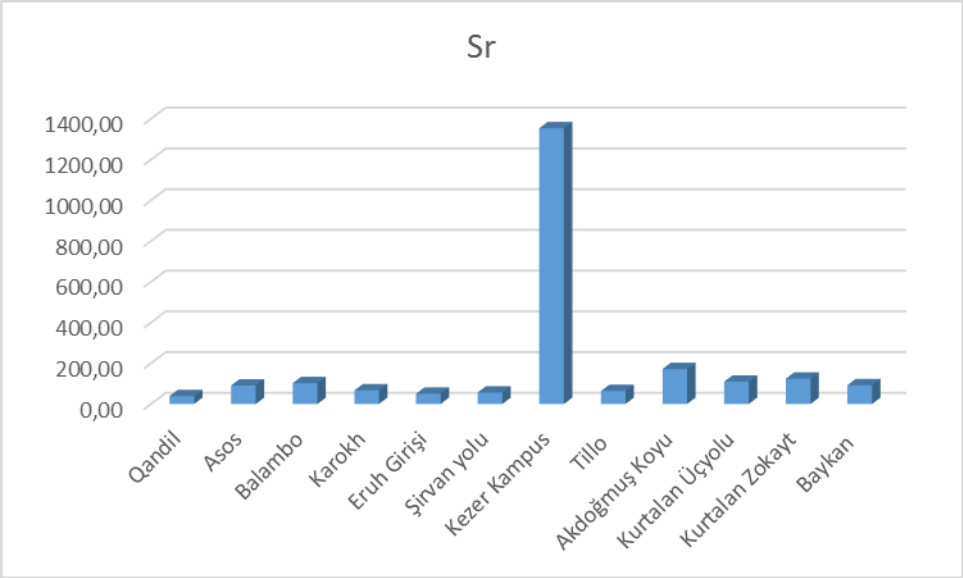

Figure 4.20. The concentration of Sr in different locations (µg g^{-1})

The level of Strontium in thyme plant grown in different locations. Obviously, the variation in the concentration of Sr was presented in the figure 4.20. It can be monitored that the lowest level of Sr was 38.44±1.11 µg g^{-1} in thyme grown in the Qandil while the maximum concentration of Sr was 1353.63±15.42 µg g^{-1} gathered in plant cultivated at Kezer Kampus. It has to be mentioned that the concentration of Sr in thyme of Kezer Kampus has significantly higher Sr content than the other investigated locations.

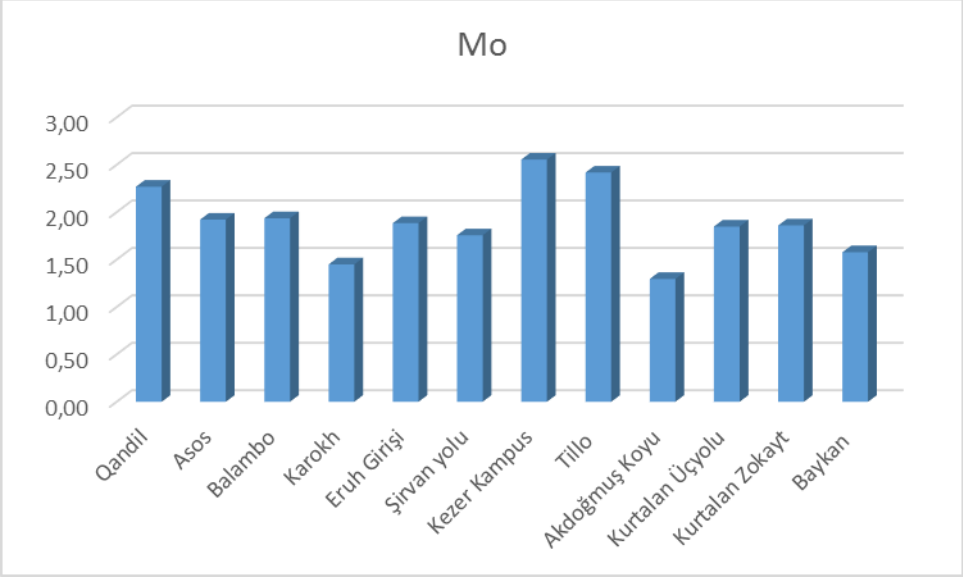

Figure 4.21. The concentration of Mo in different locations (µg g^{-1})

The concentration of molybdenum is displayed in figure 4.21. It is revealed that the content of Mo in thyme collected in Qandil, Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan Zokayt-Şelale and Baykan were 2.27, 1.92, 1.94, 1.45, 1.88, 1.76, 2.55, 2.42, 1.30, 1.85, 1.86 and 1.58 respectively. The highest level of Mo was 2.55±0.19 µg g^{-1} which recorded in thyme from Kezer Kampus area. Nonetheless, thymus plant from Akdoğmuş Koyu region contained the lowest level of Mo 1.30±0.12 µg g^{-1}

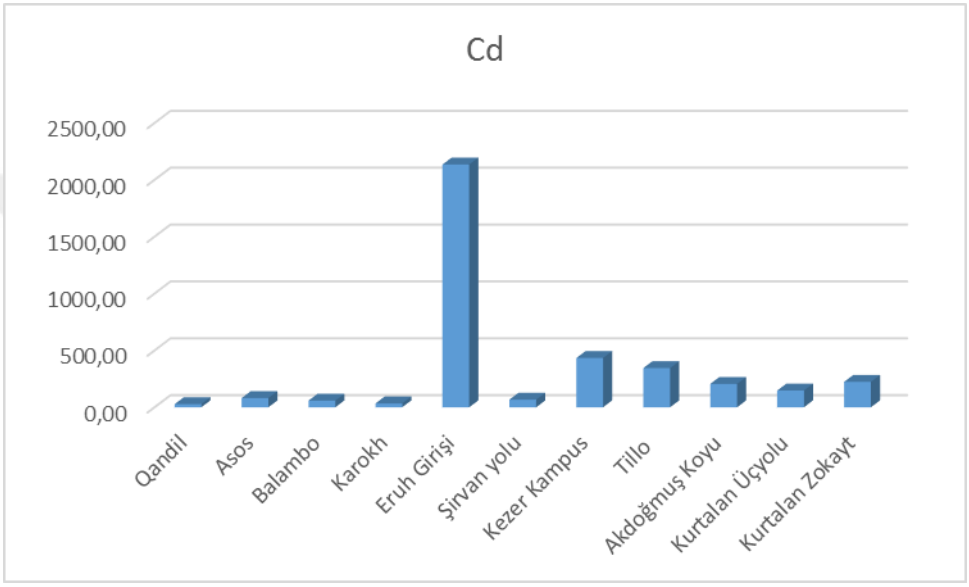

Figure 4.22. The concentration of Cd in different locations (ng g^{-1})

The concentrations of Cadmium (Cd) in Qandil were 27.89±5.23 ng g^{-1} however it was 2134.38±86.44 and 433.84±12.84 ng g^{-1} in Eruh and Kezer Kampus accordingly. It has to be mention that the level of Cd in the Qandil region was lower than the average poisonous level in the plant. Nonetheless, the Cd level in diverse studied zones were in general lower than those suggested by World Health Organization (WHO). The low level of Cd in Qandil area might be because of a low Cd content in soil of the investigated zone. Nevertheless, high concentration of Cd in the other regions could be because of growing traffic and factory density. These outcomes are agreed with the outcomes of (El-Rjoob et al 2008). Also, Abu-Darwish, Mohammad S, et al (2009) found that the level of Cd in thyme of Jordan is varied between 0.30 and 1.30 ppm.

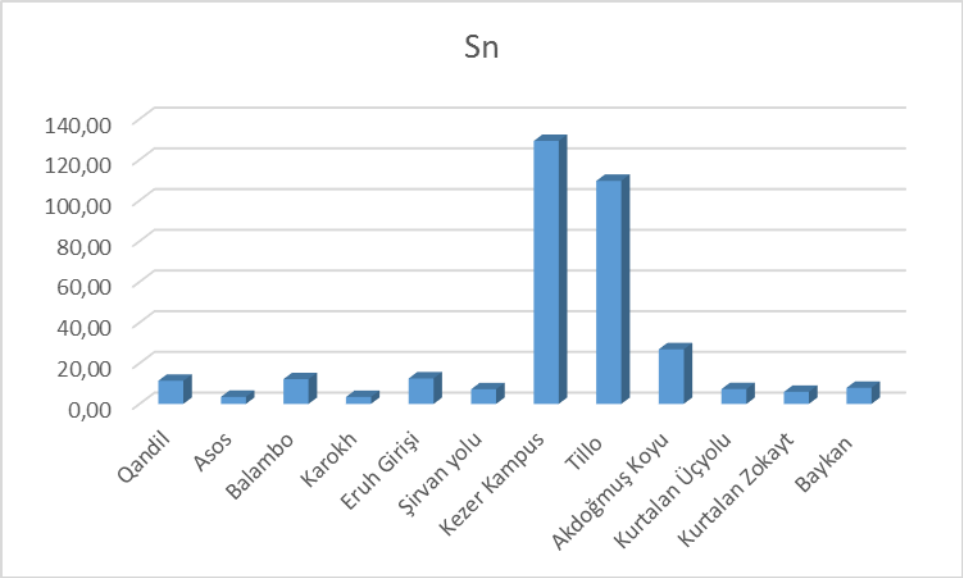

Figure 4.23. The concentration of Sn in different locations (µg g^{-1})

Figure 4.23 demonstrates the concentration of tin (Sn) in thyme collected from different regions in Kurdistan region/ Iraq and Turkey. The biggest amount of Sn content was recorded in thymus of Kezer Kampus region while the lowest concentrations of (Sn) was 3.42±0.14 µg g^{-1} in Karokh same. However, the highest concentration of Sn was 129.21±2.01 µg g^{-1} in Kezer Kampus. The level of Sn was 11.36, 3.47, 12.15, 12.52,7.22, 109.61, 26.84, 7.27, 5.94 and 7.91 in Qandil, Asos, BalamboEruh Girişi, Şirvan yolu, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan Zokayt-Şelale and Baykan respectively.

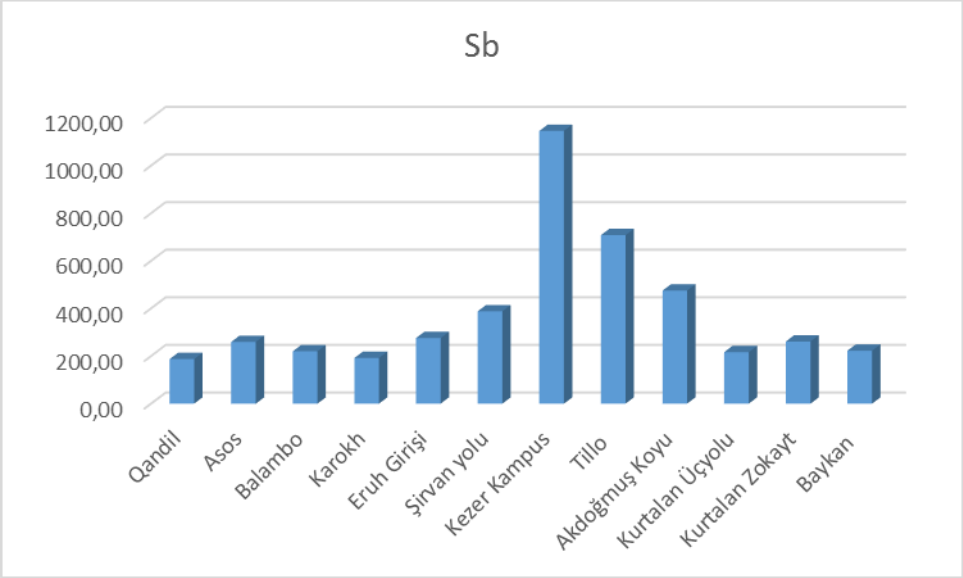

Figure 4.24. The concentration of Sb in different locations (ng g^{-1})

As displayed in figure (4.24) The mean levels of antimony (Sb) by (ng g^{-1}) were 187.33, 259.28, 220.09, 192.23, 275.85, 387.98, 1146.00, 708.07, 475.44, 216.28, 260.90, and 222.60 in Qandil, Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan (Zokayt-Şelale) and Baykan (Girişi) respectively. The highest level was 1145.99±77.70 ng g^{-1} of Sb was in thyme plant taken from Kezer Kampus. Nonetheless, the lowest amount of Sb was 187.33±9.79 ng g^{-1} in Qandil. The level of 6.90 ng g^{-1} was found by (Pellerano et al, 2012).

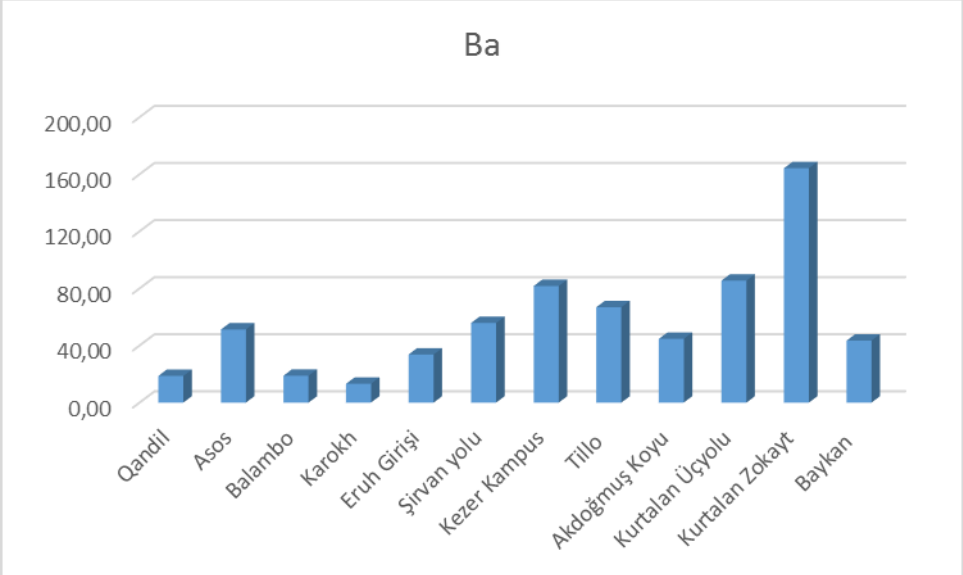

Figure 4.25. The concentration of Ba in different locations (µg g^{-1})

Figure 4.25 describes the concentration of barium (Ba) in several regions of Kurdistan region/ Iraq and Turkey. The concentration of Ba was considerably higher 164.26 µg g^{-1} in the Zokayt area in comparison to the other regions. However, the lowest level of Ba was 13.22 µg g^{-1} in Karokh. The level of Ba in Qandil, Asos, Balambo, Eruh, Şirvan yolu, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan Zokayt-Şelale and Baykan were 18.70, 51.26, 18.85, 13.22, 33.68, 55.81, 81.70, 66.84, 44.69, 85.46, 164.27 and 43.44 respectively.

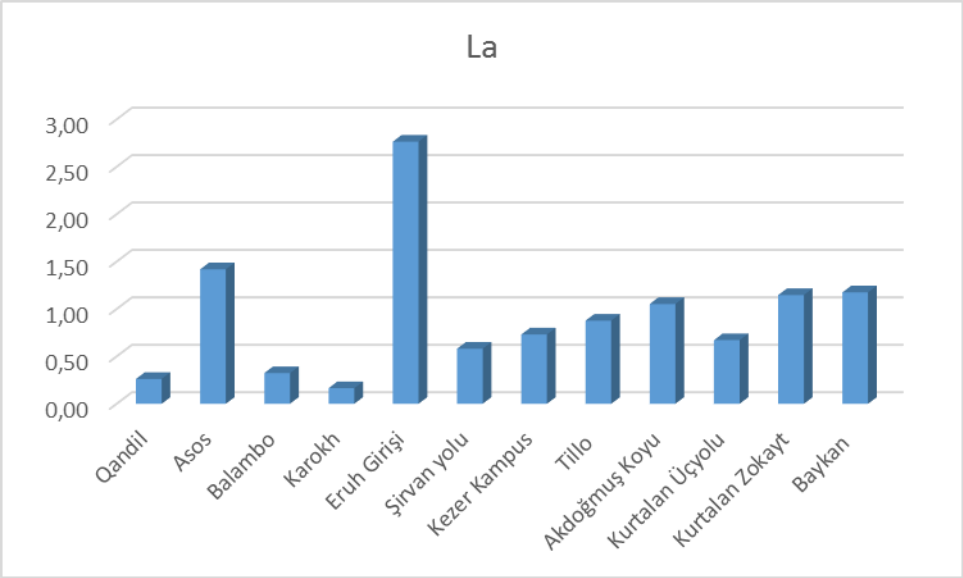

Figure 4.26. The concentration of La in different locations (µg g^{-1})

This figure describes the amount of lanthanum (La) in different locations. It can be seen than the high level of La was 2.76±0.57 µg g^{-1} while the lowest level La was 0.26±0.01 µg g^{-1} in Qandil region. The level of La were 0.26, 1.42, 0.32, 0.17, 2.76, 0.58, 0.73, 0.88, 1.05, 0.67, 1.15 and 1.18 in Qandil, Asos, BalamboEruh Girişi, Şirvan yolu, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu, Kurtalan Zokayt-Şelale and Baykan respectively. Pellerano, et al (2012) found that the average level of La was 28.84 µg g^{-1}.

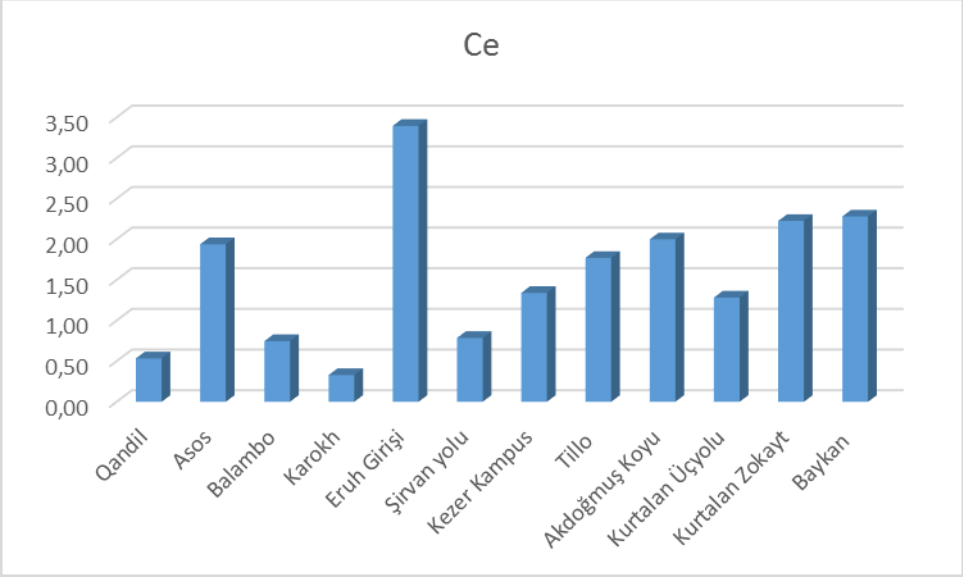

Figure 4.27. The concentration of Ce in different locations (µg g^{-1})

In this figure, the concentration of cerium (Ce) in various locations in kurdstan region/ Iraq and Tuekey is illustrated. It can be seen that there are a considerable differences in the level of Ce between the samples. The highest concentration of Ce was 3.39±0.05 µg g^{-1}in Eruh whearas the lowest concentration of Ce was 0.32±0.320.01 µg g^{-1} in Karokh. According to (Pellerano et al, 2012) found that the level of Ce is 50.22 µg g^{-1}

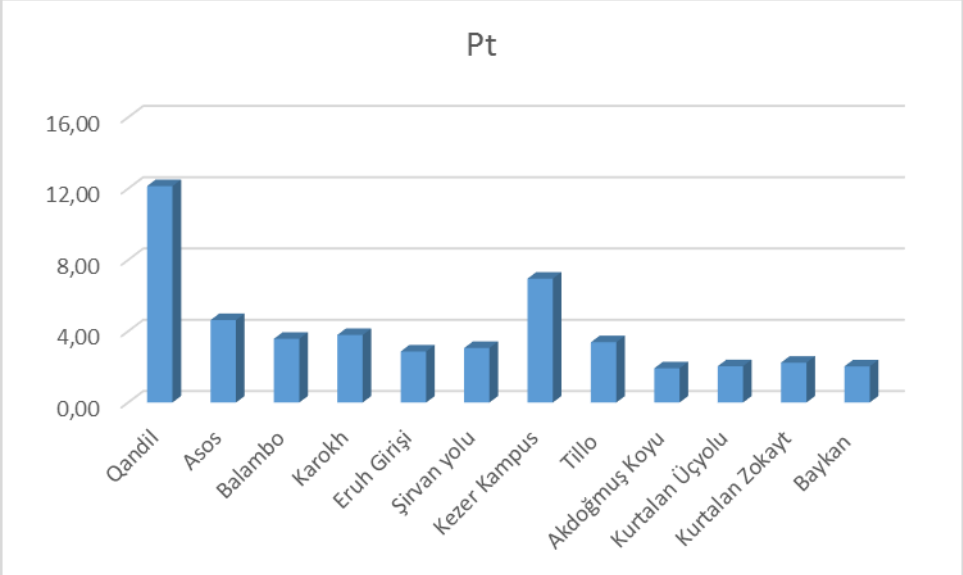

Figure 4.28. The concentration of Pt in different locations (ng g^{-1})

Figure 4.28 describes the amount of Pt content in thymus plant collected in different locations. It is revealed that the largest amount of Pt was 12.14±1.46 ng g^{-1} in thyme of Qandil region. However the lowest concentration of platnium was recorded in the sample taken from Akdoğmuş Koyu which was 1.91±0.53 ng g^{-1}

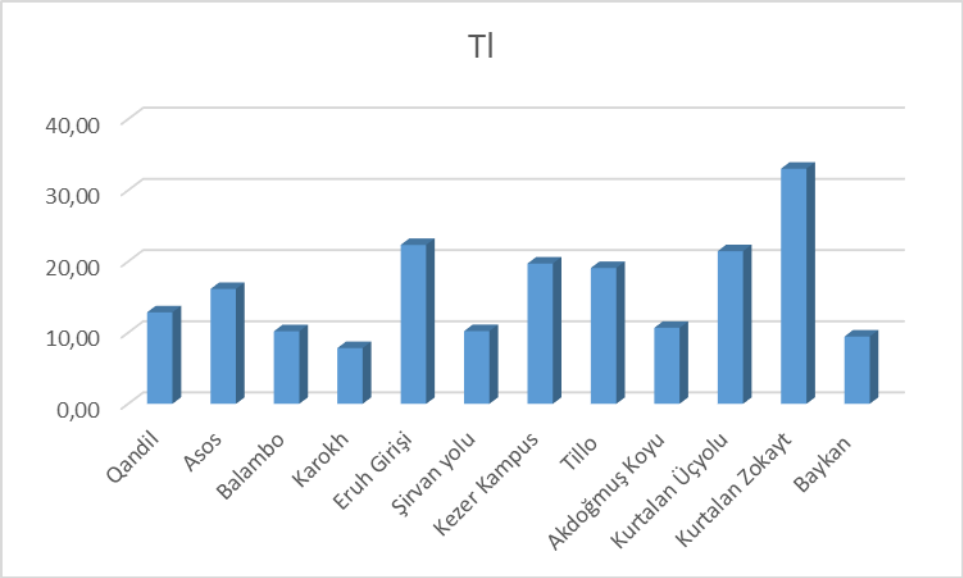

Figure 4.29. The concentration of TI in different locations (ng g^{-1})

This figure uncovered that the level of TI is broadly differed in various environmental regions of Turkey and Kurdistan region/ Iraq. In this figure, it can be seen that the concentration of TI is the highest in 33.03±1.15 ng g^{-1} in Kurtalan (Zokayt-Şelale) region. However the level of was lowest 7.83±0.28 ng g^{-1} in Karokh region.

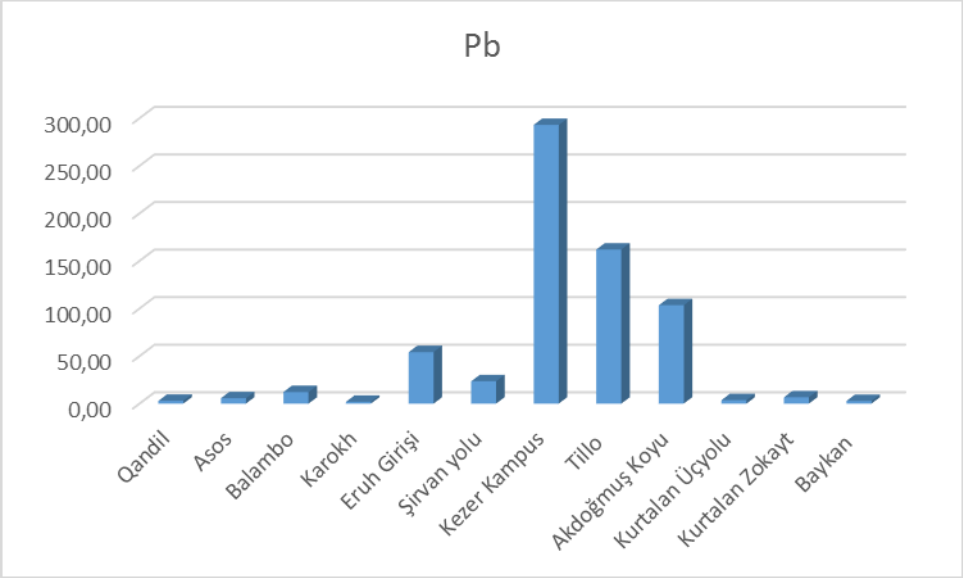

Figure 4.30. The concentration of Pb in different locations (µg g^{-1})

As displayed in figure 4.30 The mean levels of lead (Pb were 3.71, 4.72, 5.43, 5.00, 9.91, 3.71, 5.74, 5.18, 2.71, 6.55, 5.86 and 4.87 in thyme cultivated in Qandil, Asos, Balambo, Karokh, Eruh Girişi, Şirvan yolu, Kezer Kampus, Tillo, Akdoğmuş Koyu, Kurtalan Üçyolu,

Kurtalan (Zokayt-Şelale)and Baykan accordingly. The results revealed that while lower level Pb was 1.96±0.02 μg g^{-1} in Karokh areas, highest level of Pb was 292.89±1.22 μg g^{-1} in Kezer Kampus area. The outcomes of this study were between the normal limits of Pb concentration (5-10 ppm) in thyme plant. In the study of (Abu-Darwish, Mohammad et al, 2009), the Pb is 0.05 to 1.45 ppm. (Serbula et al, 2013) also found similar findings.

4.2. Antioxidant activity

In this section the results of antioxidant activity will be illustrated.

Table 4.7. The site of thyme collection which represented by K1 to K12.

Sample	K1	K2	K3	K4	K5	K6	K7	K8	K9	K10	K11	K12
Site	Qandil	Asos	Balambo	Karokh	Eruh	Şirvan yolu	Kezer Kampus	Tillo	Akdagmus	Kurtalan (üç yol)	Kurtalan zokayt	Baykan

It was thought the impact of antioxidants on DPPH radical scavenging to be because of their hydrogen-donating capability. Radical DPPH is a constant free radical and receives hydrogen radical or an electron for becoming a constant diamagnetic molecule. Capability of the scavenging of essential oils and positive control (BHA) are demonstrated in table 4.8.

Table 4.8. The extract solution in different concentration of methanol.

Sample	1mg/ml	0,5mg/ml	0,2mg/ml	1mg/ml	0,5mg/ml	0,2mg/ml	Ic 50 (mgIml)
K1	0.094	0.098	0.229	84.56	83.91	62.40	0.16
K2	0.097	0.17	0.416	84.07	72.09	31.69	0.36
K3	0.095	0.104	0.417	84.40	82.92	31.53	0.29
K4	0.17	0.327	0.472	72.09	46.31	22.50	0.61
K5	0.104	0.225	0.448	82.92	63.05	26.44	0.45
K6	0.115	0.312	0.482	81.12	48.77	20.85	0.56
K7	0.104	0.252	0.458	82.92	58.62	24.79	0.48
K8	0.101	0.154	0.391	83.42	74.71	35.80	0.3
K9	0.114	0.27	0.462	81.28	55.67	24.14	0.51
K10	0.1	0.207	0.396	83.58	66.01	34.98	0.36
K11	0.173	0.328	0.481	71.59	46.14	21.02	0.62
K12	0.375	0.477	0.546	38.42	21.67	10.34	1.32

Table 4.8 shows the inhibition activity of DPPH in thymus plant collected in different location, which are represented by letter K. the DPPH inhibition activity varied significantly

among the samples. It can be observed that the maximum DPPH inhibition activity was occurred in the sample K12. Nonetheless, DPPH activity was the lowest in sample K1. The heavy metal activities rely on numerous molecules structural characteristics. This might be attributed chiefly to their phenolic content, especially thymol and carvacrol. In addition, the strong activity of DPPH radical scavenging of those composites is well identified. Moreover, antioxidant activity might be influenced by several other elements like temperature, concentration, physical state of the system, light, kinds of materials and micro-components acting as pro-oxidants or synergists.

4.2.1. Tatol phenolic Compounds

Total phenolic analysis was done according to (Sinkard and Singleton, 1977). According to the results of the analysis, the total phenolic concentration in 1 ml extract in the extracts prepared with methanol was given. Total phenolic concentrations were given as gallic acid equivalents. Calculation of gallic acid equivalence is done by our laboratory during our work. The given gallic acid was calculated according to the standard regression curve. According to these results, the highest value showed the K1 sample (226,830 ug / ml) and the lowest value K12 sample (46.862 ug / ml). Concentration results of the samples are plotted (Figure 4.31).

Table 4.9 The total phenolic

Sample	Absorbance 1	Absorbance 2	Absorbance 3	Mean	SD	Concentration (ug/ml)
K1	1.321	1.39	1.72	1.48	0.21	226.830
K2	0.921	0.914	0.967	0.93	0.03	144.641
K3	1.171	1.183	1.217	1.19	0.02	183.440
K4	0.502	0.498	0.5	0.50	0.00	78.950
K5	0.738	0.763	0.848	0.78	0.06	121.785
K6	0.645	0.66	0.686	0.66	0.02	103.723
K7	0.673	0.706	0.734	0.70	0.03	109.878
K8	0.744	0.76	0.763	0.76	0.01	117.648
K9	0.62	0.619	0.591	0.61	0.02	95.600
K10	0.832	1.048	1.058	0.98	0.13	151.503
K11	0.636	0.649	0.534	0.61	0.06	95.045
K12	0.304	0.28	0.28	0.29	0.02	46.862

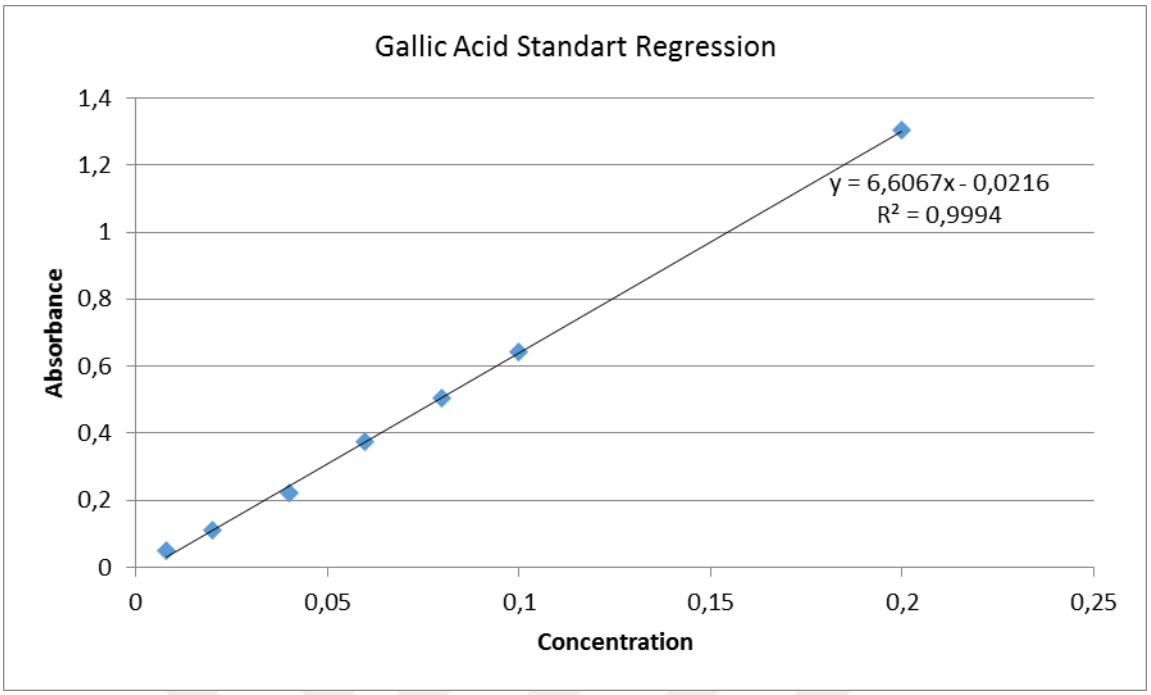

Figure 4.31. Gallic Acid Standart Regression

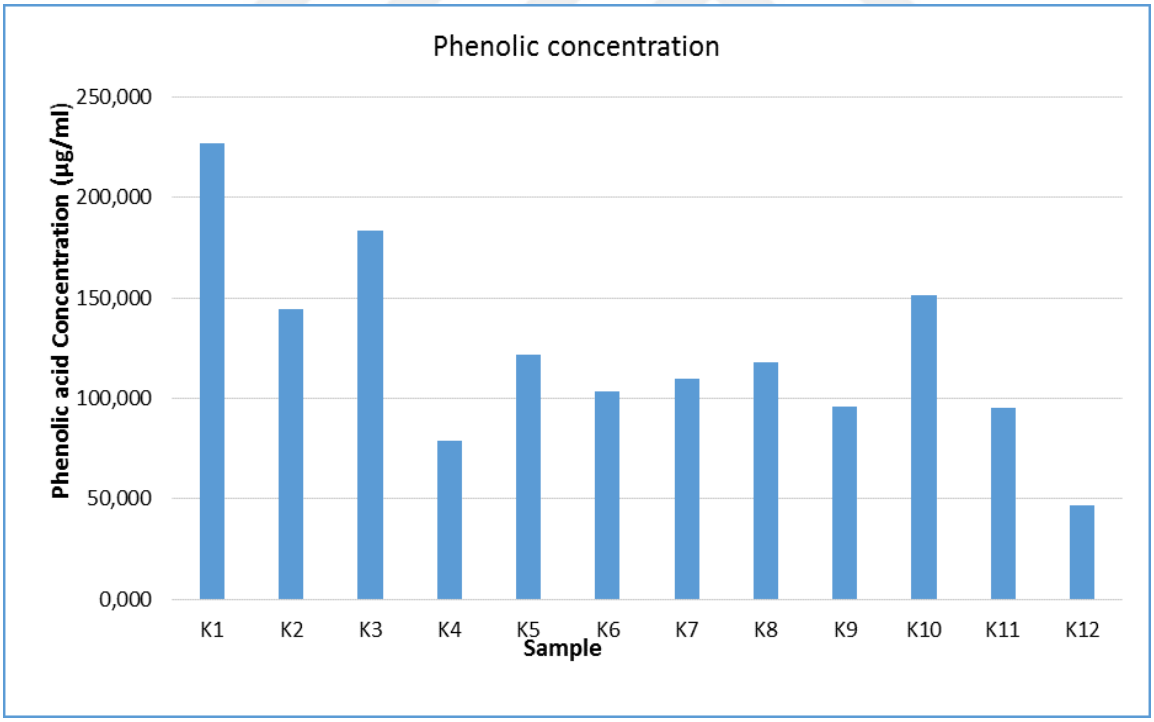

Figure 4.32. Phenolic concentration (K1(Qandil), K2(Asos), K3(Balambo), K4(Karokh), K5(Eruh), K6(Şirvan yolu), K7(Kezer Kampus), K8(Tillo) ,K9(Akdagmus), K10(Kurtalan (üç yol)) ,K11(Kurtalan zokayt) ,K12(Baykan))

4.2.2. Flavonoid analysis

Total flavonoid analysis was carried out according to Zhishen et al. 1999, Zou et al. It was made in 2004. According to the results of analysis, total flavonoid concentrate in 1 ml of extract in methanol extracts was given. Total flavonoid concentrations are given as routine equivalents. The calculation of routine equivalence is the shape that is prepared by us during our lab work. Is calculated according to the routine standard regression curve given. According to these results, the highest value was the K3 sample (2.59 mg / ml) and the lowest value was the K12 sample (0.60 mg / ml). Concentration results of the samples are plotted (figure 4.33).

Table 4.10. Show total flavonoid analysis

Sample	**Absorbance 1**	**Absorbance 2**	**Absorbance 3**	**Mean**	**SD**	**1mg/ml (1 ml)**
K1	1.047	1.063	1.019	1.04	0.022	2.26
K2	0.988	1.04	1.045	1.02	0.032	2.21
K3	1.181	1.196	1.191	1.19	0.008	2.59
K4	0.606	0.614	0.59	0.60	0.012	1.26
K5	0.792	0.93	0.845	0.86	0.070	1.83
K6	0.67	0.702	0.71	0.69	0.021	1.47
K7	0.79	0.789	0.806	0.80	0.010	1.69
K8	0.887	0.921	0.941	0.91	0.027	1.97
K9	0.788	0.804	0.802	0.80	0.009	1.70
K10	0.801	0.921	0.921	0.88	0.069	1.89
K11	0.607	0.508	0.613	0.58	0.059	1.20
K12	0.301	0.314	0.315	0.31	0.008	0.60

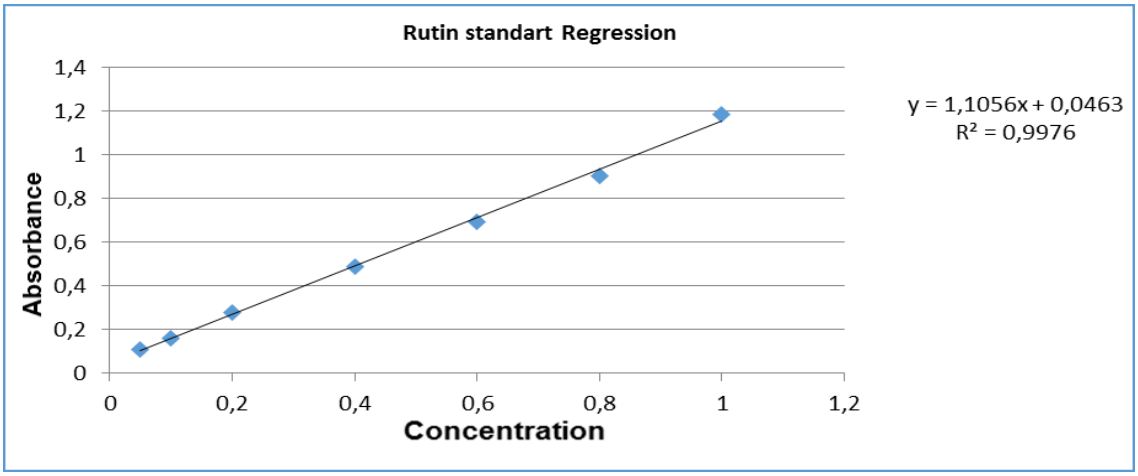

Figure 4.33. Rutin standart Regression

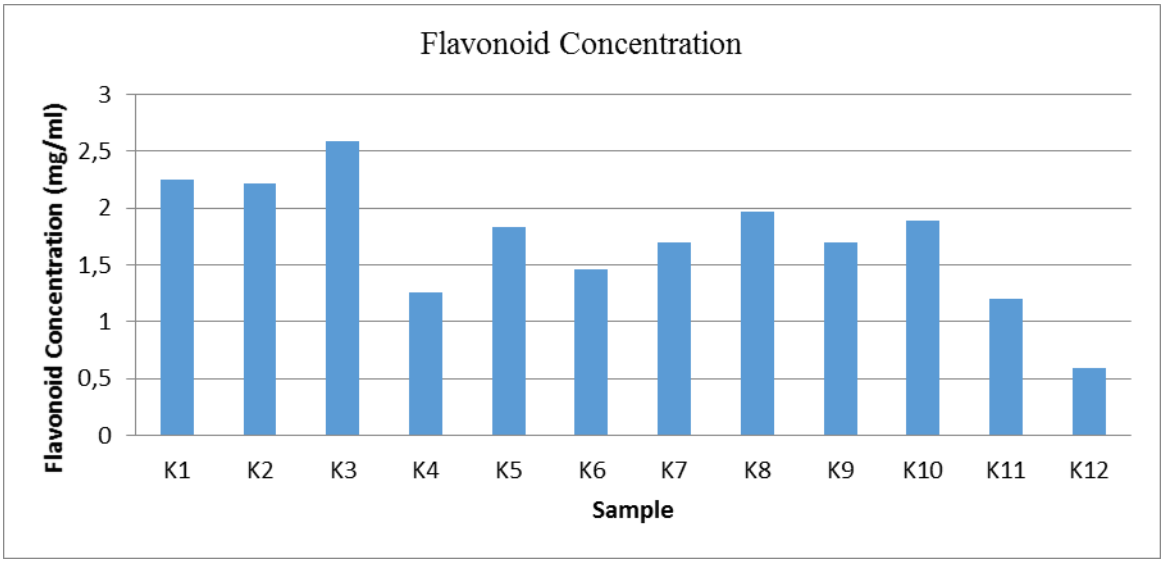

Figure 4.34. Flavonoid Concentration

4.2.3. DPPH analysis

DPPH analysis (Villano et al, 2007). According to the results of analysis, Ic50 values of DPPH inhibition of 1 ml extract in extracts prepared with methanol were given. According to these results, the highest inhibition was the K1 sample (0.16mg / ml) and the lowest inhibition was the K12 sample (1.32mg / ml). Concentration results of the samples are plotted (figure 4.35)

Table 4.11. DPPH analysis

Sample	mg/ml	,5mg/ml	),2mg/ml	mg/ml	),5mg/ml	),2mg/ml	c 50 (mgIml)
K1	0.094	0.098	0.229	84.56	83.91	62.40	0.16
K2	0.097	0.17	0.416	84.07	72.09	31.69	0.36
K3	0.095	0.104	0.417	84.40	82.92	31.53	0.29
K4	0.17	0.327	0.472	72.09	46.31	22.50	0.61
K5	0.104	0.225	0.448	82.92	63.05	26.44	0.45
K6	0.115	0.312	0.482	81.12	48.77	20.85	0.56
K7	0.104	0.252	0.458	82.92	58.62	24.79	0.48
K8	0.101	0.154	0.391	83.42	74.71	35.80	0.3
K9	0.114	0.27	0.462	81.28	55.67	24.14	0.51
K10	0.1	0.207	0.396	83.58	66.01	34.98	0.36
K11	0.173	0.328	0.481	71.59	46.14	21.02	0.62
K12	0.375	0.477	0.546	38.42	21.67	10.34	1.32

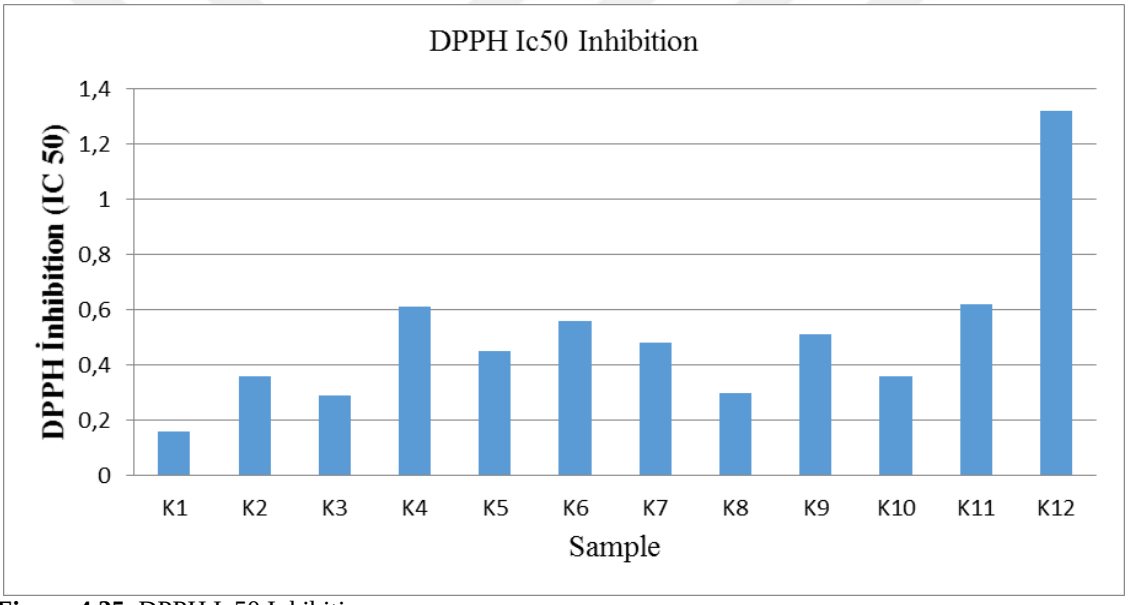

Figure 4.35. DPPH Ic50 Inhibition

K1(Qandil), K2(Asos), K3(Balambo), K4(Karokh), K5(Eruh), K6(Şirvan yolu), K7(Kezer Kampus), K8(Tillo) ,K9(Akdagmus) ,K10(Kurtalan (üç yol)) ,K11(Kurtalan zokayt) ,K12(Baykan))

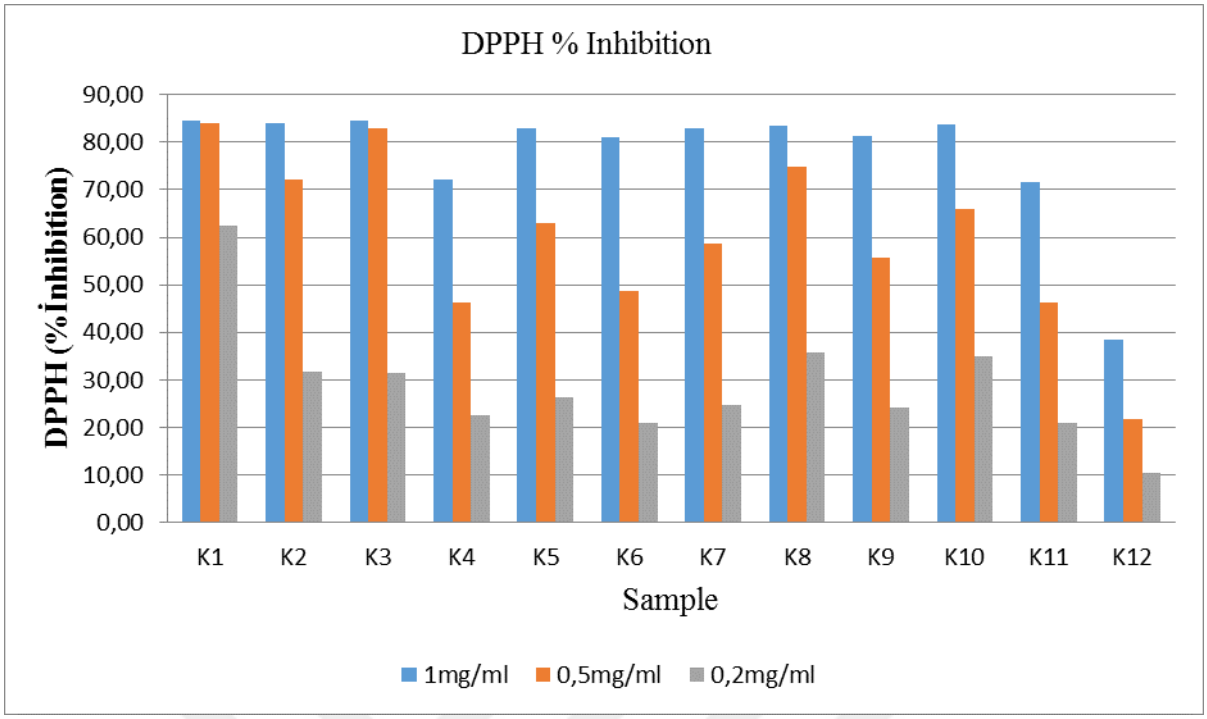

Figure 4.36. DPPH % Inhibition

K1(Qandil), K2(Asos), K3(Balambo), K4(Karokh), K5(Eruh), K6(Şirvan yolu), K7(Kezer Kampus), K8(Tillo) ,K9(Akdagmus) ,K10(Kurtalan (üç yol)) ,K11(Kurtalan zokayt) ,K12(Baykan))

4.2.4. FRAP analysis

FRAP analysis was performed according to (Benzie and Strain, 1996). According to the results of analysis, antioxidant capacities (total antioxidant amount) were given according to FRAP method in 1 ml extract in extracts prepared with methanol. FRAP antioxidant capacities are given as $FeSO_4$ equivalents. Calculation of $FeSO_4$ equilibrium is done by our laboratory during our work. Calculated based on the FRAP standard regression curve given. These results show that the highest antioxidant effect is the K5 sample (2.05 mg / ml) and the lowest antioxidant effect K12 sample (1.07 mg / ml). The antioxidant capacity results of the samples are plotted (figure .4.37)

Table 4.12. FRAP analysis

Sample	Absorbance 1	Absorbance2	Absorbance3	Mean	SD	1 mg/ml
K1	2.387	2.404	2.372	2.388	0.016	2.01
K2	2.314	2.372	2.356	2.347	0.030	1.97
K3	2.387	2.404	2.287	2.359	0.063	1.98
K4	1.944	1.841	1.956	1.914	0.063	1.59
K5	2.815	2.228	2.251	2.431	0.332	2.05
K6	1.9	1.932	1.87	1.901	0.031	1.58
K7	2.103	2.11	2.12	2.111	0.009	1.77
K8	2.3	2.318	2.314	2.311	0.009	1.94
K9	2.138	2.166	2.156	2.153	0.014	1.80
K10	1.956	2.238	2.275	2.156	0.174	1.81
K11	2.12	2.196	2.175	2.164	0.039	1.81
K12	1.336	1.328	1.295	1.320	0.022	1.07

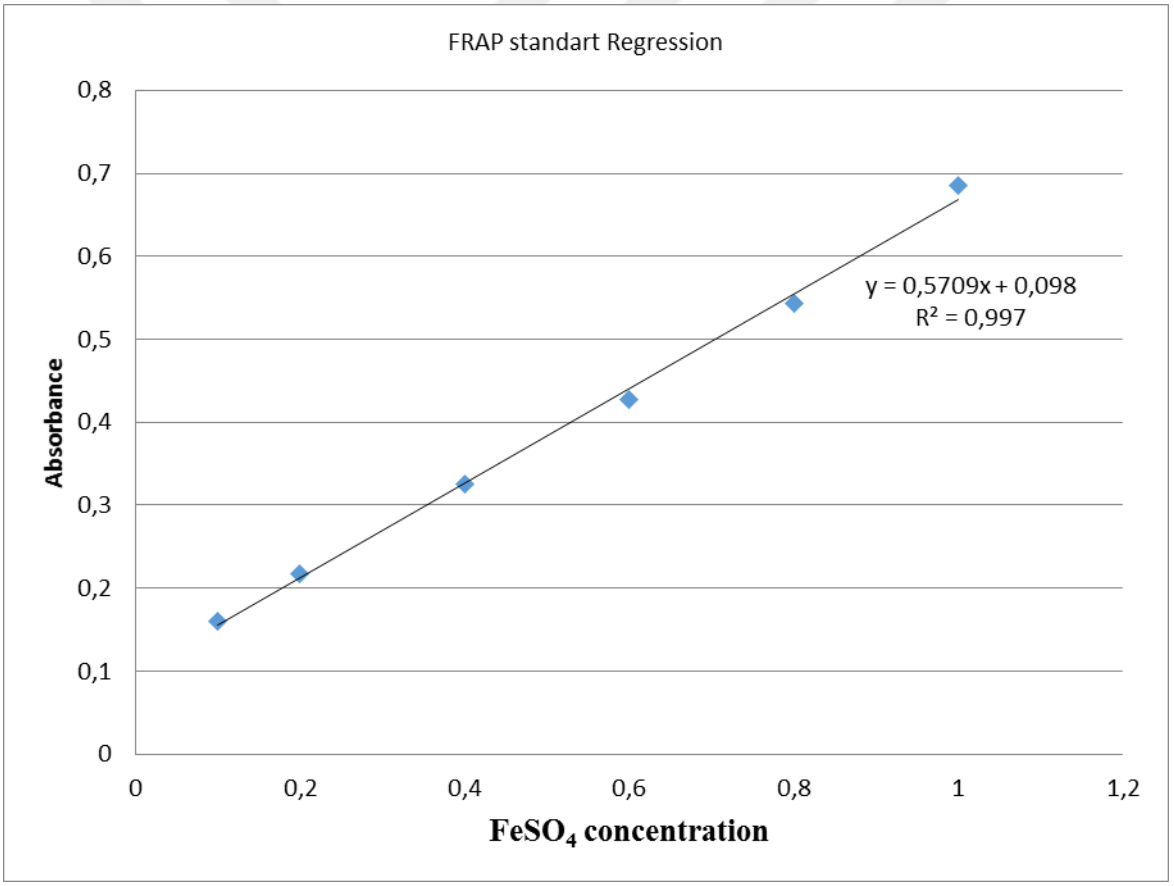

Figure 4.37. FRAP standart Regression

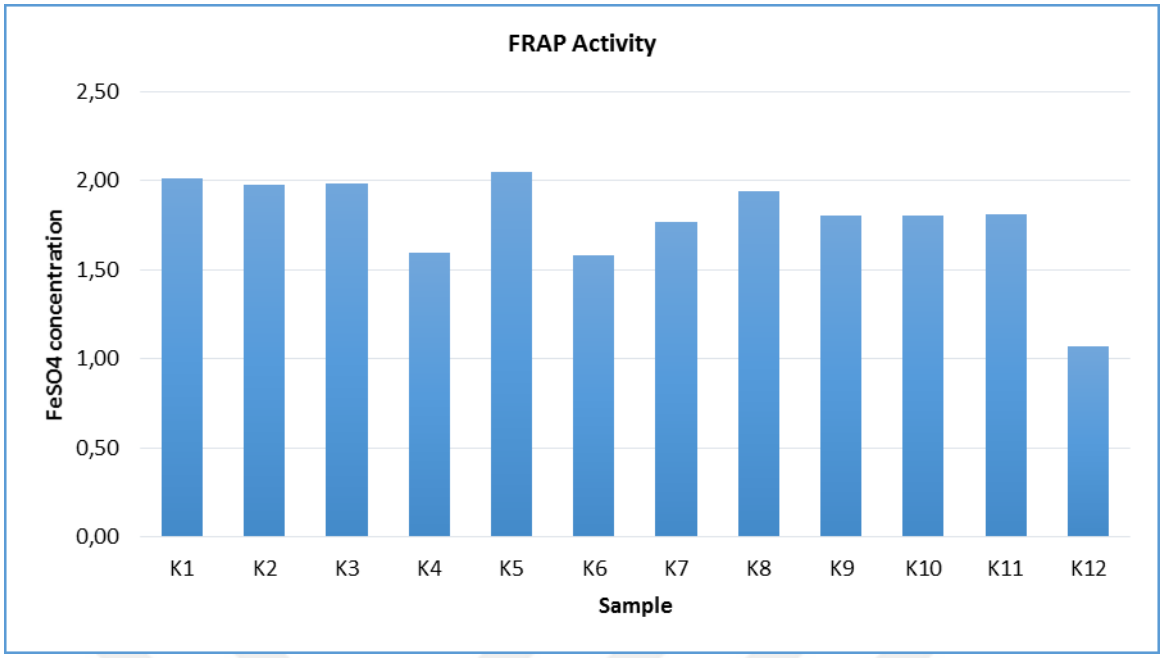

Figure 4.38. FRAP Activity

4.2.5. Iron Chelating analysis

The iron chelating analysis was carried out according to (Rival et al, 2001). According to the results of analysis, 1 ml of extract was prepared by adding iron chelating agent to the extracts prepared with methanol. According to these results, the highest iron chelating capacity showed a K11 sample (22.920mg / ml) and the lowest iron chelating capacity K9 sample (9.174mg / ml). The iron chelating capacities of the samples are given in graphical form (figure.4.39)

Table 4.13. Iron chelating analysis

Sample	Absorbance 1	Absorbance2	Absorbance 3	Average	1 mg/ml % inhibition
K1	0.415	0.387	0.402	0.401	14.484
K2	0.469	0.467	0.395	0.444	10.737
K3	0.37	0.32	0.34	0.343	19.617
K4	0.346	0.36	0.401	0.369	17.345
K5	0.338	0.292	0.375	0.335	20.354
K6	0.386	0.354	0.374	0.371	17.139
K7	0.445	0.41	0.391	0.415	13.245
K8	0.479	0.444	0.443	0.455	9.705
K9	0.501	0.431	0.452	0.461	9.174
K10	0.414	0.312	0.316	0.347	19.263
K11	0.315	0.303	0.3	0.306	22.920
K12	0.345	0.338	0.362	0.348	19.174

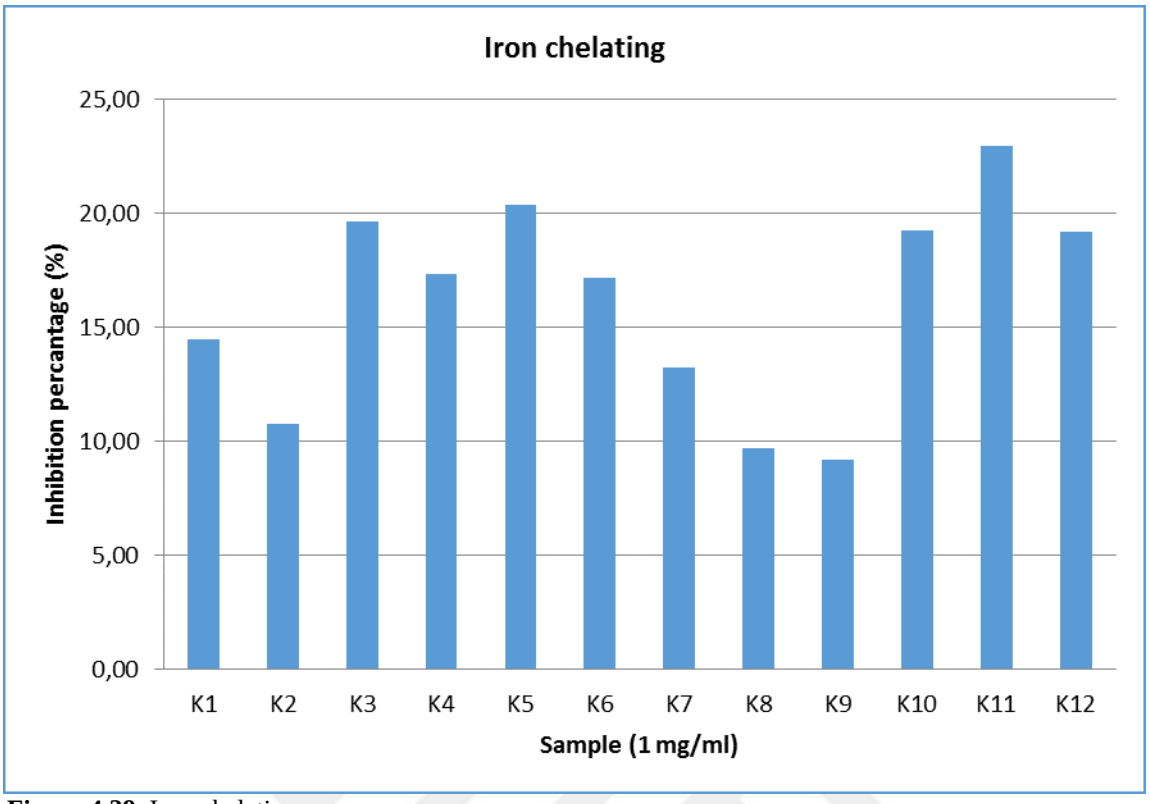

Figure 4.39. Iron chelating

4.3. Phenolic compounds

4.3.1. The results of phenolic compounds:

In this section of the research the concentration of phenolic compounds were determined in accordance to gallic acid standard and the reactivity to Folin-Ciocalteu (Slinkard and Singleton, 1977). Additionally, the total phenolic concentration in 1 ml extract in the extracts prepared with methanol was observed. Furthermore more 30 phenolic acids were identified, (Küçükbay, F. Zehra et al, 2014).

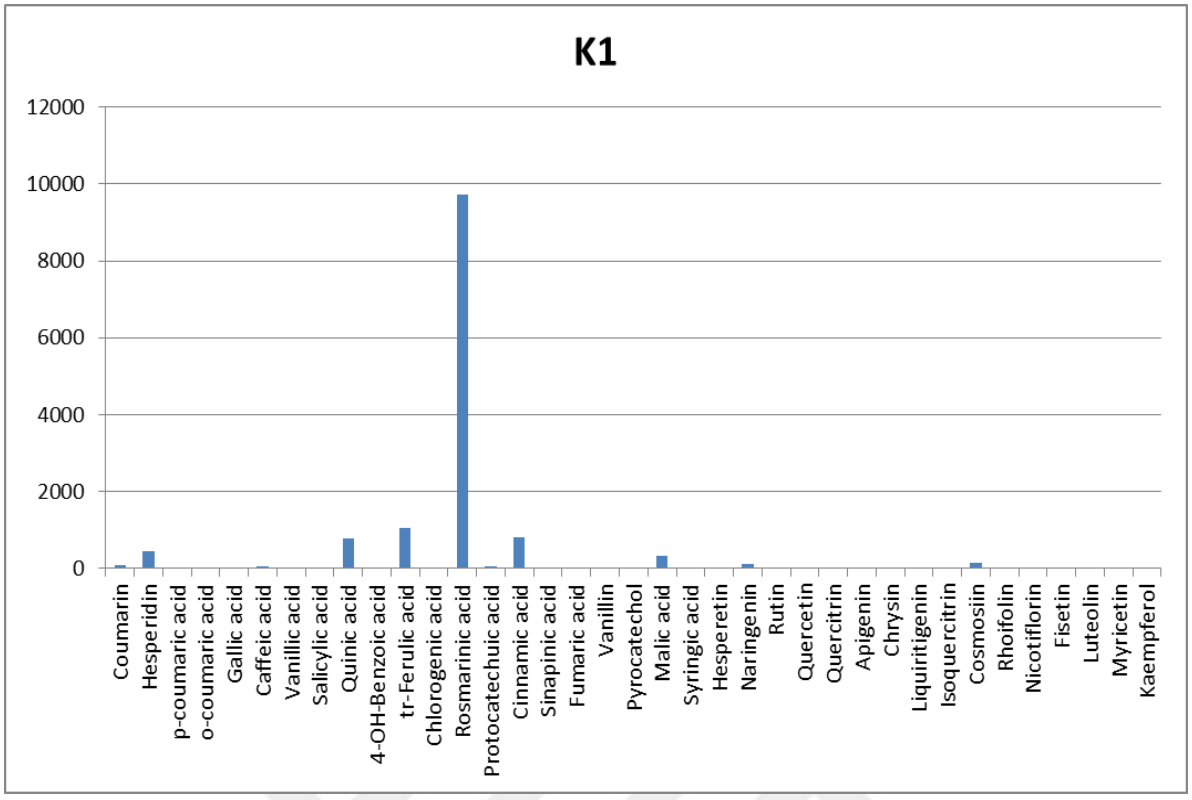

Figure 4.40. The concentration of phenolic compound in sample K1

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Figure 4.40 describes the connection of phenolic compounds in the investigated samples K1 which collected in Qandeel. From this figure, it is uncovered that the concentration of Rosmarinic acid was the highest (9721.56) while the concentration of Vanillin was the lowest (1.03), even though some compounds such as Nicotiflorin, Fisetin, Luteolin, Myricetin, and Kaempferol were not detected. Similar finding found by Ozen, Tevfik, Ibrahim Demirtas, and Huseyin Aksit (2011).

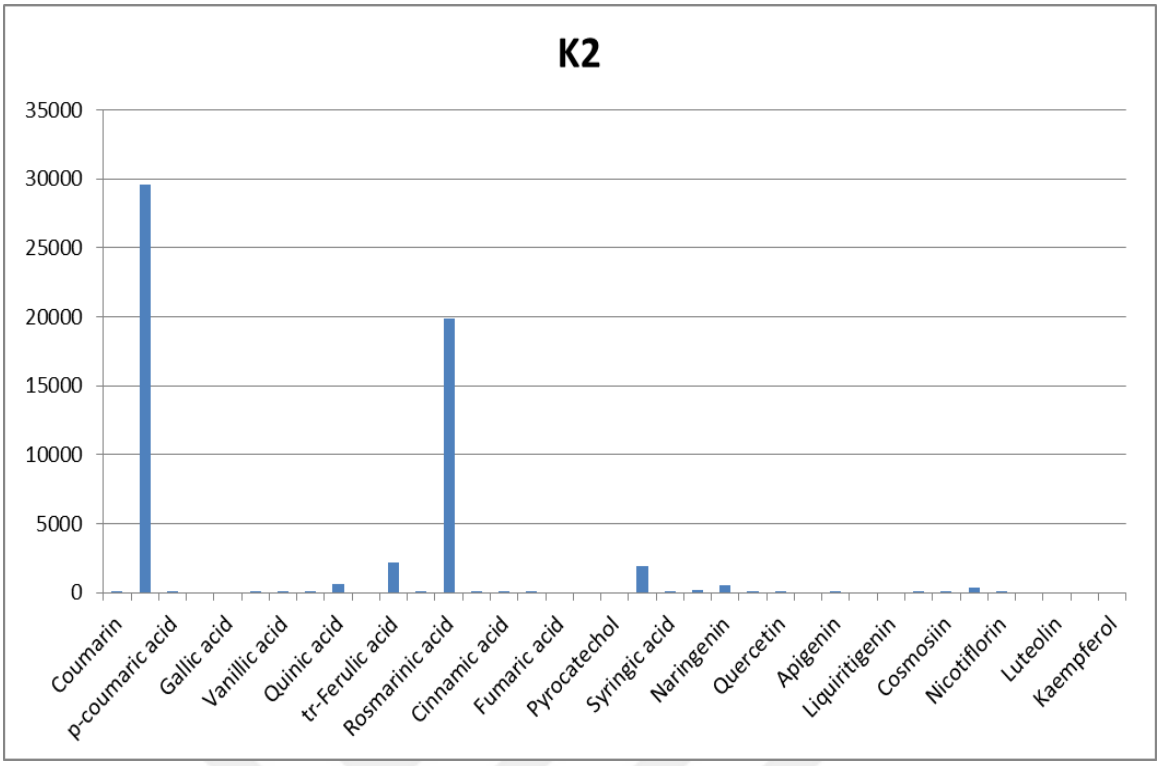


Figure 4.41. The concentration of phenolic compound in sample K2

The connection of phenolic compounds in the investigated samples K2 which collected in Asos is described Figure 4.41. In this figure, it is revealed that although some compounds like Nicotiflorin, Fisetin, Luteolin, Myricetin, and Kaempferol were not detected, the level of Hesperidin was the highest (29605.78). However, the concentration of Sinapinic acid was the lowest (1.89).

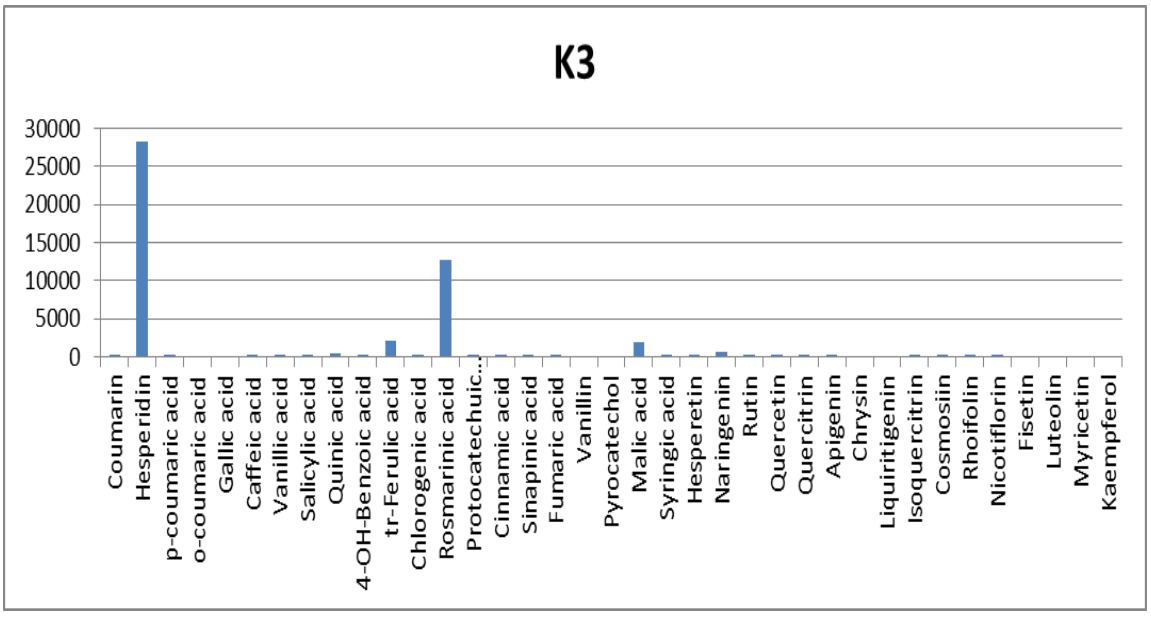


Figure 4.42. The concentration of phenolic compound in sample K3.

Figure 4.42 displays the amount of phenolic acid in the examined samples K3 that gathered in Balambo. It is revealed that several compounds like Nicotiflorin, Fisetin, Luteolin, Myricetin, and Kaempferol were not identified. As the level of Fumaric acid was the minimum (3.48) the concentratio of Hespardin was the maximum (28247.46).

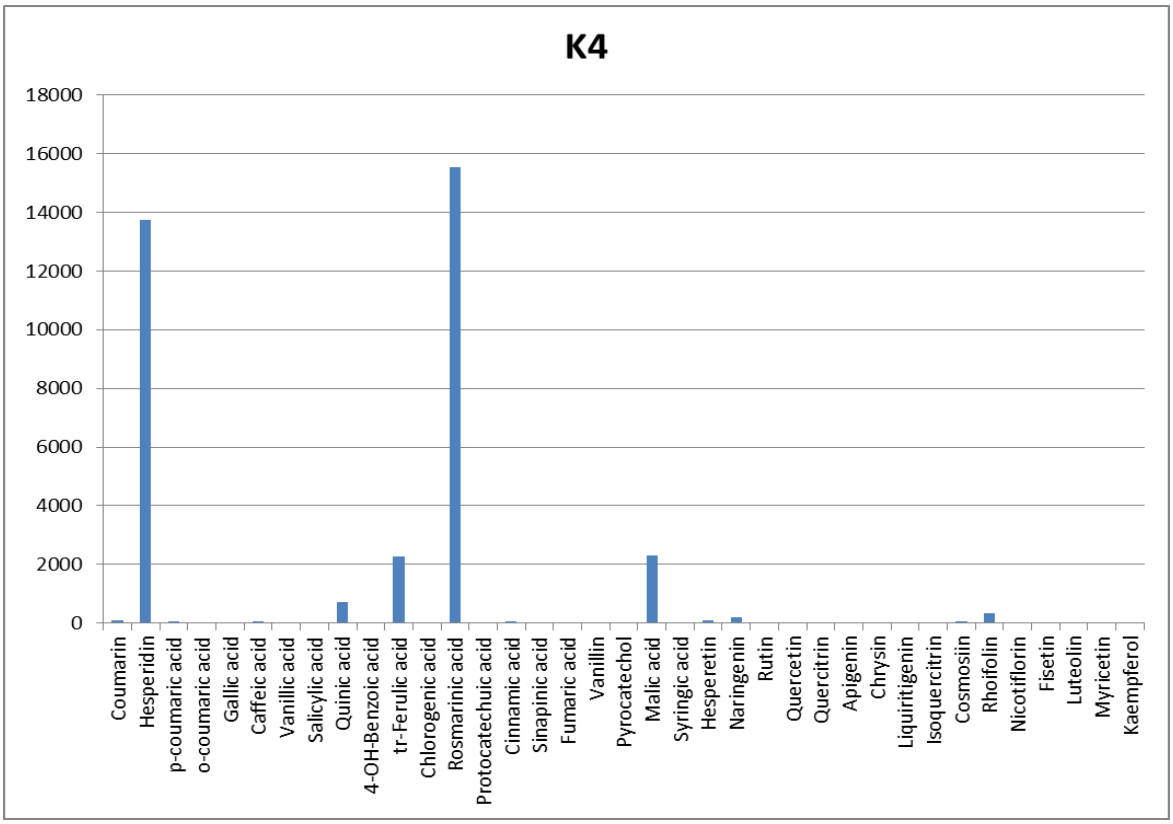


Figure 4.43. The concentration of phenolic compound in sample K4.

The amount of phenolic acid in the examined samples K4 that gathered in Karokh is indicated in Figure (4.43). It is revealed that several compounds like Nicotiflorin, Fisetin, Luteolin, Myricetin, and Kaempferol were not identified. As the level of Syringic acid was the minimum (3.67) the concentration of Rosmaininc acid was the maximum (15542.62).

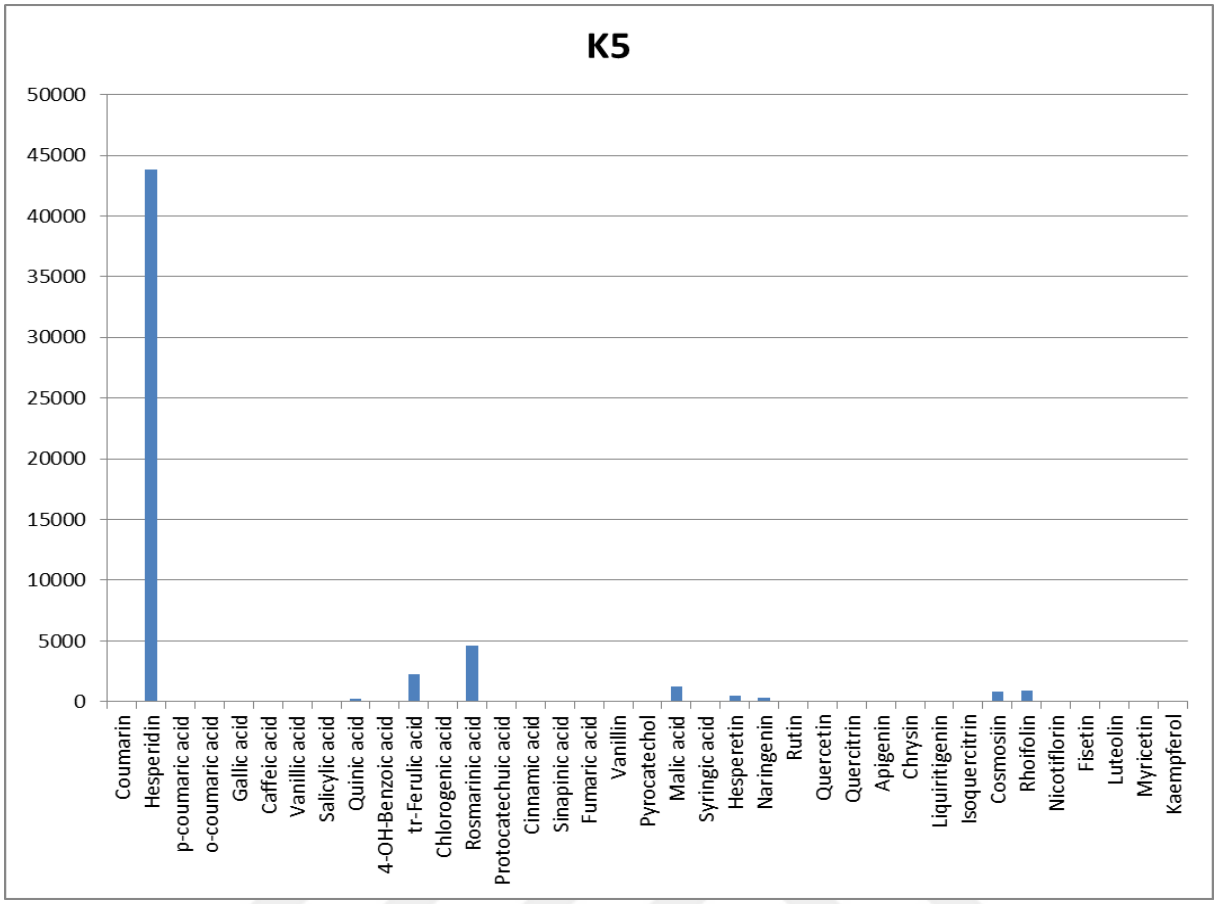


Figure 4.44. The concentration of phenolic compound in sample K5

Figure 4.44 describes the level of phenolic acid in the examined samples K5 that collected in Eruh. It is uncovered that several compounds like Nicotiflorin, Fisetin, Luteolin, Myricetin, and Kaempferol were not detected. While the level of Vanillin was the lowest (1.45) the level of Hesperidin acid was the maximum (43861.65). (Ozen, et al, 2011) also found similar results.

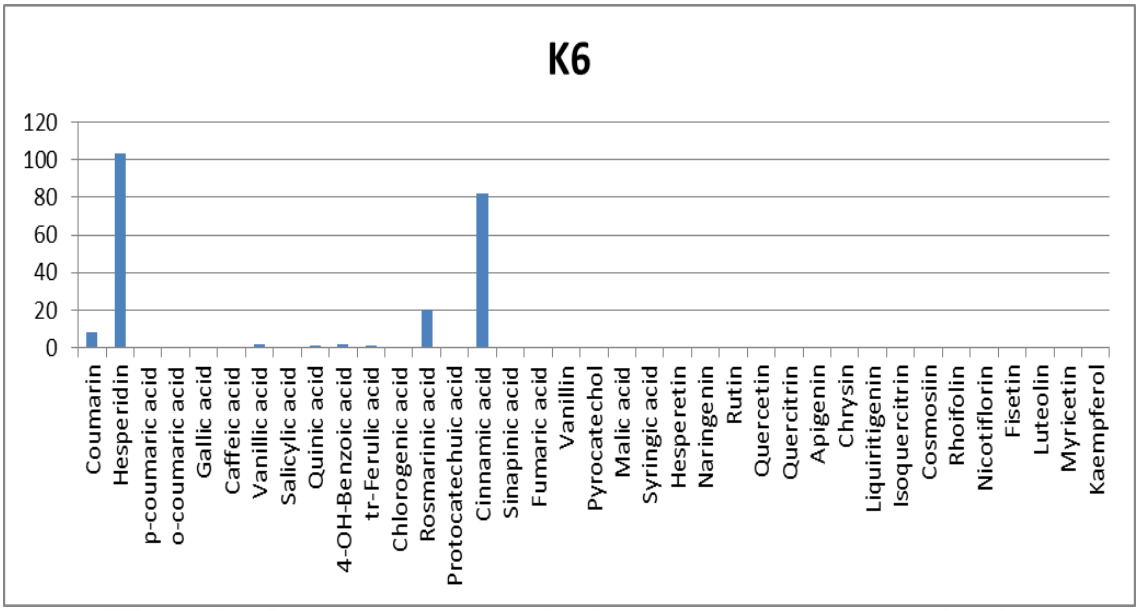


Figure 4.45. The phenolic compound in K6

The concentration of phenolic acid in the investigated samples K6 that collected in Sirvan Yolu is demonstrated in figure 4.45. It is uncovered that several compounds like Fumaric acid, Vanillin,Pyrocatechol, Malic acid, Syringic acid, Hesperetin, Naringenin, Rutin, Quercetin, Quercitrin, Apigenin, Chrysin, Liquiritigenin, Isoquercitrin, Cosmosiin, Rhoifolin, Nicotiflorin, Fisetin, Luteolin, Myricetin, Kaempferolwere not detected. While the level of Caffeic acid was the lowest (0.08) the level of Hesperidin acid was the maximum (103.49). (Küçükbay, F. Zehra, et al, 2014) have investigated three thymus plant and found around defferent compounds.

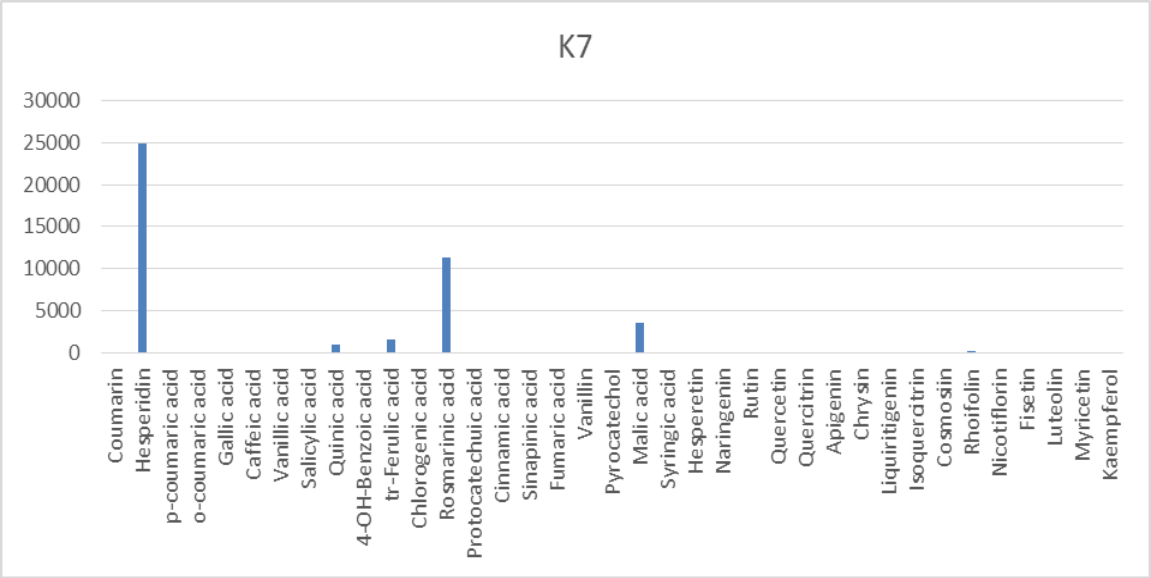


Figure 4.46. The phenolic compound in K7

Figure 4.46 presents the concentration of phenolic acid in the investigated samples K7 that collected in Kezer Kampus. It is revealed that numerous compounds such as Fumaric acid, Vanillin, Pyrocatechol, Nicotiflorin, Fisetin, Luteolin, Myricetin, and Kaempferol were not detected. While the level of Quercetin was the lowest (0.23) the level of Hesperidin acid was the highest (24896.47). (Roby, M et al, 2013) discovered similar outcomes.

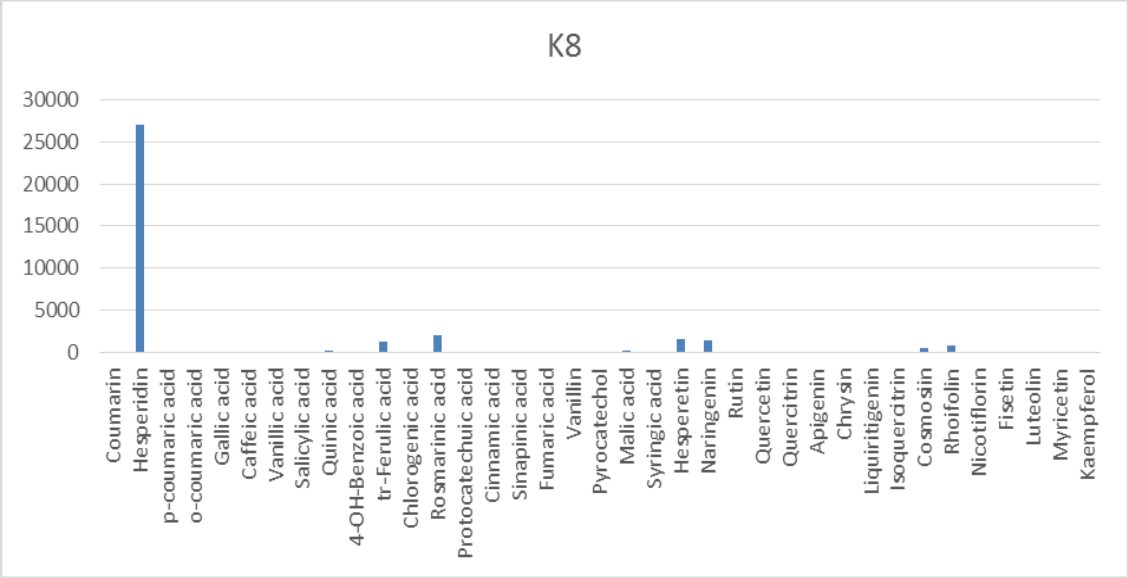


Figure 4.47. The phenolic compound in K8

The concentration of phenolic acid in the investigated samples K8 that collected in Tillo is presented in figure 4.47. It is uncovered that several compounds like o-coumaric acid, Gallic acid, Nicotiflorin, Fumaric acid, Vanillin, Pyrocatechol, Fisetin, Luteolin, Myricetin, and Kaempferol were not detected. While the concentration of Hesperidin acid was the largest (26971.65), the level of Isoquercitrin was the lowest (2.45).

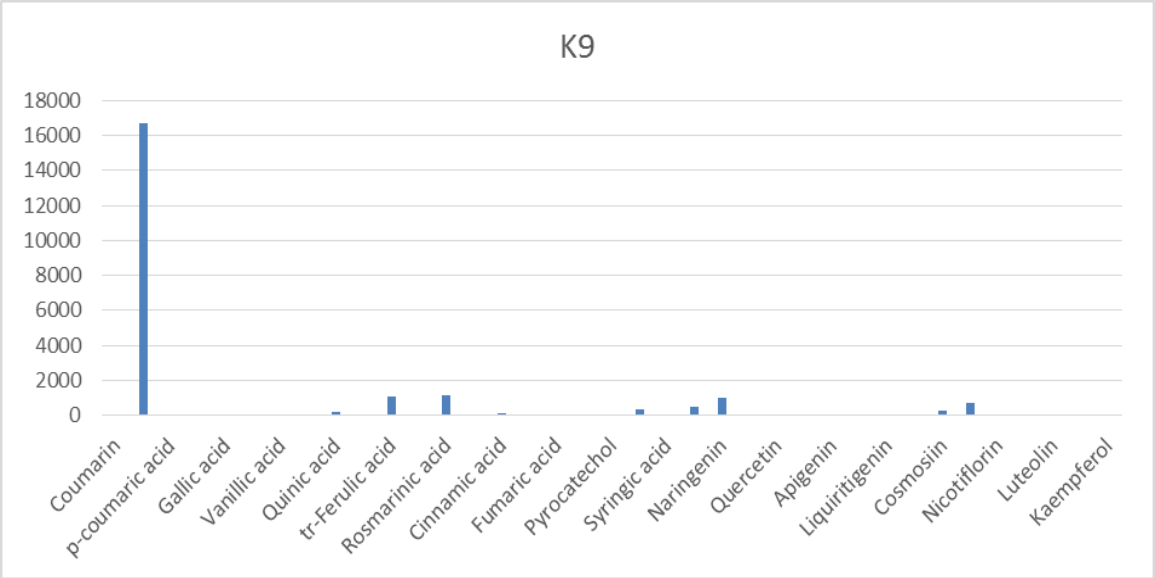


Figure 4.48. The phenolic compound in K9

Figure 4.48 displays the concentration of phenolic acid in the investigated samples K9 that collected in Akdoğmuş Koyu. It is uncovered that several compounds such as o-coumaric acid, Gallic acid, Nicotiflorin, Fisetin, Fumaric acid, Vanillin, Pyrocatechol, Luteolin, Myricetin, and Kaempferol were not identified. Eventhough the concentration of Chlorogenic acid was the minimum (0.04) the concentration of Hesperidin acid was the biggest (16668.15).

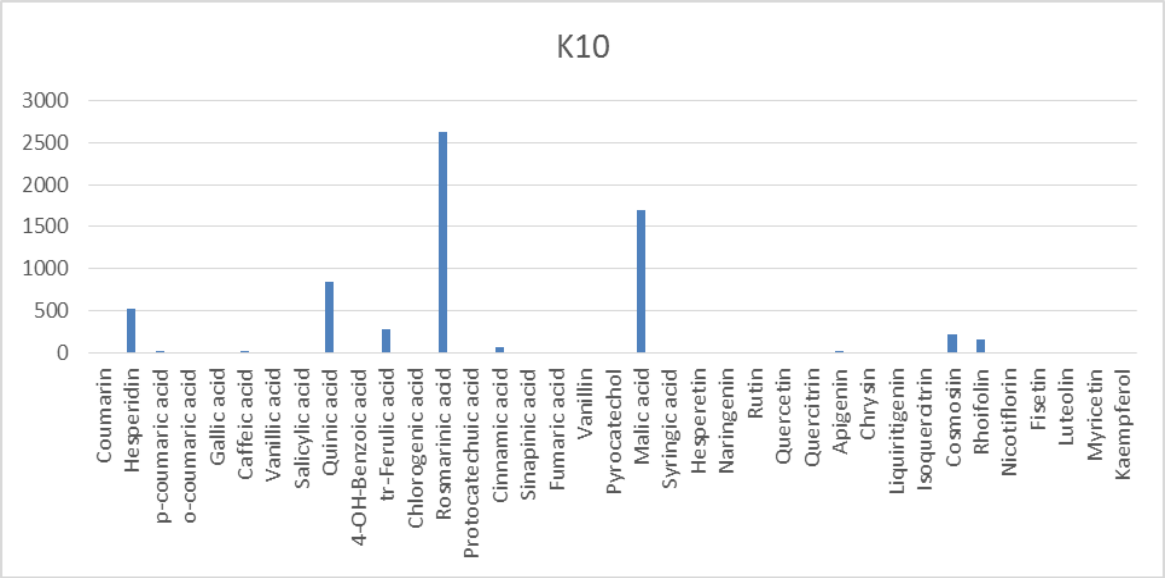


Figure 4.49. The phenolic compound in K10

In this figure, the content of phenolic acid in the examined samples K10 that gathered in Kurtalan Üçyolu is shown. It is revealed that some compounds such o-coumaric acid, 4-OH-Benzoic acid, Fumaric acid, Vanillin, Pyrocatechol, Naringenin, Rutin, Quercetin, Quercitrin, Fisetin, Chrysin, Liquiritigenin, Luteolin, Myricetin and Kaempferol were not identified. Moreover, the amount of Nicotiflorin content was the lowest (0.38) while the level of Rosmarinic acid was the highest (2621.07).

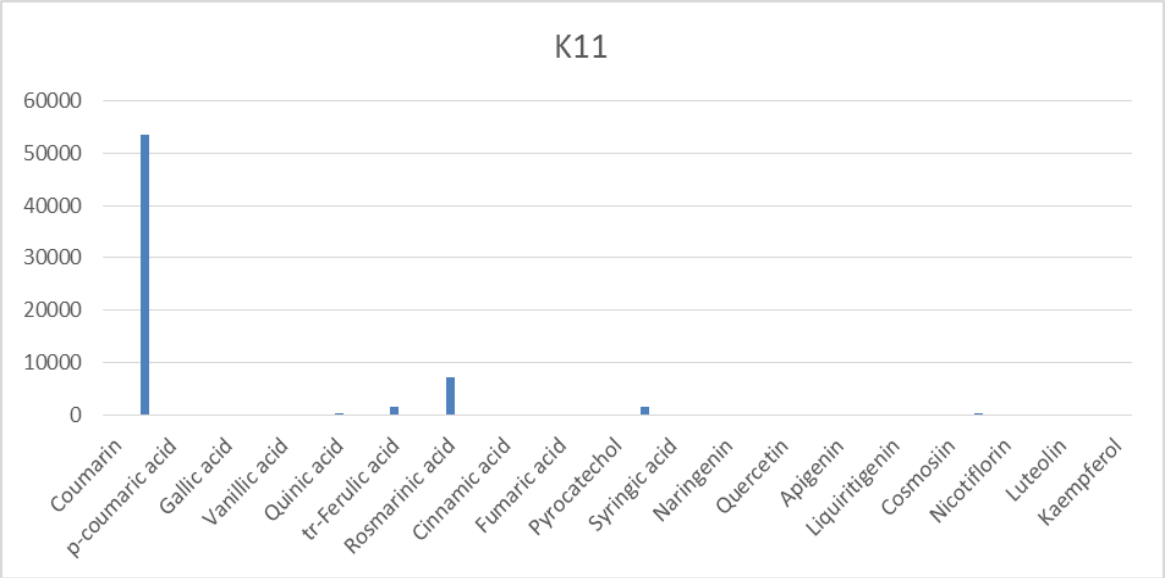


Figure 4.50. The phenolic compound in K11

Figure 4.50 displays the content of phenolic acid in the investigated samples K11 that taken from in Kurtalan (Zokayt-Şelale) region. It is discovered that some compounds such as o-coumaric acid, Gallic acid, Vanillin, Pyrocatechol, Chrysin, Liquiritigenin, Nicotiflorin, Fisetin, Luteolin, Myricetin, and Kaempferol were not detected. While the level of 4-OH-Benzoic acidwas the lowest (1.48) the level of Hesperidin acid was the maximum (53442.23).

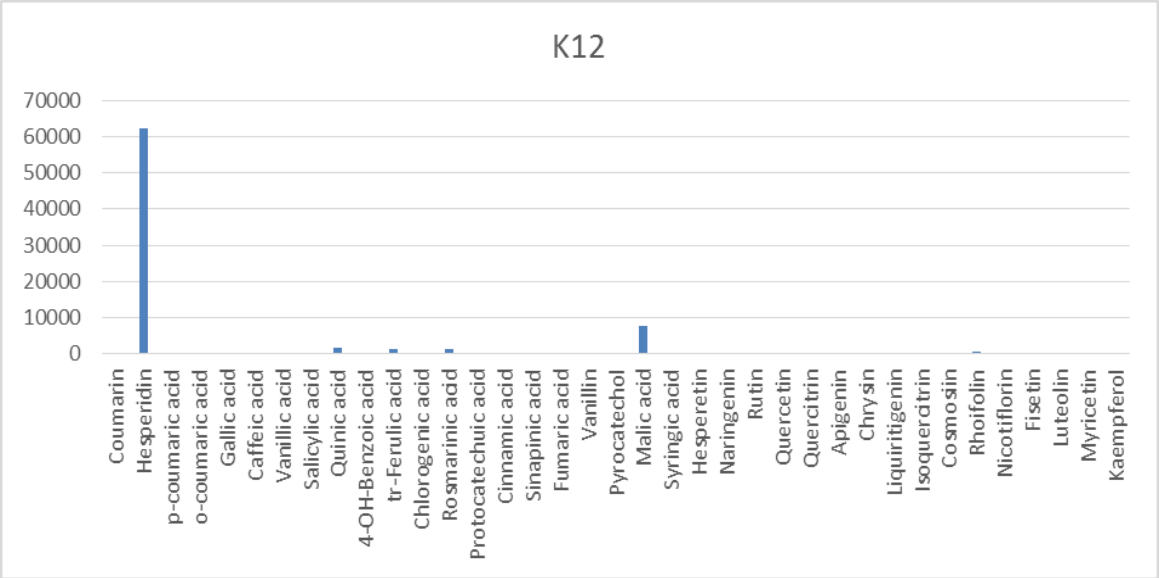


Figure 4.51. The phenolic compound in K12

In this figure, the level of phenolic acid in the studied samples K12 that gathered in Baykan (Girişi) is demonstrated. It is found that several compounds such as o-coumaric acid, Gallic acid, 4-OH-Benzoic acid, Sinapinic acid, Quercetin, Quercitrin, Nicotiflorin, Fisetin, Luteolin, Myricetin, and Kaempferol were not detected. While the level of Isoquercitrin was the lowest (1.48) the level of Hesperidin acid was the maximum (62368.19). Similar results was reprted by Roby M et al, (2013).

Table 4.14. Phenolic compounds mean

Analyte	Qandil	Asos	Balambo	Karokh	Eruh Girişi	Şirvan yolu	Kezer Kampus	Tillo	Akdoğmuş Koyu	Kurtalan Üçyolu	Kurtalan Zokayt	Baykan
Coumarin	81.740	66.320	75.800	81.210	59.010	8.170	78.170	58.080	39.490	15.520	31.430	11.040
Hesperidin	444.140	29605.780	28247.460	13739.340	43861.650	103.490	24896.470	26971.650	16668.150	530.430	53442.230	62368.190
p-coumaric acid	31.070	43.370	56.370	74.030	97.770	0.180	38.580	130.450	38.280	19.890	35.010	24.070
o-coumaric acid	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D
Gallic acid	1.060	N.D	N.D	20.460	N.D	N.D	N.D	N.D	N.D	5.900	N.D	N.D
Caffeic acid	58.970	116.230	160.420	61.470	83.790	0.080	53.600	42.440	25.670	30.530	84.830	38.140
Vanillic acid	17.950	17.970	17.740	12.090	12.050	2.110	21.850	15.700	11.790	12.850	42.200	7.300
Salicylic acid	1.850	16.970	16.950	24.170	3.490	0.070	13.580	3.330	5.600	3.560	14.760	30.260
Quinic acid	772.380	653.900	540.400	702.520	255.880	1.020	979.570	283.780	225.140	838.460	361.370	1534.690
4-OH-Benzoic acid	2.020	N.D	1.100	N.D	N.D	1.890	N.D	N.D	N.D	N.D	1.480	N.D
tr-Ferulic acid	1045.750	2228.260	2022.420	2269.440	2224.040	1.240	1572.470	1301.530	1096.100	275.070	1736.360	1130.740
Chlorogenic acid	26.360	11.230	14.440	11.150	9.510	N.D	12.260	2.870	0.040	12.160	9.940	4.750
Rosmarinic acid	9721.560	19894.510	12661.050	15542.620	4608.950	20.500	11355.940	2055.790	1163.440	2621.070	7242.920	1337.360
Protocatechuic acid	45.740	22.760	24.090	29.280	22.040	N.D	21.450	7.860	10.550	13.240	27.200	29.500
Cinnamic acid	813.780	111.790	85.020	72.390	55.180	81.980	74.960	84.090	99.770	71.650	84.990	53.470
Sinapinic acid	1.740	1.890	8.810	N.D	3.140	N.D	2.040	2.760	3.380	2.210	31.820	N.D
Fumaric acid	4.380	N.D	3.480	4.030	2.600	N.D	N.D	N.D	N.D	N.D	2.740	8.050
Vanillin	1.030	N.D	N.D	N.D	1.450	N.D	N.D	N.D	N.D	N.D	N.D	N.D
Pyrocatechol	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D
Malic acid	314.870	1904.710	1855.890	2299.180	1233.900	N.D	3649.250	301.040	329.310	1697.930	1593.950	7625.280
Syringic acid	1.620	2.500	2.260	3.670	1.490	N.D	2.040	2.700	1.960	5.940	4.030	2.970
Hesperetin	2.110	210.210	55.500	83.060	523.730	N.D	38.940	1669.850	509.320	2.780	198.310	166.960
Naringenin	122.080	509.580	553.160	188.240	337.270	N.D	86.370	1395.350	967.110	N.D	122.460	28.070
Rutin	17.860	6.000	5.480	18.280	8.010	N.D	5.590	N.D	N.D	N.D	6.020	4.330
Quercetin	N.D	13.910	12.090	19.800	N.D	N.D	0.230	12.560	28.460	N.D	N.D	N.D
Quercitrin	N.D	N.D	2.260	N.D	3.460	N.D	N.D	4.750	N.D	N.D	6.580	N.D
Apigenin	13.780	18.190	14.950	13.960	8.080	N.D	4.860	26.890	8.360	19.420	5.950	3.670
Chrysin	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D
Liquiritigenin	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D
Isoquercitrin	9.700	15.910	13.660	7.960	2.280	N.D	7.130	2.450	N.D	13.900	3.040	1.480
Cosmosiin	137.160	65.120	31.270	63.320	791.680	0.360	67.810	536.170	277.760	215.070	86.050	185.500
Rhoifolin	1.470	382.150	252.750	337.070	883.860	N.D	303.430	915.940	671.000	160.140	483.300	616.810
Nicotiflorin	N.D	6.280	10.730	5.570	67.090	N.D	7.390	39.470	31.660	0.380	N.D	N.D
Fisetin	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D
Luteolin	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D
Myricetin	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D
Kaempferol	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D	N.D

Table 4.15. Phenolic compounds Standart deviation (±)

Analyte	Qandil	Asos	Balambo	Karokh	Eruh Girişi	Şirvan yolu	Kezer Kampus	Tillo	Akdoğmuş Koyu	Kurtalan Üçyolu	Kurtalan Zokayt	Baykan
Coumarin	0.019	0.016	0.018	0.019	0.014	0.002	0.019	0.014	0.009	0.004	0.007	0.003
Hesperidin	0.116	7.757	7.401	3.600	11.492	0.027	6.523	7.067	4.367	0.139	14.002	16.340
p-coumaric acid	0.016	0.022	0.029	0.038	0.050	0.000	0.020	0.067	0.020	0.010	0.018	0.012
o-coumaric acid	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Gallic acid	0.000	N.D.	N.D.	0.006	N.D.	N.D.	N.D.	N.D.	N.D.	0.002	N.D.	N.D.
Caffeic acid	0.030	0.000	0.019	0.015	0.009	0.011	0.030	0.014	0.000	0.000	0.000	0.000
Vanillic acid	0.009	0.009	0.009	0.006	0.006	0.001	0.011	0.008	0.006	0.007	0.021	0.004
Salicylic acid	0.001	0.006	0.006	0.008	0.001	0.000	0.004	0.001	0.002	0.001	0.005	0.010
Quinic acid	0.063	0.054	0.044	0.058	0.021	0.000	0.080	0.023	0.018	0.069	0.030	0.126
4-OH-Benzoic acid	0.001	N.D.	0.000	N.D.	N.D.	0.001	N.D.	N.D.	N.D.	N.D.	0.000	N.D.
tr-Ferulic acid	0.517	1.101	0.999	1.121	1.099	0.001	0.777	0.643	0.541	0.136	0.858	0.559
Chlorogenic acid	0.002	0.001	0.001	0.001	0.001	N.D.	0.001	0.000	0.000	0.001	0.001	0.000
Rosmarinic acid	6.931	14.185	9.027	11.082	3.286	0.015	8.097	1.466	0.830	1.869	5.164	0.954
Protocatechuic acid	0.019	0.009	0.010	0.012	0.009	N.D.	0.009	0.003	0.004	0.005	0.011	0.012
Cinnamic acid	0.116	0.016	0.012	0.010	0.008	0.012	0.011	0.012	0.014	0.010	0.012	0.008
Sinapinic acid	0.000	0.001	0.002	N.D.	0.001	N.D.	0.001	0.001	0.001	0.001	0.009	N.D.
Fumaric acid	0.001	N.D.	0.000	0.000	0.000	N.D.	N.D.	N.D.	N.D.	N.D.	0.000	0.001
Vanillin	0.000	N.D.	N.D.	N.D.	0.000	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Pyrocatechol	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Malic acid	0.036	0.215	0.210	0.260	0.139	N.D.	0.412	0.034	0.037	0.192	0.180	0.862
Syringic acid	0.004	0.006	0.005	0.009	0.004	N.D.	0.005	0.006	0.005	0.014	0.010	0.007
Hesperetin	0.001	0.118	0.031	0.047	0.294	N.D.	0.022	0.938	0.286	0.002	0.111	0.094
Naringenin	0.064	0.265	0.288	0.098	0.176	N.D.	0.045	0.727	0.504	N.D.	0.064	0.015
Rutin	0.003	0.001	0.001	0.003	0.001	N.D.	0.001	N.D.	N.D.	N.D.	0.001	0.001
Quercetin	N.D.	0.006	0.005	0.009	N.D.	N.D.	0.000	0.006	0.013	N.D.	N.D.	N.D.
Quercitrin	N.D.	N.D.	0.045	N.D.	0.069	N.D.	N.D.	0.095	N.D.	N.D.	0.132	N.D.
Apigenin	0.009	0.012	0.010	0.009	0.005	N.D.	0.003	0.017	0.005	0.013	0.004	0.002
Chrysin	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Liquiritigenin	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Isoquercitrin	0.001	0.002	0.002	0.001	0.000	N.D.	0.001	0.000	N.D.	0.002	0.000	0.000
Cosmosiin	0.082	0.039	0.019	0.038	0.473	0.000	0.040	0.320	0.166	0.128	0.051	0.111
Rhoifolin	0.001	0.360	0.238	0.317	0.832	N.D.	0.286	0.862	0.631	0.151	0.455	0.580
Nicotiflorin	N.D.	0.002	0.003	0.002	0.019	N.D.	0.002	0.011	0.009	0.000	N.D.	N.D.
Fisetin	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Luteolin	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Myricetin	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.
Kaempferol	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.	N.D.

4.4. Correlation analysis of elements in thyme

Correlation analysis of element contents of thyme plant is very important to explain factors such as the some spices and herbs ((Kara, 2009; Karadaş and Kara, 2012). Correlation coefficients between the elements analyzed in thyme plants were calculated in the form of correlation matrix and given in Table 4.16. The correlations given in Table 4.16 indicate that the relationships between the elements are complex and are difficult to explain individually. The element to element correlation data in terms of linear correlation coefficient values that are significant at 95% and 99% confidence level was examined. The pairs of Fe–V (0.917), Zn–Ni (0.958), Ni–Cr (0.970), Se–V (0.902), Sr–Co (0.928), Sn–Cu (0.970), Sn–Zn (0.953), Sb–Ni (0.945), Sb–Cu (0.963), Sb–Zn (0.991), Sb–Sn (0.944), La–Se (0.924), Ce–Se (0.907), Ce–La (0.943), Pb–Ni (0.959), Pb–Cu (0.972), Pb–Zn (0.993), Sb–Sn (0.950) and Pb–Sb (0.983) showed very high and significant correlations at 99% confidence level. Besides, the pairs of K–B (0.720), Ca–Ti (0.862), Ni–Cr (0.807), Cu–Cr (0.711), Cu–Ni (0.880), Zn–Cr (0.805), As–Ni (0.819), As–Cu (0.817), As–Zn (0.812), Se–Fe (0.778), Sr–Cr (0.848), Sr–Mn (0.795), Sr–Zn (0.852), Cd–Mg (0.827), Cd–V (0.991), Sb–Sn (0.944), La–Se (0.924), Ce–Se (0.907), Ce–La (0.943), Pb–Ni (0.959), Pb–Cu (0.742), Cd–Se (0.714), Sn–Co (0.869), Sn–As (0.790), Sn–Sr (0.725), Sn–Cr (0.854), Sb–As (0.797), Sb–Sr (0.864), La–V (0.839), La–Fe (0.746), La–Cd (0.844), Ce–Ti (0.858), Ce–Fe (0.765), Pb–Cr (0.786), Pb–As (0.843) and Pb–Sr (0.832) showed high significant correlations at 99% confidence level. But Sb–Mo (0.584) showed moderate correlations at 99% confidence level. The pairs of Ni–Mn (0.8705), As–Fe (0.704), Se-Co (0.703), Sr–Cu (0.702), Mn–Ca (0.706), Sn–Mo (0.702) and Ce–Cd (0.706) showed high correlations at 95% confidence level. Besides, the pairs of Mg–Li (0.582), Na–B (0.602), Ti–Be (0.604), Mg–V (0.612), Mn–Cr (0.636), Fe–Na (0.608), Co–V (0.655), Se-Ti (0.590), Sr–As (0.646), Cd–Fe (0.687), Sn–V (0.678), Ba–Li (0.582), La–Na (0.667), La–Ti (0.592), La–Co (0.669), Ce–Ti (0.596), Ce–Co (0.601) and Pt–Mo (0.579) show moderate correlations at 95% confidence level. Table 4.16 Correlation matrix for the element concentrations in thyme samples.

Table 4.16. Correlation matrix for the element concentrations in thyme samples.

Element	Li	Be	B	Na	Mg	P	K	Ca	Ti	V	Cr	Mn	Fe	Co	Ni	Cu	Zn	As	Se	Sr	Mo	Cd	Sn	Sb	Ba	La	Ce	Pt	Tl	Pb
Li	1.000	0.281																												
Be	0.281	1.000																												
B	-0.238	-0.004	1.000																											
Na	-0.154	0.067	**0.602***	1.000																										
Mg	**0.582***	0.332	0.089	-0.049	1.000																									
P	-0.376	0.344	-0.043	-0.193	-0.087	1.000																								
K	-0.290	0.139	**0.720****	0.297	0.051	0.408	1.000																							
Ca	0.294	0.370	0.197	0.349	0.119	-0.047	0.351	1.000																						
Ti	0.173	**0.604***	0.224	0.434	0.186	0.125	0.267	**0.862****	1.000																					
V	0.556	0.331	-0.450	-0.256	**0.612***	0.021	-0.304	0.353	0.455	1.000																				
Cr	0.240	-0.315	-0.180	-0.268	-0.040	-0.161	0.099	0.430	-0.001	0.062	1.000																			
Mn	0.451	0.289	0.016	-0.129	0.214	0.160	0.475	**0.706***	0.381	0.262	**0.636***	1.000																		
Fe	0.567	0.264	-0.362	-0.255	**0.608***	-0.004	-0.156	0.503	0.463	**0.917****	0.397	0.475	1.000																	
Co	0.402	0.374	-0.002	0.420	**0.669***	-0.040	0.100	0.469	0.546	**0.655***	-0.062	0.279	0.623*	1.000																
Ni	0.289	-0.328	-0.039	-0.172	0.054	-0.270	0.184	0.545	0.111	0.241	**0.807****	**0.705***	0.490	0.105	1.000															
Cu	0.111	-0.475	-0.112	-0.272	-0.062	-0.304	-0.006	0.311	-0.019	0.247	**0.711****	0.379	0.481	-0.051	**0.880****	1.000														
Zn	0.207	-0.435	-0.100	-0.241	-0.050	-0.332	0.058	0.402	-0.009	0.194	**0.805****	0.539	0.456	-0.026	**0.958****	**0.970****	1.000													
As	0.241	-0.189	-0.039	0.070	0.284	-0.297	0.087	0.559	0.316	0.525	0.537	0.469	**0.704***	0.511	**0.819****	0.817**	**0.812****	1.000												
Se	0.294	0.423	-0.322	-0.071	0.520	0.320	-0.111	0.358	**0.590***	**0.902****	-0.113	0.162	**0.778****	**0.703***	-0.006	-0.004	-0.078	0.343	1.000											
Sr	0.412	-0.219	-0.045	-0.129	0.051	-0.300	0.190	0.529	0.038	0.083	**0.848****	**0.795****	0.361	0.074	**0.928****	**0.702***	**0.852****	**0.646***	-0.180	1.000										
Mo	0.228	-0.206	0.216	-0.252	0.204	-0.053	0.078	0.115	-0.108	0.060	0.421	0.318	0.275	-0.222	0.554	0.567	0.571	0.366	-0.161	0.503	1.000									
Cd	0.356	0.050	-0.119	-0.227	**0.827****	0.065	0.010	0.108	0.168	**0.742****	0.058	0.169	**0.687***	0.611*	0.180	0.116	0.082	0.396	**0.714****	0.035	0.104	1.000								
Sn	0.153	-0.414	0.027	-0.237	0.019	-0.293	0.086	0.287	-0.051	0.177	**0.678***	0.418	0.441	-0.064	**0.869****	0.970**	**0.953****	**0.790****	-0.092	**0.725****	**0.702***	0.083	1.000							
Sb	0.228	-0.404	-0.112	-0.243	-0.042	-0.291	0.055	0.434	0.031	0.209	**0.854****	0.558	0.495	-0.021	**0.945****	**0.963****	**0.991****	**0.797****	-0.045	**0.848****	**0.584***	0.083	**0.944****	1.000						
Ba	**0.582***	0.495	-0.539	-0.308	0.134	0.133	-0.352	0.228	0.206	0.478	0.247	0.384	0.556	0.179	0.136	0.151	0.173	0.157	0.336	0.229	0.188	-0.054	0.177	0.235	1.000					
La	0.303	0.431	-0.118	-0.123	**0.667***	0.298	0.110	0.408	**0.592***	**0.839****	-0.056	0.285	**0.746****	**0.669***	0.072	-0.008	-0.052	0.332	**0.924****	-0.091	-0.074	**0.844****	-0.068	-0.029	0.136	1.000				
Ce	0.335	0.522	-0.192	-0.227	0.557	0.330	0.119	0.420	**0.596***	**0.858****	-0.061	0.364	**0.765****	**0.601***	0.114	0.076	0.024	0.356	**0.907****	-0.054	-0.089	**0.706***	0.022	0.040	0.312	**0.943****	1.000			
Pt	-0.276	-0.388	0.157	-0.118	-0.222	0.194	0.134	-0.179	-0.384	-0.384	0.112	0.077	-0.328	-0.433	0.244	0.118	0.178	-0.067	-0.426	0.261	**0.579***	-0.141	0.193	0.140	-0.293	-0.343	-0.445	1.000		
Tl	0.702*	0.481	-0.443	-0.319	0.542	0.103	-0.372	0.179	0.216	0.732**	0.083	0.328	**0.737****	0.403	0.148	0.127	0.127	0.275	0.556	0.167	0.375	0.374	0.182	0.165	0.841**	0.437	0.511	-0.130	1.000	
Pb	0.246	-0.432	-0.115	-0.265	0.037	-0.327	0.049	0.398	0.005	0.279	**0.786****	0.536	0.524	0.040	**0.959****	**0.972****	**0.993****	**0.843****	0.006	**0.832****	0.568	0.190	**0.950****	**0.983****	0.164	0.042	0.107	0.149	0.166	1.000

*. Correlation is significant at the 0.05 level (2-tailed).
**. Correlation is significant at the 0.01 level (2-tailed).

More interpretations between elements and spas may be obtained using more powerful chemometric techniques such as principal component analysis (PCA).

4.5. Principal component analysis for the element concentrations in thyme samples.

By using the PCA subroutine of the IBM SPSS statistic V22 package (Release 22.0.0.0, 1989–2003), principal component analysis (PCA) was subsequently carried out on the correlation matrix to identify the underlying pattern of metals and to help the data interpretation. The PCA was applied to the data matrix of total metal concentrations (30 × 12 cases) of the thyme samples. The results show that five eigenvalues explain about 84.93% of the total variance and the fifth eigenvalue explains about 6.76 % of the variance. Principal component loading for thyme samples extracted three components in which it explains about 84.93% of the total variance with the contribution of each factor being 31.33%, 22.52%, 12.52%, 11.82% and 6.76%, respectively. For this reason, the first five eigenvalues were selected for further analysis. The five factor loadings extracted after performing the maximum rotation and communalities are given in Table 4.17. The factor 1 has high loadings for Zn, Sb, Pb, Ni, Sn, Cu, Sr, Cr, As and Mo, explains about 31.33 of the total variance. The factor 2 is loaded La, Cd, Se, Ce, V, Mg, Fe and Co explains 22.52% of the total variance. Factor 3 is loaded by Ba, Tl and Li, explains 12.52% of the total variance. The loading were for B, K, Ca, Ti, Be, Mn and Na on the fourth component and for P and Pt on the fifth component. A 3-D plot of the PCA loadings is illustrated in Figure 4.17 and the relationships among the elements are readily seen.

Table 4.17. The loading and the scores of the first five rotated principal components.

	The loading					The score					
Element	PC1	PC2	PC 3	PC 4	PC 5	Vocational	PC1	PC2	PC 3	PC 4	PC 5
Zn	**0.988**	0.001	0.053	-0.014	-0.063	Qandil	-0.350	-0.922	-0.445	-1.554	1.948
Sb	**0.982**	0.007	0.097	0.023	-0.043	Asos	-0.429	0.181	-0.576	2.037	-0.169
Pb	**0.982**	0.107	0.047	-0.044	-0.069	Balambo	-0.380	-0.759	-0.670	-0.183	-0.795
Ni	**0.971**	0.085	0.004	0.133	0.017	Karokh	-0.534	-0.527	-1.238	-0.044	-1.444
Sn	**0.944**	0.028	0.009	-0.084	-0.008	Eruh	-0.224	2.868	-0.482	-0.608	0.204
Cu	**0.940**	0.072	0.028	-0.125	-0.089	Şirvan	-0.238	-0.762	0.189	-0.504	0.171
Sr	**0.888**	-0.106	0.122	0.245	0.030	Kezer Kampus	2.845	-0.306	0.232	0.695	0.230
Cr	**0.831**	-0.098	0.159	0.107	0.063	Tillo	0.979	0.403	-0.263	-0.929	-0.220
As	**0.790**	0.431	-0.052	0.164	-0.257	Akdoğmuş Koyu	0.134	0.048	0.381	-0.612	-1.078
Mo	**0.646**	-0.013	0.054	-0.133	0.374	Kurtalan Üçyolu	-0.591	-0.352	1.323	-0.138	-0.812
La	-0.052	**0.945**	0.053	0.204	0.134	Kurtalan Zokayt	-0.642	0.248	2.356	0.507	0.466
Cd	0.115	**0.933**	-0.064	-0.181	0.046	Baykan	-0.570	-0.121	-0.807	1.334	1.499
Se	-0.113	**0.890**	0.243	0.181	0.011						
Ce	-0.010	**0.868**	0.202	0.264	0.168						
V	0.153	**0.859**	0.436	0.062	-0.109						
Mg	0.010	**0.771**	0.064	0.039	-0.023						
Fe	0.436	**0.761**	0.419	0.166	-0.047						
Co	-0.022	**0.727**	-0.001	0.428	-0.323						
Ba	0.131	0.076	**0.904**	0.263	0.084						
Tl	0.124	0.458	**0.762**	0.096	0.126						
B	-0.020	-0.124	-0.753	**0.364**	0.076						
K	0.122	0.012	-0.628	**0.501**	0.466						
Li	0.227	0.313	**0.595**	0.204	-0.223						
Ca	0.412	0.233	-0.003	**0.826**	-0.065						
Ti	-0.021	0.407	-0.010	**0.810**	-0.079						
Be	-0.443	0.258	0.393	**0.638**	0.221						
Mn	**0.580**	0.141	0.188	**0.586**	0.359						
Na	-0.214	-0.110	-0.499	**0.547**	-0.447						
P	-0.312	0.157	-0.003	0.120	**0.817**						
Pt	0.267	-0.321	-0.255	-0.296	**0.527**						
Eigenvalue	9.40	6.76	3.76	3.55	2.03						
Variance (%)	31.33	22.52	12.52	11.82	6.76						
Cumulative (%)	31.33	53.85	66.37	78.19	84.95						

Table 4.17 shows the two way loadings and score plots. Every principal component was plotted against PC1 to show high percentage of the total variance (31.33–84.95).

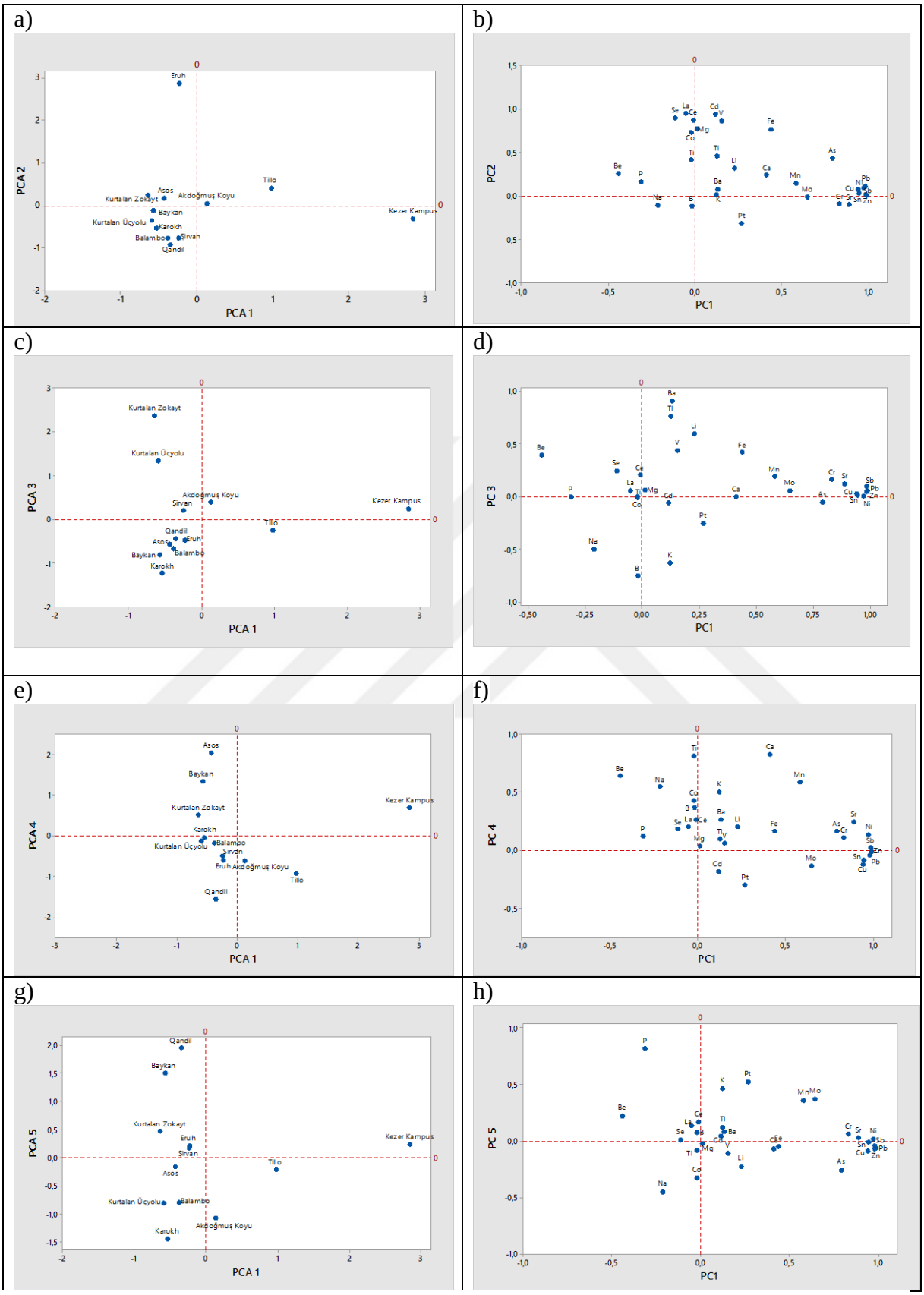


Figure 4.52. The score and loading plots (a, c, e and g are the score plots and b, d, f and h are the loading plots).

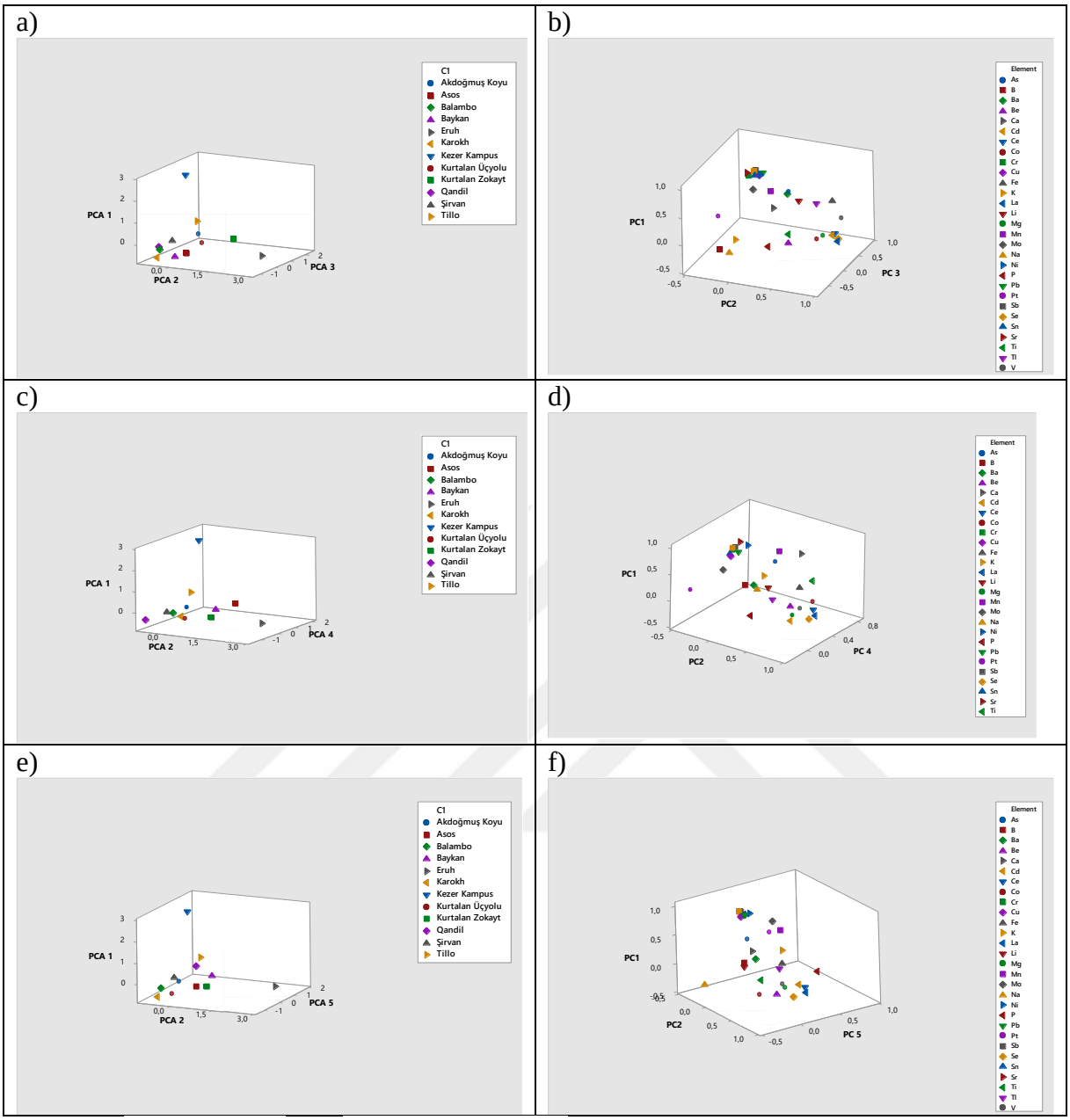


Figure 4.53. 3D Scatterplot of PCA score and loading plots)

The classification of the species from the view point of metal contents can be made using three way PC score graphs. The highest percentage of total variance of about 53.85 was observed with PC 1–2 while PC 1–3 score graph shows about 43.85 % of total variance. It can be seen from the PC 1–2, PC 1–3 graphs (Figure 4.52), Table 4.17 and Figure 4.53 that the thermal waters of spas can be classified into five groups. These groups were:

Group 1: Kezer Kampus and Tillo

Group 2: Eruh

Group 3: Şirvan, Akdoğmuş Köyü, Kurtalan uçyol and Kurtalan-zokayt

Group 4: Asos, Karokh and Balambo

Group 5: Qandil and Baykan

4.6. Cluster analysis

Cluster analysis is the most widely used unsupervised pattern recognition technique in chemometrics (Brereton, 2003). Objects will be grouped in clusters in terms of their nearness or similarity. The cluster analysis was applied using the Minitab 17 (Licensing: 17.1.0.0, 2013) package. The measurement is based on the squared Euclidean distance. Seven clusters were obtained from the analysis. These groups contain:

Group 1: Qandil and Baykan

Group 2: Asos, Karokh

Group 3: Balambo, Kurtalan uçyol and Kurtalan-zokayt

Group 4: Şirvan

Group 5: Eruh

Group 6: Akdoğmuş Köyü

Group 7: Kezer Kampus and Tillo

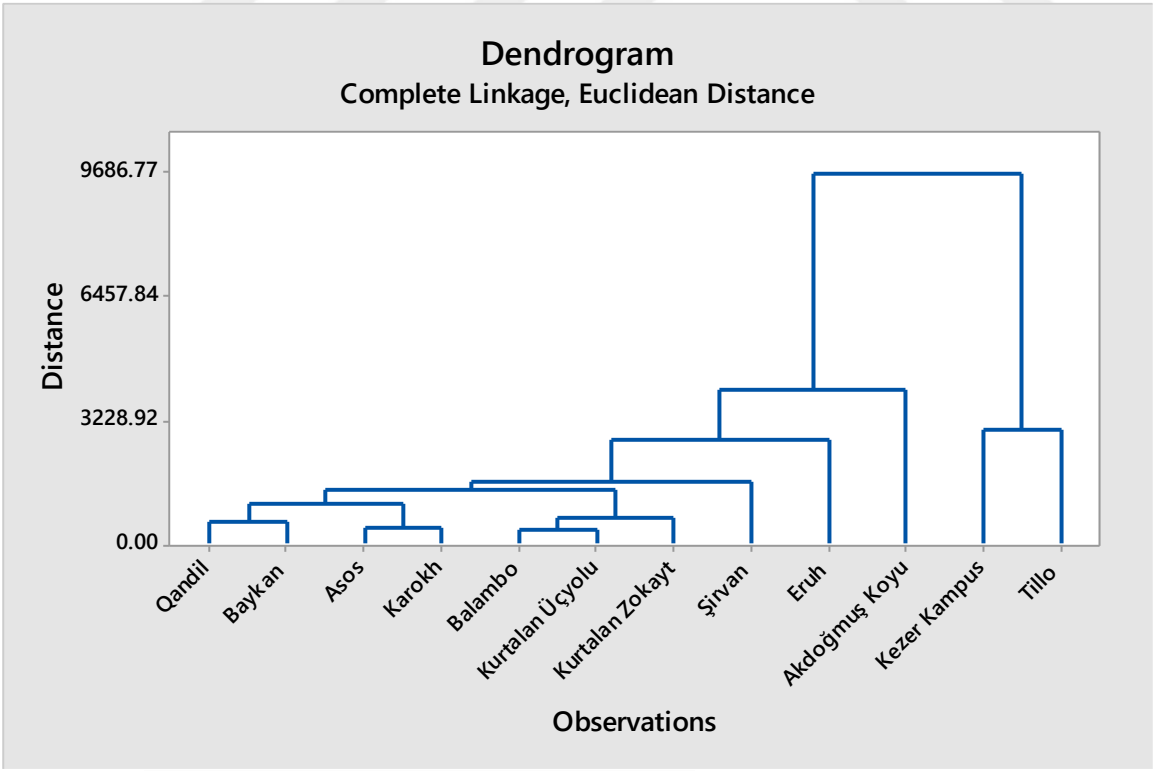


Figure 4.54. Dendrogram of cluster analysis for thyme samples

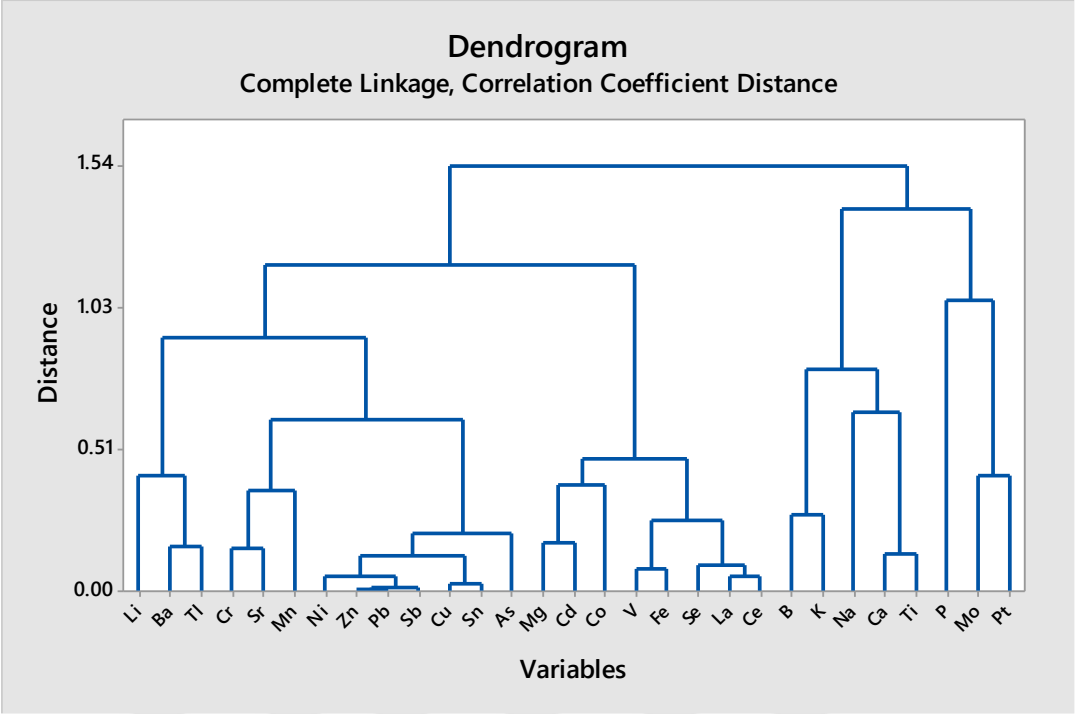


Figure 4.55. Dendrogram of cluster analysis for metal concentrations

Figure 4.55 shows that the metal concentrations are divided into 7 groups. For elements: Group 1 had Li, Ba and Tl, Group 2 had Cr, Sr and Mn, Group 3 had Ni, Zn, Pb, Sb, Cu, Sn and As, Group 4 had Mg, Cd, Co, V, Fe, Se, La and Ce, Group 5 had B, K, Na, Ca, and Ti, Group 6 had P; Group 7 had Mo and Pt.

5. CONCLUSION

In this study, twelve examples of thyme taken from the Iraqi Kurdish region and around Siir-Turkeyt were examined. The metal concentrations, total phenolic amount, total flavonoid amount, antioxidant amount, and phenolic substance components of these samples were analyzed. The concentrations of elements such as Li , B , Be , Na, Mg , P , K , Ca , Ti , V , Cr , Mn, Fe , Co, Ni , Cu , Zn , As , Se , Sr , Mo , Cd , Sn , Sb , Ba , La , Ce , Pt ,Tl and Pb were performed with a Thermo Scientific ICAP Q brand ICP MS device. The content of minerals in thyme are mainly affected by variable natural climate condition in different geographical are distributed between the Siirt area in Turkey and Kurdistan regional of Iraq. The level of metals obtained shows a comparable result with other reported values in some cases. , the contents of Pb , Cd, Fe, Ni, Na, Mg, P, K , Ca, Ti, V, Zn, Co, As and other all selected metals in thyme are low and non- toxic which can be safely used for pharmaceutical and edible purposes without any hazard effects on human health. Thirty minerals were quantified for each sample Classification of thyme according to their origin (mountains) was achieved by pattern recognition techniques on the mineral data. By means of correlation analysis, principal component analysis and cluster a good separation by geographical origin is obtained when scores for the five first principal components are plotted. The results showed differences in the metal concentrations of all sample. According to principle component analysis and cluster analysis, the thyme collected regions can be divided into five groups. The first group is Kezer campus and Tillo, the second group is Eruh, and the third group is Şirvan, Akdoğmuş Köyü, Kurtalan uçyol and Kurtalan-zokayt. The forth group is Asos, Karokh and Balambo. The fifth group is Qandil and Baykan.

Total phenolic content was assessed by Folin Ciocalteu's method and varied between 84.015 - 387.526 µg gallic acid/mL of extract. The total flavonoid content was determined by the aluminum chloride colorimetric method and varied 69.980-935.750 rutin µg/mL of extract. The antioxidant potency was determined by analyzing FRAP and DPPH methods. In FRAP assay the highest antioxidant activity (IC_{50}) was found 25.175 ± 0.037 µg/mL extract. In DPPH method the maximum antioxidant activity percentages of extract was found 88.113%. Metal chelating activity was estimated by method of (Rival et al, 2001). Iron-chelating activities of the all samples were greater than 70%.

Thyme leaves are utilized for various uses in different areas in all the world Therefore, to utilize thyme for a day-to-day consumption, it has to be taken care in cultivation,

processing, transporting and selling of thyme leaves to the consumer (added to staple foods for flavoring, utilized for making tea, utilized for medication, etc)

6. REFERENCES

Abu-Darwish, M. S., Abu-Dieyeh, Z. H., Mufeed, B., Al-Tawaha, A. R. M., & Al-Dalain, S. Y. A. (2009). Trace element contents and essential oil yields from wild thyme plant (Thymus serpyllum L.) grown at different natural variable environments, Jordan. J. Food Agric. Environ, 7(3-4), 920-924.

Abu-Arab, A.A.K. and Abou Donia, M.A. 2000. Heavy metals in Egyptian spices and medicinal plants and the effect of processing on their levels. J. Agric. Food Chem. 48:2300-2304

Acquarone C, Buera P, Elizalde B (2007) Pattern of pH and electrical conductivity upon honey dilution as a complementary tool for discriminating geographical origin of honeys. Food Chem 101:695–703.

Agency for Toxic Substances and Disease Registry. (2008). ToxFAQs. Retrieved March 2009 from http://www.atsdr.cdc.gov/toxfaq.html.

Agrios G(1997). Plant pathology. 4th ed. San Diego: Academic Press;.

Ajasa AMO, Bell MO, Ibrahim AO, Ogunwande IA, Olawore NO (2004) Heavy trace metals and micronutrients status in herbal plants of Nigeria. Food Chemistry 85, 67-71

Ajasa, A.U.O., M.O. Bello, A.O. Ibrahim, I.A. Ogunwarde and N.O. Olawore, 2004. Heavy trace metals and macronutrients status in herbal plants of Nigeria. Food Chem., 85: (67–71)

Al-Alawi, M.M. and K.L. Mandiwana, 2007. The use of Aleppo pine needles as a bio-monitor of heavy metals in the atmosphere. J. Haz. Mat., 148: 43–46

Ali Ghasemzadeh, and Neda Ghasemzadeh(2011), Flavonoids and phenolic acids: Role and biochemical activity in plants and human Journal of Medicinal Plants Research Vol. 5(31), pp. 6697-6703, 23 December,

Al-Jundi, B., 2000. Determination of trace elements and heavy metals in the Zarka River sediments by instrumental neutron activation analysis. Nucl. Instr. Meth. Phys, Res., 170: (180–186)

Amacher, M. C., (1996) "Nickel, Cadmium and Lead" in Methods of Soil Analysis, Part 3, SSSA, pp. 739-768.

Amrutha VA, Bhaskar VSC (2010). Antioxidative and Antimicrobial Activity of Methanol and Chloroform Extracts of Gmelina Arborea Roxb. Int. J. Biotechnol. Biochem., 6: (139-144).

Anass Terrab ,† Dolores Hernanz ,§ and Francisco J. Heredia Inductively Coupled Plasma Optical Emission Spectrometric Determination of Minerals in Thyme Honeys and Their Contribution to Geographical Discrimination Agric. Food Chem., 2004, 52 (11), pp 3441–3445

Andlauer, W., & Fürst, P. (1998). Antioxidative power of phytochemicals with special reference to cereals. Cereal Foods World, 43, 356–359.

Ansari R, Kazi TG, Jamali MK, Arain MB, Wagan MD, Jaibani N, Afridi HI, Shah AQ (2009) Variation in accumulation of heavy metals in different verities of sunflower seed oil with the aid of multivariate technique. Food Chemistry 115, (318-33)

Asfaw. N, H. J. Storesund, L. Skattebol, F. Tonnesen, A. J. Aasen, Volatile oil constituents of two Thymus species from Ethiopia, Flavor Fragr. J., 2000, 15, (123-125)

Atoui, A. K., Mansouri, A., Boskou, G. And Kefalas, P., 2005. Tea and Herbal infusions: Their antioksidant activity and phenolic profile, Food Chemistry, 89, (27-36).

Baianu, I.C., Costescu, D.M., and You, T. 2002. Novel Techniques for Microspectroscopy and Chemical Imaging Analysis of Soybean Seeds and Embryos.(2002). Soy2002 Conference, Urbana, Illinois.

Barlow SM (1990). Toxicological Aspects of Antioxidants Used as Food Additives, in Food Antioxidants, Hudson BJF (Ed.), Elsevier, Amsterdam, p. 23.

Bartosz, G. (1997). Oxidative stress in plants. Acta Physiologiae Plantarum, 19(1), 47–64.

Baytop, T., 1999. Türkiye'de Bitkiler İle Tedavi, Geçmişte Ve Bugün. İstanbul Üniversitesi, Eczacılık Fakültesi, İstanbul, 550s.

Bazylko A, Strzelecka H (2007). A HPTLC densitometric determination.
Basgel S, Erdemoglu SB (2006). Determination of mineral and trace elements in some medicinal herbs and their infusions consumed in Turkey. Sci. Total Env. 359:82-89.

Blagojević, N., Damjanović-Vratnica, B., Vukašinović-Pešić, V., & Đurović, D. (2009). Heavy Metals Content in Leaves and Extracts of Wild-Growing Salvia Officinalisfrom Montenegro. The Polish Journal of Environmental Studies, 18(2), 167-173.

Benzie IFF, Strain JJ (1996). Ferric reducing ability of plasma (FRAP) as a measure of antioxidant power: The FRAP assay. *Anal Biochem. 239*:70-7

Bin, C., Xiaouru, W. and Lee, F.S.C. 2001. Pyrolysis coupled with atomic absorption spectrometry for determination of mercury in Chinese medicinal materials. Anal. Chim. Acta 447:161–169.

Bourne, G.H. (1985) Minerals in Food and Nutritional Topics. S. Karger AG, Basel.

Boullata JI: Trace elements in critically ill patients. J Infus Nurs 2013; 36:16-23.

Brereton, R. G. (2003). *Chemometrics: Data Analysis for the Laboratory and Chemical Plant*. *Book (Chemo)* (Vol. 8). https://doi.org/10.1198/tech.2004.s738

Buenz, I.F.F; Schenepple, D.J; and Motley, T.J. (2004) Technique: bio prospecting historical herbal texts by hunting for new leads in old tomes. Trends in pharmacological Sciences 25, 494-498.

Burda S, Oleszek W (2001). Antioxidant and antiradical activities of flavonoids. J. Agric. Food Chem., 49(6): 2774-2779.

Cacar, 2003: Canadian Contaminants Assessment Report II, Sources, Occurrence, Trends and Pathways in the physical environment, Northern Contaminants program, Minister of Indian Affairs and Northern Development, Minister of Public Works and Government Services Canada, p,p. 332.

Caldas E.D., Machado L.L(2004). Cadmium, mercury and lead in medicinal herbs in Brazil. Food Chem. Toxicol. (42, 599),.

Cavero S, Jaime L, Martin-Ălvarez PJ, Señoráns FJ, Reglero G, Ibañez E (2005). In vitro antioxidant analysis of supercritical fluid extracts from rosmary (Rosmarinus officinalis L). J. Eur. food Res. technol. 221 (478-486).

Coogan, T.P., Latta, D.M., Snow, E.T., Costa, M. and Lawrence, A. (1989) Toxicity and Carcinogenicity of Nickel Compounds. Critical Reviews in Toxicology, 19, 341-384.

Cruz Cabral L, Fernandez Pinto V, Patriarca A. Application of plant derived compounds to control fungal spoilage and mycotoxin production in foods. Int J Food Microbiol. 2013; 166 (1):1–14

Ukbay∗, Ebru Kuyumcuturk J (2009). Determination of trace element contents of Thymus species from Turkey F. Zehra K¨UC Chem 34, 911 – 919.

Davis P. H. (1982). Flora of Turkey and the East Aegean Islands. Vol.7. Edinburgh University Press, Edinburgh, pp. (349-382)

Demsew .S. N. Asfaw, Some useful indigenous labiates from Ethiopia, Lmiales Newsletter, 1994, 3, 5.

Demsew. S. the Genus Thymus (Labiatae) in Ethiopia, Opera. Bot.1993, 121, (57-60).

Dixon RA & Paiva NL. 1995. Stress-induced phenylpropanoid metabolism, Plant Cell, 7: (1085-1097).

Diaspro, A., and Robello, M. (1999). Multi-photon Excitation Microscopy to Study Biosystems. European Microscopy and Analysis. 5:(5-7).

Djukic-Cosic, D. and M. Curcic, 2007. Cmiljanovic, M. Vasovic, I. and V. Matovic. 2007. Heavy metal contents in samples of hypericum and thymus spec. Graz, Austria September 2–6 during legume nodulation, Trends in Plant Sci. 12(7):282-283.

Dorman HJ, Bachmayer O, Kosar M, Hiltunen R. Antioxidant properties of aqueous extracts from selected lamiaceae species grown in Turkey. J Agric Food Chem. 2004; 52(4):(762–70).

Duh PD, Tu YY, Yen GC (1999). Antioxidants activity of aqueous extract of Harnjyur (Chrysanthemum morifolium Ramat). Lebensmwiss Technol. 32: (269-277).

Dwyer J. 1995. Overview: dietary approaches for reducing cardiovascular disease risks, J Nutr, 125:656S-665S.

Eastern Research Group. (2001). Summary report for the ATSDR expert panel meeting on tribal exposures to environmental contaminants in plants. Retrieved on January 28, 2009fromhttp://www.engg.ksu.edu/CHSR/outreach/ tosnac/docs/NAreport_fnl032301.pdf

Ebrahimzadeh MA, Pourmmorad F, Hafezi S (2008). Antioxidant activities of Iranian corn silk, Turk. J. Biol. 32(43-49)

Edrir. A. E. (2007). Pharmaceutical and therapeutic potentials of essential oils and their individual volatile constituents: A review. *Phytother. Res.* 21, 308-323. D. Kalemba and A. Kunicka (2003). Antibacterial and antifungal properties of essential oils, *Curr. Med. Chem.* 10, 813-829

El-Mokhtar El Ouariachi1, Julien Paolini2*, Abdelhamid Bouyanzer1, Pierre Tomi2, Belkheir Hammouti1, Rachid Salghi3, Lhou Majidi4 and Jean Costa2(2011). Chemical composition and antioxidant activity of essential oils and solvent extracts of Thymus capitatus (L.) Hoffmanns and link from Morocco Journal of Medicinal Plants Research Vol. 5(24), pp. 5773-5778, 30 October,

El-Rjoob, A. W. O, Massadeh, A. M., & Omari, M. N. (2008). Evaluation of Pb, Cu, Zn, Cd, Ni and Fe levels in Rosmarinus officinalis labaiatae (Rosemary) medicinal plant and soils in selected zones in Jordan. Environmental monitoring and assessment, 140(1), (61-68).

Ersn.E.coon.J.T.2011.heavy metals in traditional chines medicines: asystematic review clinical pharmacoloji and therapeutics, 70,497, 504.

Evans W. C. (1989), Trease and Evans' Pharmacognosy. Bailliere Tindall, London, 13th ed., pp. 216D217.

Faleiro, L., Miguel, G., Gomes, S., Costa, L., Venâncio, F., Teixeira A., Figueiredo, A.C., Barroso, J.G., and Pedro, L.G. 2005. Antibacterial and antioxidant activities of essential oils isolated from Thymbra capitata L. (Cav.) and Origanum vulgare L. Journ. Agri. Food Chem. 53☹8162-8168).

Farman, M. (1990). Isolation and characterization of flavonoids from Indigofera hebepetala, Quaid-i- A Azam University, Islamabad.

Fecka I, Raj D, Krauze-Baranowska M (2007). Quantitative determination of four water-soluble compoundsin herbal drug from Lamiaceae using different chromatographic techniques, Chromatographia, 66: (87-93).

Field, T. S., Lee, D. W. & Holbrook, N. M. (2001). Why leaves turn red in autumn, the role of anthocyanins in senescing leaves of Red-Osier Dogwood, Plant Physiol., 127: (566–574).

Fraga, C.G. (2005) Relevance, Essentiality and Toxicity of Trace Elements in Human Health. Molecular Aspects of Medicine, 26, (235-244).

Früchtenicht K, Vetter H. Charakterisierung der Schwermetallbelastung durch Messung der Schwermetallgehalte in Pflanzen. In: Landwirtschaftliche Forschung, Kongressband. Frankfurt am Main, Germany: J.D. Sauerländer's Verlag; 1982; 39:(154-64).

Ghadir Zayzafoon, Adnan Odeh, Abdul Wahab Allaf Determination of essential oil composition by GC-MS and integral antioxidant capacity using photochemiluminescence assay of two Thymus leaves: Thymus syriacus and Thymus cilicicus from different Syrian locations Vol. 58 No. 4 2012)

Ghasemi Dehkordi, N. Pharmacopia of Iranian plants, First Edition, Isfahan. Ministry of Health and Medical Education. Deputy of Food and Medicine 2002.

Grigore1 A., Ina Paraschiv1, S. Colceru-Mihul1, C. Bubueanu1, E. Draghici1, M. Ichim(2010). Chemical Composition and Antioxidant Activity of Thymus vulgaris L. volatile oil obtained by two different methods Vol. 15, No. 4

Goldhaber, S. B. Regul. Toxicol. Pharm. 2003, 38, 232-242.

Goldman, P. Ann. Int. Med. 2001, 135, 594-600.

Grayer, R.J., Bryan, S.E., Veitch, N.C., Goldstone, F.J., Paton, A., Wollenweber, A. (1996) External flavones in sweet basil, Ocimum basilicum, and related taxa. Phytochemistry, Vol.43, Nr. 5, pp (1033–1039).

Gu L, Wu T, Wang Z. TLC bioautography-guided isolation of antioxidants from fruit of Perilla frutescens var. acuta. LWT - Food Sci Technol. 2009; 42:131e136

Guillen, M.D., Manzanos, M.J., (1998) Study of the composition of the different parts of Spanish Thymus vulgaris L. plant. Food Chemistry, Vol. 63, pp. 373.

Gülçin at G, Beydemir ElmastaM, Küfreviolu Ö (2004). Comparison of antioxidant activity of clove (Eugenia caryophylata Thunb) buds and lavender (Lavandula stoechas L.). Food Chem., 87: (393-400).

Gumus. T (2010). Determination of the changes of antifungal properties of Satureja hortensis, Thymus vulgaris and Thymbra spicata exposed to gamma irradiation, Radiat. Phys. Chem. 79, (109-114).

Guyton, C. A., Hall, J. E. (2006). Text book of medical physiology, 11th edition, Mc-Graw Hill.

Hall, C.A.; Cuppet, S.L. Strcture activities of natural antioxidants. In: Antioxidant Methodology in vivo and in vitro Concepts. Aruoma 01, Cuppet SL (eds). Champaign, IL, 1997; pp. 2-29.

Halliwell B (1995). Antioxidant characterization methodology and mechanism. Biochem Pharmacol., 49: (1341-1348).

Halliwell B, Gutteridge J. (1999). Free Radicals in Biology and Medicine. Oxford University Press, New York,.

Halliwell B, Gutteridge JM (1984). Lipid peroxidation, oxygen radicals, cell damage, and antioxidant therapy, *Lancet.* 8391:1396-1397, Wallace DC (1999). Mitochondrial diseases in man and mouse, *Science.* 283:1482-1488.

Halliwell B. Gutteridge J. M. C. (2000). Free radicals in Biology and medicine. Oxford University Press, 148-149.

Halliwell, B., & Gutteridge, J. M. C. (2003). Free Radicals in Biology and Medicine. Oxford, UK: Oxford University Press.

Halliwell. B (1989) and J. M. Gutteridge, Free Radicals in Biology and Medicine, Clarendon Press, Oxford, UK.

Halliwell. B(1997), "Antioxidants and human disease: a general introduction," Nutrition Reviews, vol. 55, no. 1, pp. S44–S52.

Harborne, J. B. (ed.) (1998). Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis, 3rd ed. Chapman and Hall, London.

Harborne JB. 1994. The flavonoids: Advances in research since 1986, New York: Chapman and Hall.

Harborne JB & Baxter H. 1999. Handbook of Natural Flavonoids Chichester: Wiley.

Harnjyur (Chrysanthemum morifolium Ramat). Lebensmwiss Technol. 32: (269-277).

Raja Sudarajan N, Ahamad H, Kumar V (2006). Cytisus scoparius Link-A natural antioxidant. 6: (1-7)

Havsteen BH (2002). The biochemisty and medicinal significance of the flavonoids. Pharmacol. Ther. 96: (67-202).

Hazzit, M., Baaliouamer, A., Verissimo, A.R., Faleiro, M.L., Miguel, M.G., 2009. Chemical composition and biological activities of Algerian Thymus oils, Food Chemisty. 116, (714-721).

Hollman PCH, Hertog MG.L, Katan MB (1996). Analysis and health effects of flavonoids. Food Chem., 57(1): 43-46.

Hostettmann, K., MARSTON*(2005) Andrew,* Hostettmann, *Maryse ..., Pages* 50-*127 ... Sarker, S.D. (*Ed.*)*

Hras, A. R., Hadolin, M., Knez, Z., & Bauman, D. (2000). Comparison of antioxidative and synergistic effects of rosemary extract with a-tocopherol, ascorbyl palmitate and citric acid in sunflower oil. Food Chemistry, 71, (229–233).

Hsieh PC, Mau JL, Huang SH. Antimicrobial effect of various combinations of plant extracts. Food Microbiol. 2001; 18(1):35–43

Ibironke A. Ajayi1, Rotimi A. Oderinde1, Adewale Adewuyi1 Joseph I. Uponi2 Evaluation of Selected Heavy Metals and Macronutrients Status of 10 Medicinal Plants from Nigeria e-ISSN: 2278-3008, p-ISSN:2319-7676. Volume 8, Issue 2 (Nov. – Dec. 2013), PP 12-17).

Imelouane B., Amhamdi H., Wathelet J. P., Ankit M., Khedid K. and El Bachiri A. (2009) Chemical composition of the essential oil of thyme (Thymus vulgaris) from Eastern Morocco. International Journal of Agriculture and Biology 11: 205–208.

Ismail, F., Anjum, M.R., Mamon, A.N. and Kazi, T.G. (2011) Trace Metal Contents of Vegetables and Fruits of Hyderabad Retail Market. Pakistan Journal of Nutrition, 10, (365-37).

Ioannis K. Karabagias & Maria V. Vavoura & Anastasia Badeka & Stavros Kontakos & Michael G. Kontominas Differentiation of Greek Thyme Honeys According to Geographical Origin Based on the Combination of Phenolic Compounds and Conventional Quality Parameters Using ChemometricFood Anal. Methods (2014) (7:2113–2121).

Jamil, D. M. (2009). Characterization, antioxidant and antimicrobial activity of Thymus syriacus Boiss.Var. Thymus and Thymbra Spicata L. from Kurdistan – Iraq.PhD. Thesis.

Jamil, D. M.; J .E. Brown, D. Driscoll and N. K. Howell. (2010). Characterization and antioxidant activity of the volatile oils of Thymus syriacus Boiss.var syriacus and Thymbra spicata L. grown wild in Kurdistan-Iraq.;6(OCE1).

Javanmardi, J., Khalighi, A., Kashi, A., Bais, H.P., Vivanco, J.M. (2002) Chemical characterization of basil (Ocimum basilicum L.) found in local accessions and used in traditional medicines in Iran. Journal of Agricultural and Food chemistry, Vol. 50, pp. (5878–5883).

Jayasingne C., Gotoh N., Aoki T., Wada S. (2003) Phenolic Composition and Antioxidant activity of Sweet basil (Ocimum basilicum L.). Journal of Agricultural and Food chemistry, Vol. 51, pp. (4442–4449).

Johnsson, L. 2003. Hazards of heavy metal contamination. British Medical Bulletin 68☺167–18).

Jones, J.B. and V.W. Case, 1990. Sampling, Handling and Analyzing Plant Tissue Samples. In: Westerman, R.L. (ed.), Soil Testing and Plant Analysis, pp: 404–409. Madison

Justesen, U. (2000) Negative atmospheric pressure chemical ionization low-energy collision activation mass spectrometry for the characterization of flavonoids in extracys of fresh herbs. Journal of Chromatography A, Vol. 902, pp. 369–379.

Kara, D. (2009). Evaluation of trace metal concentrations in some herbs and herbal teas by principal component analysis. *Food Chemistry*, *114*(1), 347–354.

Karadaş, C., & Kara, D. (2012). Chemometric approach to evaluate trace metal concentrations in some spices and herbs. *Food Chemistry*, *130*(1), 196–202.

Koca, N., Karadeniz, F., 2003. Productıon Mechanısms of Free Radıcal and Antıoxıdant Defence Systems in The Body. Gıda Mühendisliği Dergisi. Ankara. (16), 32-37.

Küçükbay, F. Z., Kuyumcu, E., Çelen, S., Azaz, A. D., & Arabaci, T. (2014). Chemical composition of the essential oils of three Thymus taxa from Turkey with antimicrobial and antioxidant activities. *Records of Natural Products*, *8*(2), 110.

Lee S, Suh S, Kim S (2000). Protective effects of the green tea polyphenol (-)-epigallocatechin gallate against hippocampal neuronal damage after transient global ischemia in gerbils, Neurosci. Lett. 287:191-194.

Lee SC, Kim JH, Nam KC, Ahn DU (2003). Antioxidant properties of far infrared-treated rice hull extract in irradiated raw and coked turkey breast. J. Food Sci. 68: 1904-1909

Lee, S. E., Hwang, H. J., Ha, J. S., Jeong, H. S., & Kim, J. H. (2003). Screening of medicinal plant extracts for antioxidative activity. Life Science, 73, 167–179.

Li HB, Wong CC, Cheng KW, Chen F (2008). Antioxidant properties in vitro and total phenolic contents in methanol extracts from medicinal plants. Food Sci. Technol., 41: 385-390.

Li HY, Hao ZB, Wang XL, Huang L, Li JP. Antioxidant activities of extracts and fractions from Lysimachia foenum-graecum Hance. Bioresour Technol. 2009; 100:970e974

Lozak, A., Sołty, K., Ostapczuk, P. and Fijałek, Z. 2002. Determination of selected trace elements in herbs and their infusions. Sci. Total Environ.289:33–40.

Lozak, A.; Soltyk, K.; Ostapczuk, P.; Fijalek, Z. Sci. Total Environ. 2002, 289, 33-40.

Luiz, M.; Biasutti, A.; Garcia, N.A. Micellar effect on the scavenging of singlet molecular oxygen by hydroxybenzenes. Redox Rep. 2002, 7, 23–28.

Karadas C., Kara D (2012). Chemometric approach to evaluate trace metal concentrations in some spices and herbs. Food Chem. (130, 196).

Mahdavi, D.L.; Salunkhe, D.K (1995). Toxicological aspects of food antioxidant. In D. L. Mahdavi, S.S. Deshpande; D.K. Salunkhe (Eds.), Food antioxidants. New York: Marcel Dekker.

Majid AAB, Sarmani S, Yusoff NI, Wei YK, Hamzah F (1995) Trace elements in Malaysian medicinal plants. Journal of Radionanalytical Nuclear Chemistry Articles 195, 173-183

Maksimović, S., M. Jakovljević and D. Radanović, 1999. Influence of agro ecological factors on chemical composition of sage (Salvia officinalis L.), In: Brkić, D. (ed.), Sage (Salvia officinalis L.), pp: 61–80. Belgrade.

Maksimovic, M., Vidic, D., Milos, M., olic, M. E. S, Zic, S. A., & Siljak-Yakovlev, S. (2007). Effect of the environmental conditions on essential oil profile in two Dinaric Salvia species: S. brachyodonVandas and S. officinalis L. Biochemical Systematic and Ecology, 35, 473-478.

Mamani, M. C. V.; Aleixo, L. M.; Abreu, M. F.; Rath, S.J. Pharm. Biomed. Anal. 2005, 37, 709-713.

Manzoor Iqbal Khattak (2013)Determination of Trace Elements In Some Medicinal Plants In Balochistan Sci.Int (Lahore), 25(3), 599-602.

Marimuthu P, Wu CL, Chang HT, Chang ST (2008). Antioxidant activity of the ethanolic extract from the bark of Chamaecyparis obtusa var. formosana. J Sci Food Agric.; 88:1400-1405

Mensah JK, Okoli RI, Ohaju-Obodo JO, Eifediyi K (2008) Phytochemical, nutritional and medical properties of some leafy vegetables consumed by Edo people of Nigeria. African Journal of Biotechnology 7, 2304-2309

Middleton EJ, Kandaswami C, Theoharides TC., (2000) The effects of plant flavonoids on mammalian cells: implications for inflammation, heart disease, and cancer. Pharmacol. Rev. 52: 673-751

Miguel, M.G., Falcato-Simões, M., Figueiredo, A.C., Barroso, J.M.G., Pedro, L.G., and Carvalho, L. M.. 2005. Evaluation of the antioxidant activity of Thymbracapitata, Thymus mastichina and Thymus camphoratus essential oils. Journ. Food Lipids 12:181-197.

Miguel, M.G., Figueiredo, A.C., Costa, M.M., Martins, D., Duarte, J., Barroso, J.G., and Pedro, L.G. 2003a. Effect of volatile constituents isolated from Thymus albicans, Th. mastichina, Th. carnosus and Thymbra capitata in sunflower oil. Nahrung/Food, 47: 397-402.

Miguel, M.G., Figueiredo, A.C., Costa, M.M., Martins, D., Duarte, J., Barroso, J.G., and Pedro, L.G. 2003b Effect of the essential oils isolated from Thymbra capitata (L.) Cav. on olive and sunflower oils. Grasas Y Aceites 54:219-223

Miroslowki, J.; Wiechula, D.; Kwapuliski, J.; Rochel, R.; Loska, K.; Ciba, J. Bromat. Chem. Toksykol. 1995, 28, 363-372.

Mohammad S. Abu-Darwish 1, Ziad H. Abu Dieyeh 2, Batarseh Mufeed 3, Abdel Rahman M. Al-Tawaha 4*and Sati Yassin Ahmed Al-dala, 2009, Trace element contents and essential oil yields from wild thyme plant (Thymus serpyllum L.) grown at different natural variable environments, Jordan Journal of Food, Agriculture & Environment, Vol.7 (3&4), 920 924.

Morales Morales R. (1997), Synopsis of the genus Thymus L. inthe Mediterranean area. Logascalia 19, 249D262

Mustafa Abdullah YILMAZ, Bazi *Achillea L*. Türlerinin LCMS-IT/TOF ve LC MS/Ms İle Metabolik Profillerinin Çikarilmasi Ve Biyolojik Aktivitelerinin Belirlenmesi, Doktora Tezi, Eylül 2015)

Nasuti C, Gabbianelli R, Falcioni G, Cantalamessa F (2006) Antioxidative and gastroprotective activities of anti-inflammatory formulationsderived from chestnut honeyin rats.Nutr Res 26:130– 137

Noguchi N, Niki E. (1999) Diet Nutrition and Health, 20th ed. Papas M. P., CRC Press, Florida

Norins A L: Free radical formation in the skin following exposurceto ultraviolet light. J I Invest Dermatol 39:445, 1962.

Noori, M. (2002). Characterization of the Iranian species of Sophora and Ammodendron (Leguminosae; Sophoreae), PhD Thesis, University of London and Royal Botanic Gardens, Kew, UK.

Noori, M., Chehreghani, A. & Kaveh, M. (2009). Flavonoids of 17 species of Euphorbia, (Euphorbiaceae) in Iran, Toxicological and Environmental Chemistry, 91 (3): 409-418.

Nriagu, J.O., Pacyna, J.M. 1988. Quantitative assessment of worldwide contamination of air, water and soils by trace metals. Nature 333 (6169): 134-139.

Occupational Health and Safety Administration. (2004). Safety and health topics: Heavy metals. Retrieved on December 30, 2008 from http://www.osha.gov/SLTC/metalsheavy/index.html

Odetola AA, Akojeun SM (2000) Anti-diarrhea and gastro-intestinal potentials of the aqeous extract of Phyllantus of the aqeous extract of Phyllantus amarus (Eupthorbiacease). Olawoyin, R., Oyewole, S.A. and Grayson, R.L. (2012) Potential Risk Effect from Elevated Levels of Soil Heavy Metals on Human Health in the Niger Delta. Ecotoxicology and Environmental Safety, 85, 120-130.Journal o of luteolin in Thymus vulgaris and its extracts. J. Fitotherapia. 78:391 -395.

Olu-Owolabi, B.I., Fakayode, S.O., Adebowale, K.O. and Onianwa, P.C. (2007) Proximate and Elemental Composition and Their Estimated Daily Intake in Infant Formulae from Developed and Developing Countries: A Comparative Analysis. Journal of Food, Agriculture & Environment, 5, 40-44.

Palacios, G., Gomez, I., Moral, R., and Mataix, J., (1995) "Nickel Accumulation in Tomato Plants, Effect on Plant Growth" Frecenius Envir. Bull. 4: 469-474.

Park, E.J. and Pezzutto, J.M. (2002): Botanicals in Cancer Chemo preventives. Cancer and Metastasis Reviews, 21, 231-255.

Parr, A. J. & Bolwell, G. P. (2000). Phenols in the plant and in man. The potential for possible nutritional enhancment of the diet by modifying the phenols content or profile, Journal of the Science of Food and Agriculture, 80: 985–1012.

Pedersen J. A. (2000), Distribution and taxonomic impli-cations of some phenolics in the family Lamiaceae determined by ESR spectroscopy. Biochem. Syst. Ecol. 28, 229D253.

Pellerano, R. G., Uñates, M. A., Cantarelli, M. A., Camiña, J. M., & Marchevsky, E. J. (2012). Analysis of trace elements in multifloral Argentine honeys and their classification according to provenance. Food chemistry, 134(1), 578-582.

Peschel. W, F. Sanchez-Rabaneda, W. Dieckmann, A. Plescher, I. Gartzia, and D. Jimenez, (2006). An industrial approach in the search of natural antioxidants from vegetable and fruits wastes," Food Chemistry, vol. 97, pp. 137–150.

Pourmorad F, Hosseinimehr SJ, Shahabimajd N (2006). Antioxidant activity, phenol and flavonoid contents of some selected Iranian medicinal plants. Afr. J. Biotechnol., 5: (1142-1145).

Premkumar. P, K. Kannan* and M. Natesan, Thyme Extract of Thymus vulgar L. as Volatile Corrosion Inhibitor for Mild Steel in NaCl Environment Vol. 20, No. 1 (2008), (445-451)

Protect Your Family | Lead | US EPA. (2015, October 21). Retrieved from http://www2.epa.gov/lead/protect-your-family.

Quezel P, Santa S, Schotter O. Nouvelle flore de lAlgerie et des régions desertiques meridionales.Paris. 1962.

Rai V, Kakkar P, Khatoon S, Rawat AKS, Mehrotra S. Heavy metal accumulation in some herbal drugs. Pharm Biol 2001; 39:384-7.

Reddy, P. R. K.; Reddy, S. J. Chemosphere, 1997, 34,(22193-2212).

Reische D, Lillard D, Eintenmiller R. Antioxidants in food lipids. In: Ahoh C, Min D editors. Chemistry, Nutrition and Biotechnology. New York: Marcel Dekker; 1998. p. 423–48

Rice-Evans, C.A.; Miller, N.J.; Bolwell, P.G.; Bramley, P.M.; Pridham, J.B. The relative antioxidant activities of plantderived polyphenolic flavonoids. Free Radic. Res. 1995, 22, 375–283.

Rival, S.G., Boeriu, C.G. and Wichers, H.J., 2001. Caseins and Casein Hydrolysates. 2. Antioxidative Properties and Relevance to Lipoxygenase Inhibition, Journal of Agricultural and Food Chemistry, 49:295-302.

Rojas TR, Sampayo CAF, Vázquez BI, Franco CM, Cepeda A. Study of interferences by several metabolites from Aspergillus spp. in the detection of aflatoxigenic strains in media added with cyclodextrin. Food Control. 2005;16(5):445–50.

Roby, M. H. H., Sarhan, M. A., Selim, K. A. H., & Khalel, K. I. (2013). Evaluation of antioxidant activity, total phenols and phenolic compounds in thyme (Thymus vulgaris L.), sage (Salvia officinalis L.), and marjoram (Origanum majorana L.) extracts. *Industrial Crops and Products*, *43*, 827-831.

Safaei-Ghomi J, Ebrahimabadi AH, Djafari-Bidgoli Z, Batooli H (2009). GC/MS analysis and in vitro antioxidant activity of essential oil and methanol. Extracts of Thymus caramanicus Jalas and its main constituent carvacrol Food Chem., 115: 1524-1528

Safaei-Ghomi J, Ebrahimabadi AH, Djafari-Bidgoli Z, Batooli H (2009). GC/MS analysis and in vitro antioxidant activity of essential oil and methanol. Extracts of Thymus caramanicus Jalas and its main constituent carvacrol Food Chem., 115: 1524-1528.

Sattar A., Wahid M., Durrani S. K. Concentration of selected metals in spices, dry fruits and plant nuts. Plant Food. Hum. Nutr. 39, 279, 1989.

Schumacher M, Bosque MA, Domingo JL, Corbella J (1991) Dietary intake of lead and cadmium from foods in Tarrangona Province, Spain. Bulletin of Environmental Contaminant and Toxicology 46, 320-328

Schwart MK (1975) Role of trace elements in cancer. Cancer Research 35, 3481-3484.

Senyuva HZ, Gilbert J, Silici S, Charlton A, Dal C, Gürel N, Cimen D (2009) Profiling Turkish honeys to determine authenticity using physical and chemical characteristics. J Agric Food Chem 57: 3911–3919

Schwille, P., Haupts, U., Maiti, S., and Webb. W.(1999). Molecular dynamics in living cells observed by fluorescence correlation spectroscopy with one- and two-photon excitation. Biophysical Journal, 77(10):2251-2265.

Serbula, S. M., Kalinovic, T. S., Ilic, A. A., Kalinovic, J. V., & Steharnik, M. M. (2013). Assessment of airborne heavy metal pollution using Pinus spp. and Tilia spp. Aerosol Air Qual Res, 13(2), 563-573.

Sharma OP, Bhat TK (2009). DPPH antioxidant assay revisited. Food Chem., 113: 1202-1205.

Sharma KR, Agrawal M, Marshall MF (2009). Heavy metals in vegetables collected from production and market sites of a tropical urban area of India. Food Chem. Toxicol. 47:583-591.

Shi J, Gong J, Liu J, Wu X, Zhang Y. Antioxidant capacity of extract from edible flowers of Prunus mume in China and its active components. LWT-Food Sci Tech. 2009; 42(2):477–82.

Shirley, B.W. (1996) Flavonoid biosynthesis: "new" functions for an "old" pathway. Trends in Plant Science, 1, 377-382.

Slinkard, K. ve Singleton, V.L. (1977). “Total phenol analyses: Automation and Comparison with Manual Methods”, American Journal of Enology and Viticulture, 28, 49-55.

Sofowora A (1993) Medicinal Plants and Traditional Medicine in Africa (2nd Edn), Spectrum Books Ltd., Sunshine House, Ibadan pp 1-257

Somer E (1983). The toxic potential of trace metals in foods: A review. J. Food Sci. 39:215-217.

Sukumar K, Perich MJ, Boobar LR. Botanical derivatives in mosquito control: a review. J Am Mosq Control Assoc. 1991; 7(2):210–37.

Syamasri, P., Sharma, A. and Talukder, G. 1994. Effects of cobalt on plants. The Botanical Review 60(2):149-181

Takeuchi H, Lu ZG, Yu O (2004). Distinct crucial roles of flavonoids

Tevfik Ozena, Ibrahim Demirtasb,*, Huseyin Aksit Determination of antioxidant activities of various extracts and essential oil compositions of Thymus praecox subsp. skorpilii var. skorpilii Food Chemistry 124 (2011) 58–64)

Trease GE, Evans WC (1989) A Textbook of Pharmacognosy, pp 166, 469-471.

Trenary K. (1997), Visionary cactus Guide, Opuntia Sic, Retrieved 2007.

Treutter, D. (2006) Significance of flavonoids in plant resistance: a review, Environmental Chemistry Letters, 4 (3): 147-157.

Vartika R, Poonam K, Rawat AKS, Shantan M (2001) Heavy metal accumulation in some herbal drugs. Pharmaceutical Biology 39, 384-387.

Velioglu YS, Mazza G, Gao L, Oomah BD. (1998) Antioxidant activity and total phenolics in selected fruits, vegetables, and grain products. J. Agric. Food Chem. 46: 4113-4117.

Villãno D. Villãno, M.S. Fernández-Pachón, M.L. Moyá, A.M. Troncoso, M.C. García-Parrilla. (2007). Radical scavenging ability of polyphenolic compounds towards DPPH free radical. Talanta, 71, 221-230

VinsonJA, HaoY, SuX, Zubik L (1998) Phenolantioxidantquantityand quality in foods: vegetables. J Agric Food Chem 46:3630–3634

Viuda-Martos M, Ruiz-Navajas Y, Fernández-LópezJ,Pérez-Álvarez JA (2008) Functional properties of honey, propolis, and royal jelly. J Food Sci 73:117–124

Wallace DC (1999). Mitochondrial diseases in man and mouse, Science. 283:1482-1488.

Wendy Griswold, Ph.D., is a publications editor and CHSR’s expert in adult education and outreach evaluation methods and has worked with the Center since 1995 (griswold@ksu.edu).

World Health Organization (1991). Report on the intercountry expert meeting of traditional medicine and primary health care. WHO-EMTRM/1-E/l1292/168 30 November- 3 December 1991, Cairo, Egypt.

Wong MK, Tan P, Wee YC (1993). Heavy metals in some of Chinese herbal plants. Biol. Trace Elem. Res. 36:135-142.

World Health Organization, (1998) “Determination of Arsenic and Heavy Metals”. In Quality Control Methods for Medicinal Plant Material, Genova, p. 62-63.

Yanishlieva, N. V., Marinova, E. M., Gordonb, M. H. and Ranevaa, V. G. (1999). Antioxidant activity and mechanism of action of thymol and carvacrol in two lipid systems. Food Chemistry, 64, 59-66.

Yanishlivea, N. V., Marinova, E., Pokorny, J. (2006). Natural antioxidants from herbs and spice. Eur. J. Lipid Sci. Technol. 108; 776-793

Yoong K. Soon (1998) “Determination of Cd, Cr, Co, Pb, and Ni in Plant Tissue” in Handbook of Reference Methods for Plant Analysis (Y. P. Kalra ed) CRC Press, pp. 193-198.

Young IS, Woodside JV., (2001). Antioxidants in health and disease. J. Clin. Pathol. 54: 176-186

Zanda Kruma1, Mirjana Andjelkovic 2, Roland Verhe 2, Viesturs Kreicbergs (2008). Phenolic Compounds In Basil, Oregano And Thyme Foodbalt

Zheljazkov, V.D., and Fair, P., (1996) “Study of the Effect of Highly Heavy Metal Polluted Soils on Metal Uptake and Distribution in Plants from Genera Artemisia, Draccocephalum, Inula, Ruta and Symphytum” Acta Horticultura, 426: (397-417).

Zhishen J, Mengcheng T and Jianming W. (1999). The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chemistry, 64*, (555-559).

Zou Y, Lu Y and Wei D. (2004) Antioxidant activity of flavonoid-rich extract of Hypericum perforatum L in vitro. *Journal of Agriculture and Food Chemistry, 52*, (5032-5039).

APPENDICES

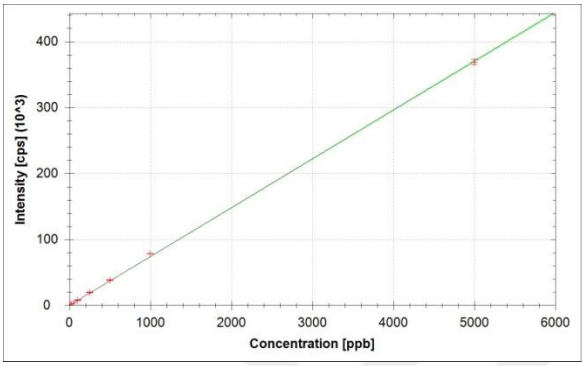
Appendix 1. Calibration charts of metal analysis

Calibration Curves:

Instrument Nam: iCAP Q

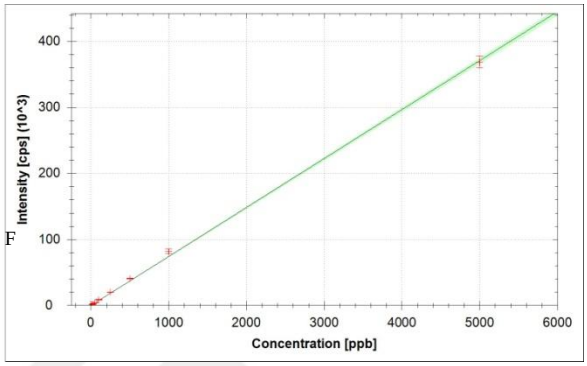
Serial Number: Undefined

$_{7}$Li



f(x) = 74.0467*x
R_2 = 0.9998
BEC = 0.000 ppb
LOD = 0.0000 ppb

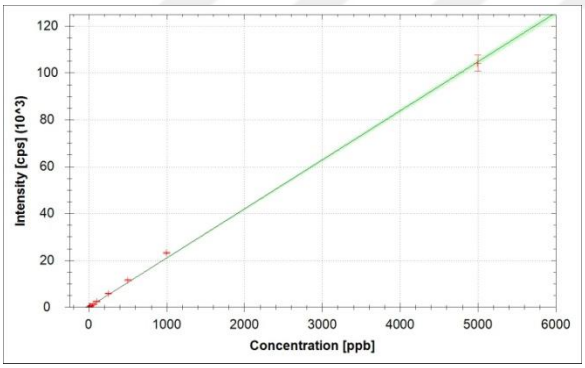
$_{9}$Be



F

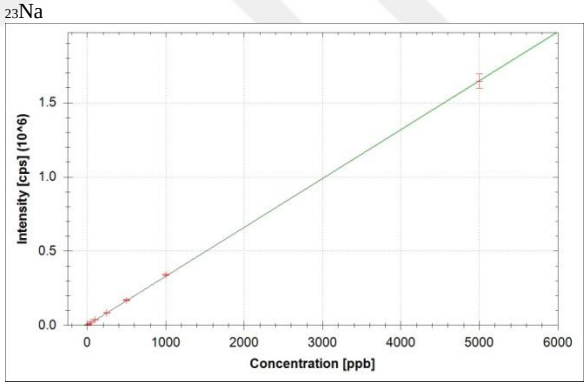
(x) = 74.1049*x + 3.3333
R_2 = 0.9993
BEC = 0.045 ppb
LOD = 0.2337 ppb

$_{11}$B



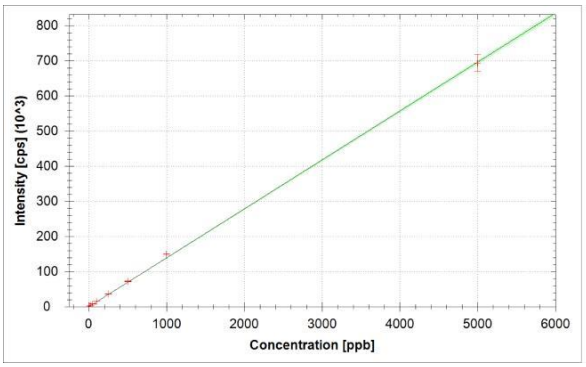
f(x) = 20.9200*x + 103.3341
R_2 = 0.9993
BEC = 4.939 ppb
LOD = 3.6089 ppb

$_{23}$Na



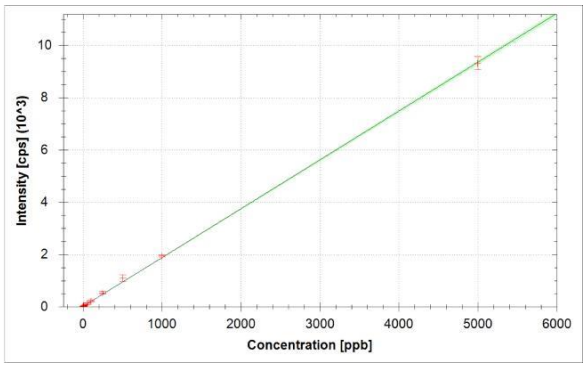
f(x) = 329.3396*x + 580.0151
R_2 = 0.9999
BEC = 1.761 ppb

$_{24}$Mg



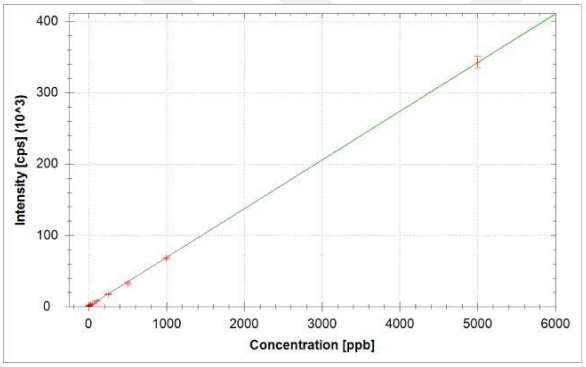
f(x) = 139.2022*x + 6.6667
R^2 = 0.9996
BEC = 0.048 ppb
LOD = 0.2489 ppb

$_{31}$P



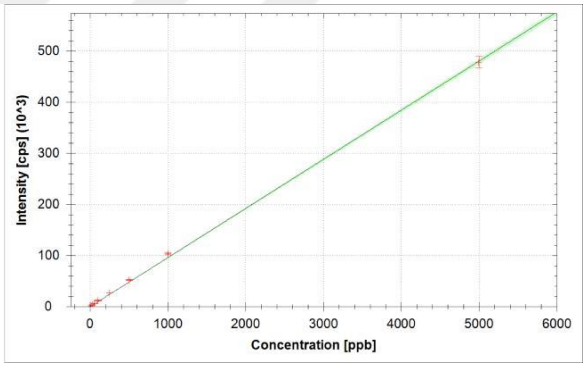
f(x) = 1.8676*x + 13.3334
R^2 = 0.9996
BEC = 7.139 ppb
LOD = 9.2745 ppb

$_{39}$K



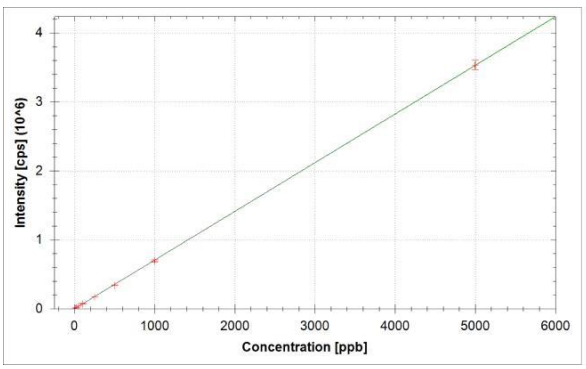
f(x) = 68.2591*x + 753.3588
R^2 = 0.9999
BEC = 11.037 ppb
LOD = 4.2690 ppb

$_{44}$Ca



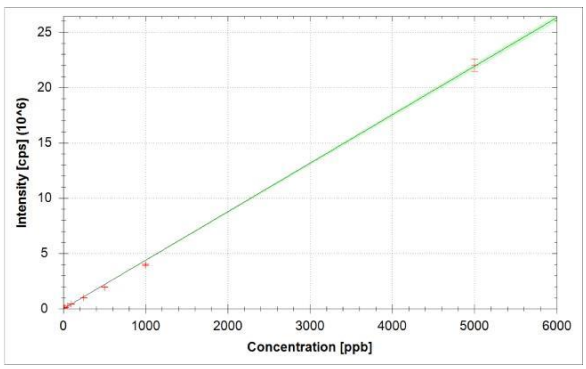
f(x) = 95.9947*x + 26.6669
R^2 = 0.9995
BEC = 0.278 ppb
LOD = 0.3609 ppb

$_{48}$Ti



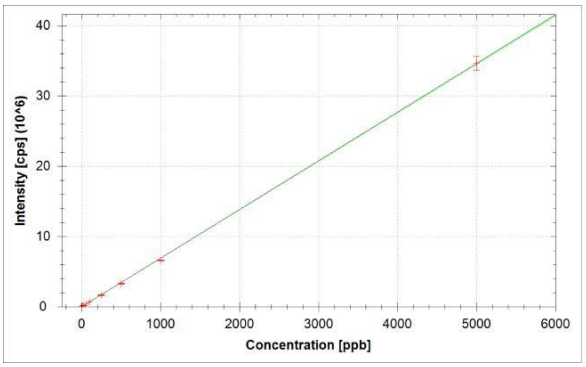
f(x) = 705.8034*x + 3.3333
R^2 = 1.0000
BEC = 0.005 ppb
LOD = 0.0245 ppb

$_{51}$V



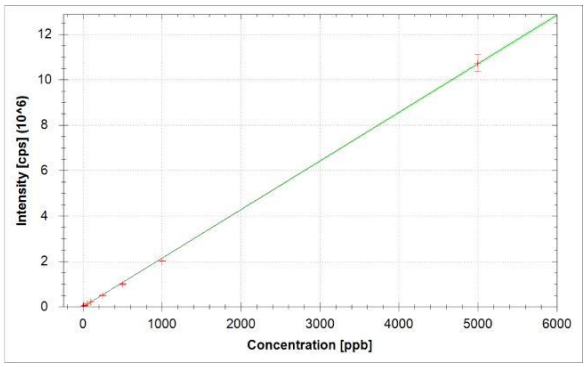
f(x) = 4383.3481*x
R^2 = 0.9994
BEC = 0.000 ppb
LOD = 0.0000 ppb

^{52}Cr



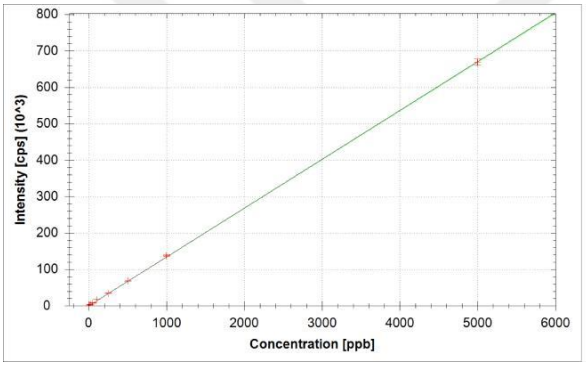
f(x) = 6917.2912*x + 26.6668
R^2 = 0.9998
BEC = 0.004 ppb
LOD = 0.0025 ppb

^{55}Mn



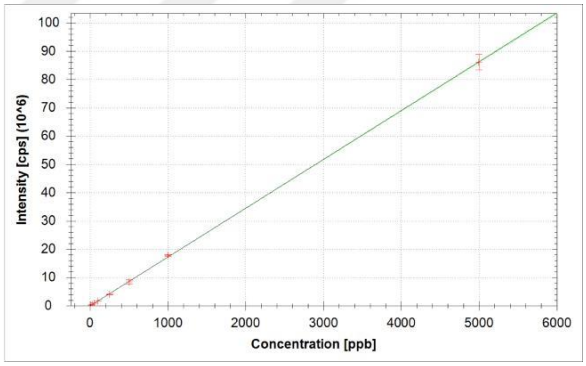
f(x) = 2139.0694*x + 33.3335
R^2 = 0.9997
BEC = 0.016 ppb
LOD = 0.0081 ppb

^{57}Fe



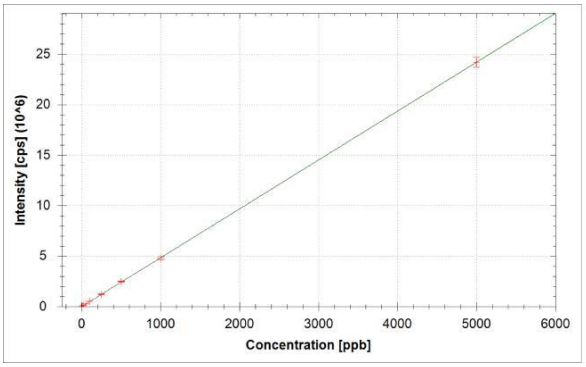
f(x) = 134.0377*x + 10.0000
R^2 = 0.9999
BEC = 0.075 ppb
LOD = 0.0000 ppb

^{59}Co



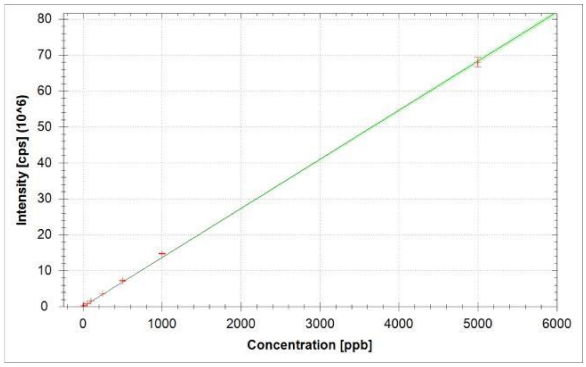
f(x) = 17246.2538*x + 113.3344
R^2 = 0.9999
BEC = 0.007 ppb
LOD = 0.0089 ppb

^{60}Ni



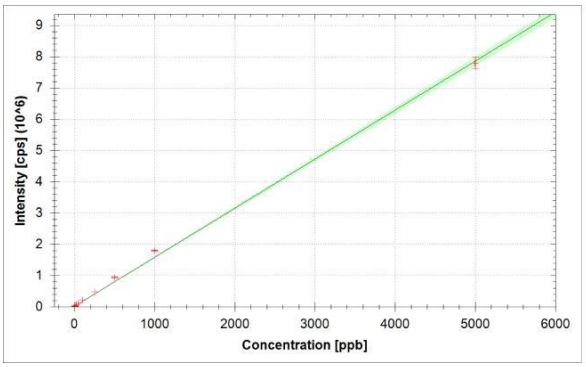
f(x) = 4838.3663*x + 20.0001
R^2 = 1.0000
BEC = 0.004 ppb
LOD = 0.0062 ppb

^{63}Cu



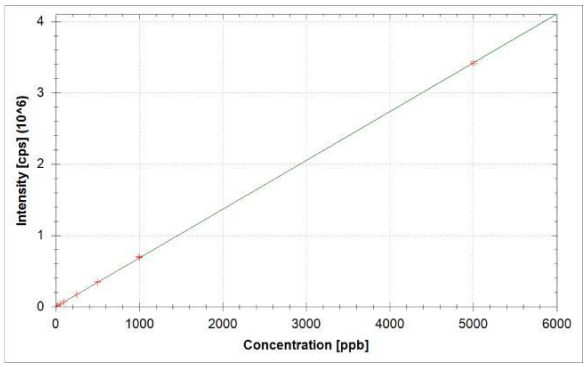
f(x) = 13652.8900*x + 116.6677
R^2 = 0.9996
BEC = 0.009 ppb
LOD = 0.0063 ppb

^{66}Zn



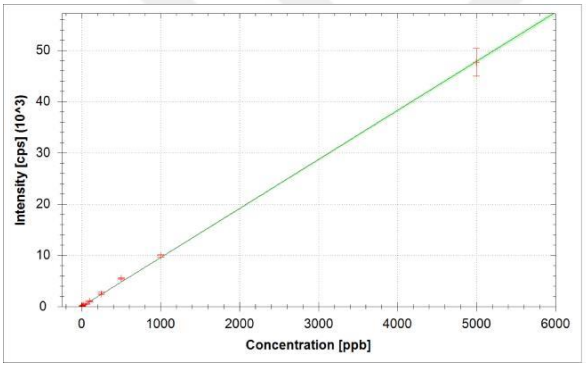
f(x) = 1568.0454*x + 15209.3923
R^2 = 0.9985
BEC = 9.700 ppb
LOD = 2.1074 ppb

^{75}As



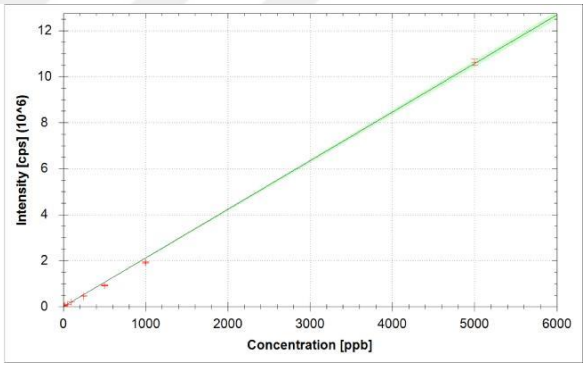
f(x) = 683.9898*x
R^2 = 1.0000
BEC = 0.000 ppb
LOD = 0.0000 ppb

^{77}Se



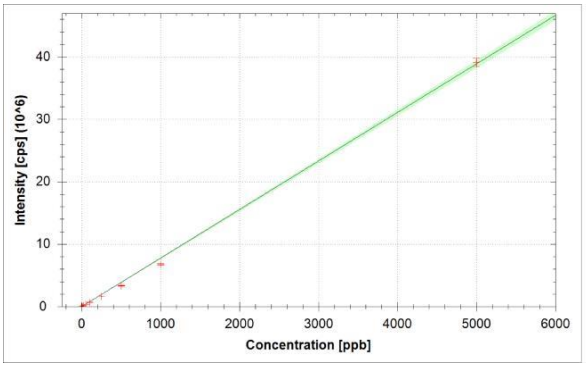
f(x) = 9.5640*x + 3.3333
R^2 = 0.9996
BEC = 0.349 ppb
LOD = 1.8110 ppb

^{88}Sr



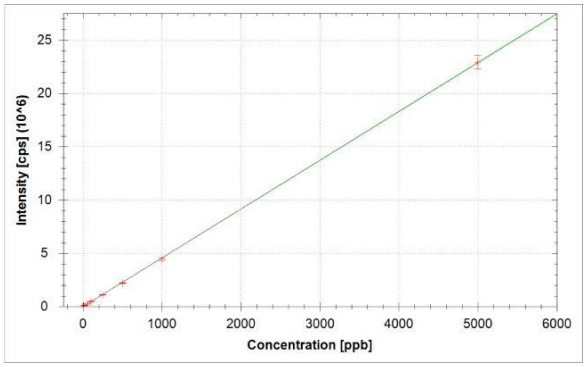
f(x) = 2112.9353*x
R^2 = 0.9993
BEC = 0.000 ppb
LOD = 0.0000 ppb

^{95}Mo



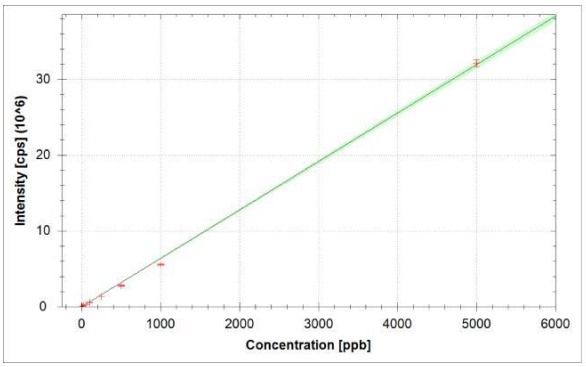
f(x) = 7768.8252*x + 80.0006
R^2 = 0.9988
BEC = 0.010 ppb
LOD = 0.0077 ppb

^{111}Cd



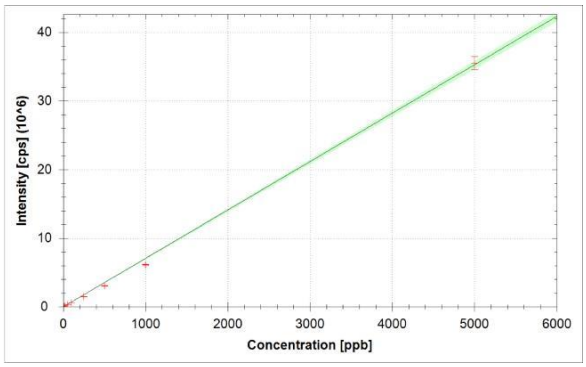
f(x) = 4577.4282*x + 33.3335
R^2 = 0.9999
BEC = 0.007 ppb
LOD = 0.0136 ppb

^{118}Sn



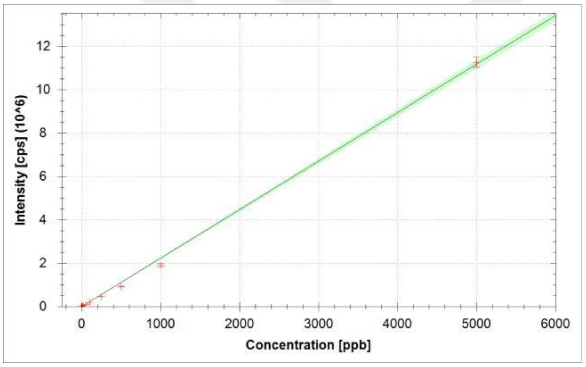
f(x) = 6383.6315*x + 76.6672
R^2 = 0.9989
BEC = 0.012 ppb
LOD = 0.0072 ppb

^{121}Sb



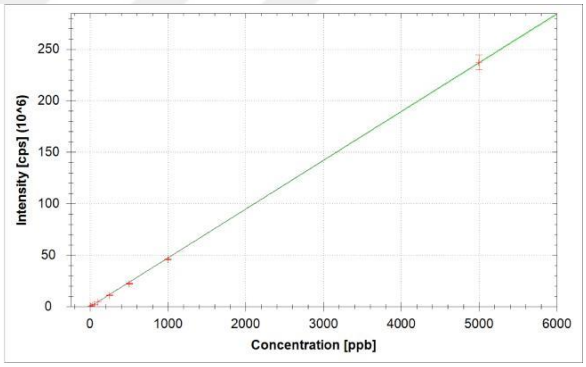
f(x) = 7055.1055*x
R^2 = 0.9988
BEC = 0.000 ppb
LOD = 0.0000 ppb

^{137}Ba



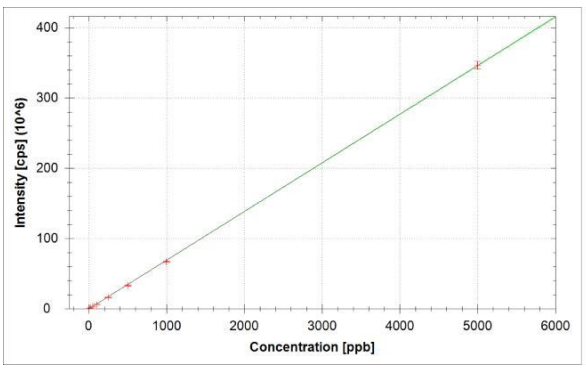
f(x) = 2235.0224*x + 13.3334
R^2 = 0.9985
BEC = 0.006 ppb
LOD = 0.0155 ppb

^{139}La



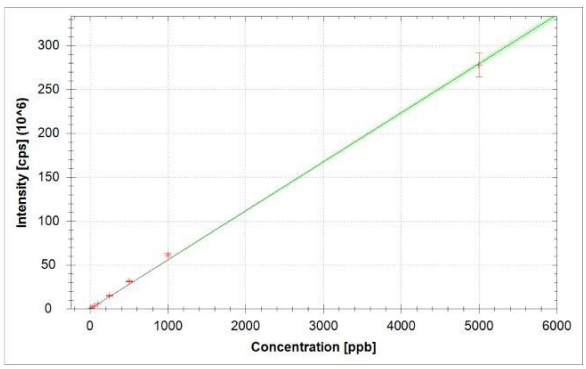
f(x) = 47360.5541*x + 10.0000
R^2 = 0.9999
BEC = 0.000 ppb
LOD = 0.0006 ppb

^{140}Ce



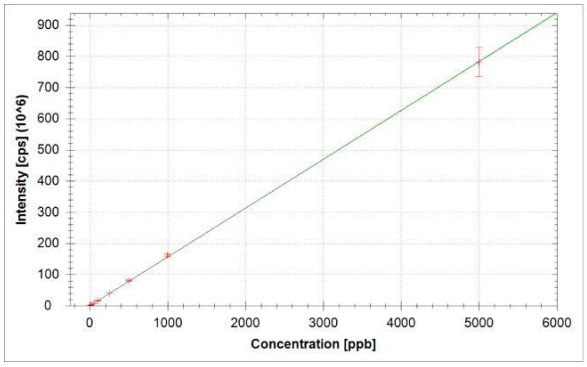
f(x) = 69223.9711*x + 13.3334
R^2 = 0.9999
BEC = 0.000 ppb
LOD = 0.0007 ppb

^{195}Pt



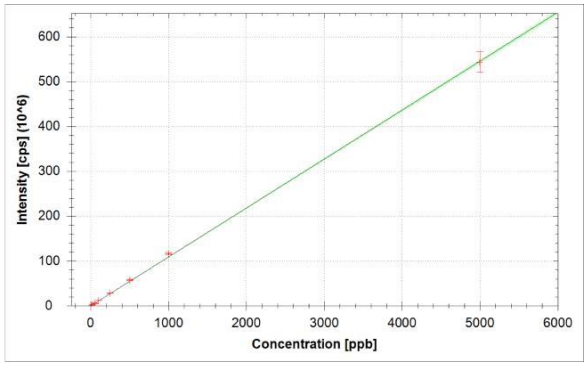
f(x) = 55859.6529*x + 40.0002
R^2 = 0.9993
BEC = 0.001 ppb
LOD = 0.0005 ppb

^{205}Tl



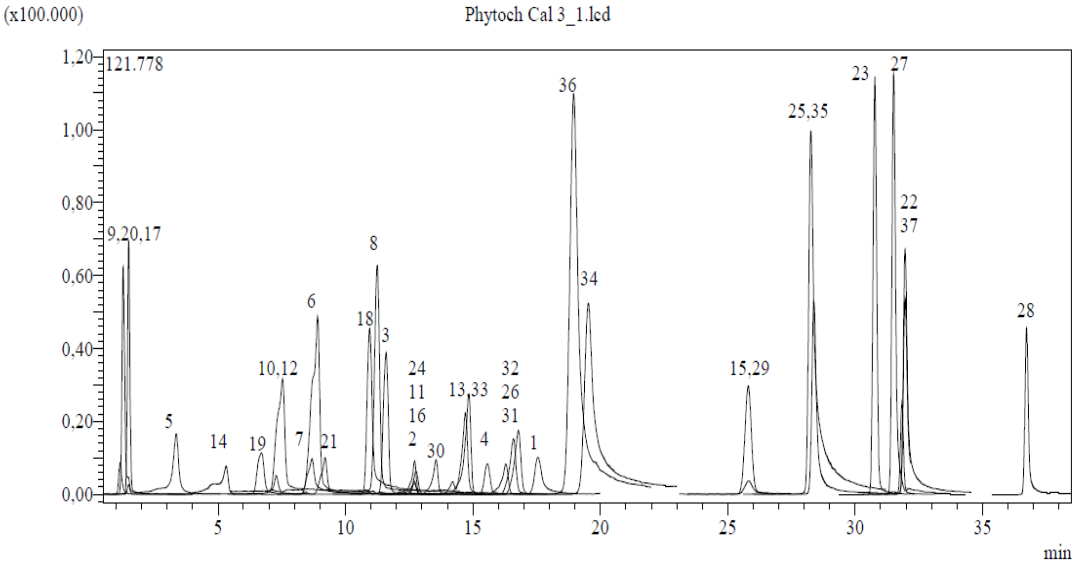
f(x) = 156622.0831*x + 20.0002
R_2 = 0.9999
BEC = 0.000 ppb
LOD = 0.0004 ppb

^{208}Pb

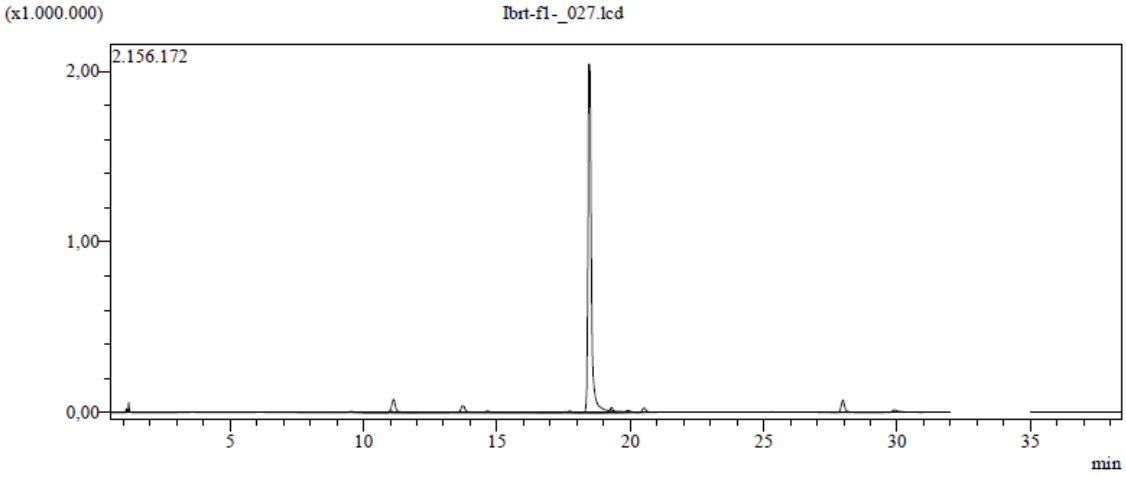


f(x) = 109032.9730*x + 2336.8958
R_2 = 0.9997
BEC = 0.021 ppb
LOD = 0.0055 ppb

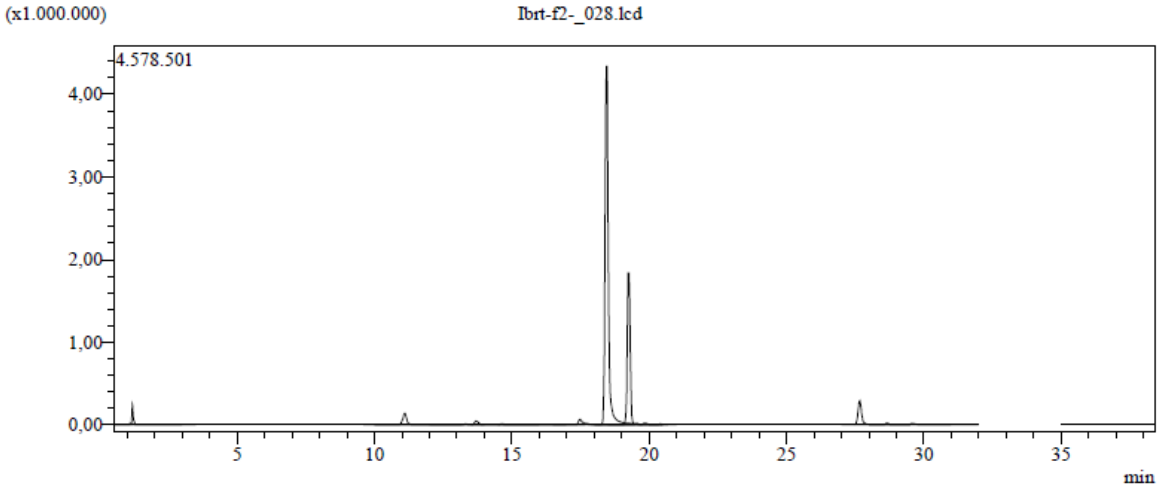
Appendix 2. Phenolic compound picks (1-13)



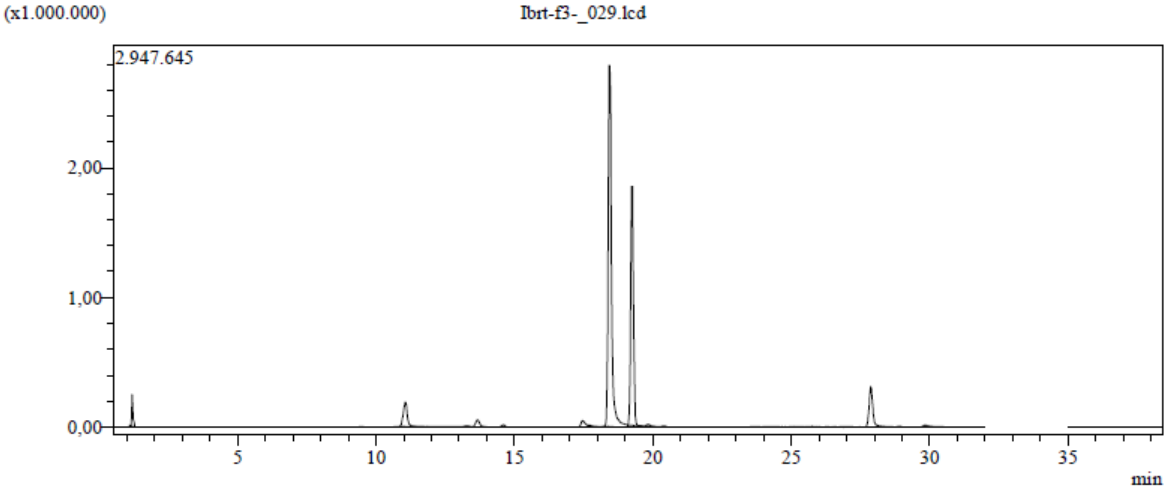
App. 2.1. The sdandart chromatograms of phenolic compound



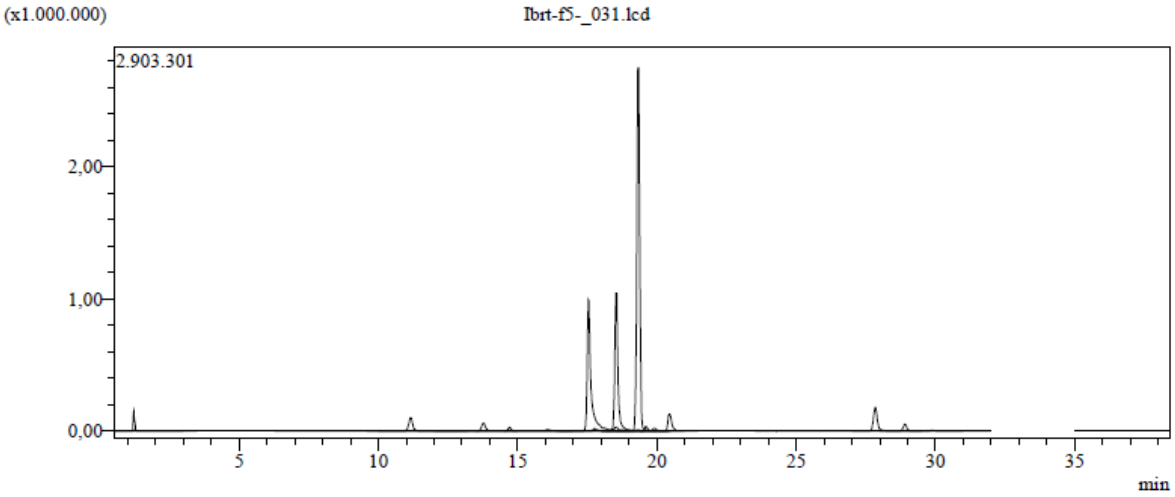
App. 2.2. The chromatograms of Qandil



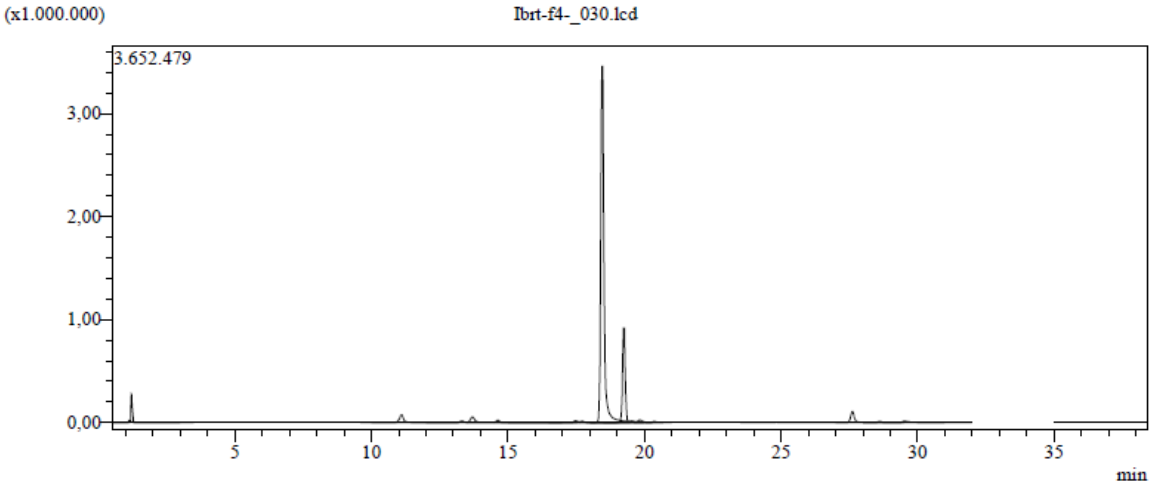
App. 2.3. The chromatograms of Asos



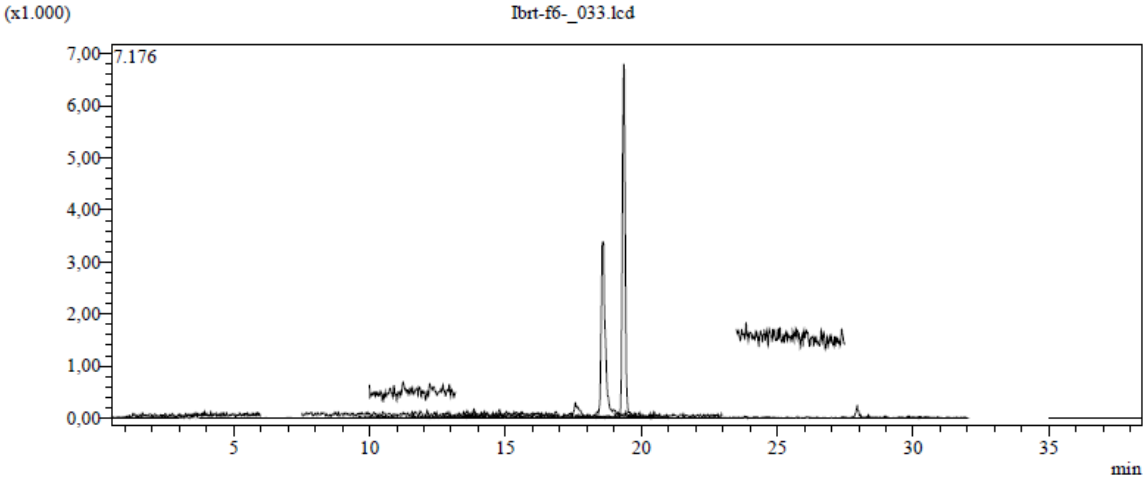
App. 2.4. The chromatograms of Balambo



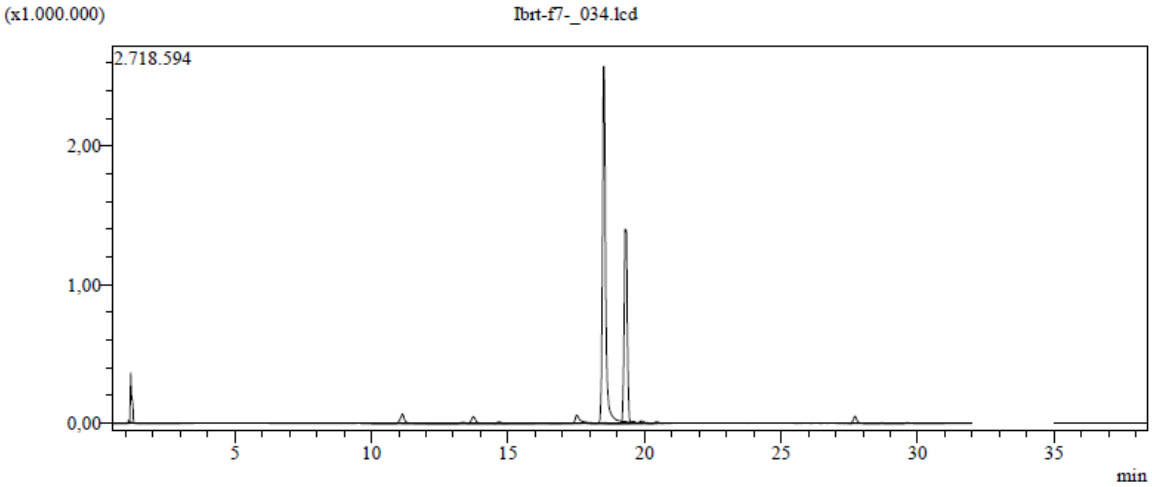
App. 2.5. The chromatograms of Eruh



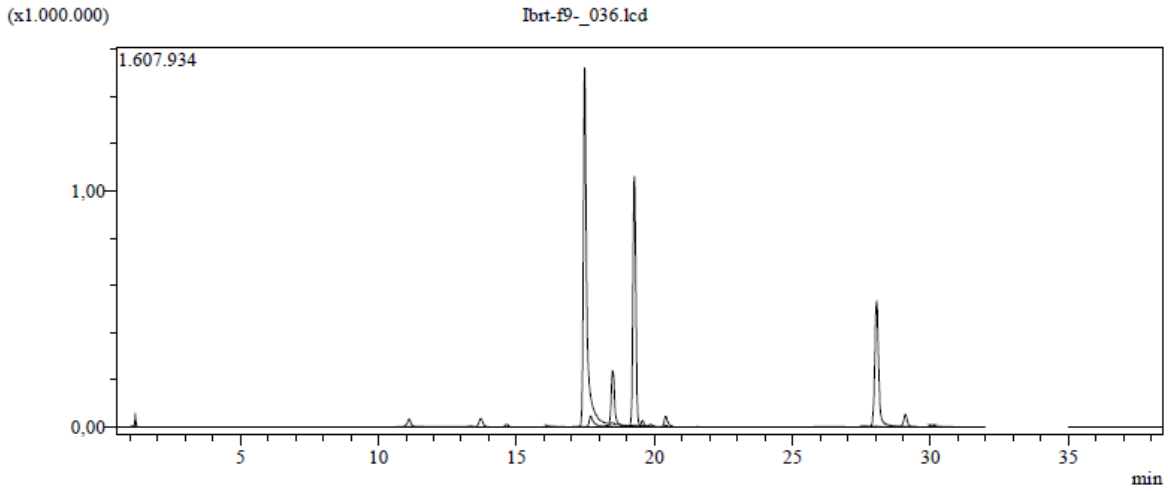
App. 2.6. The chromatograms of Karokh



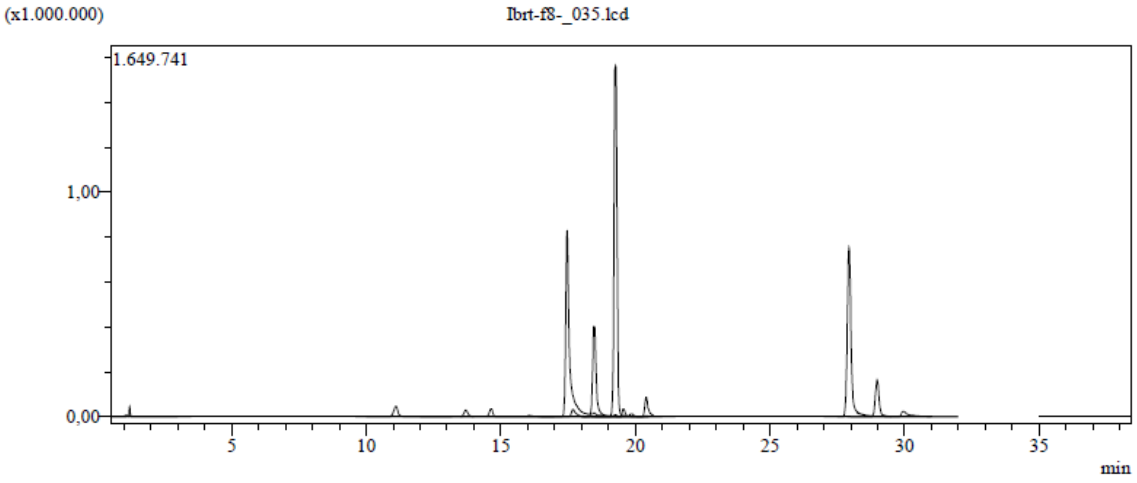
App. 2.7. The chromatograms of Şirvan yolu



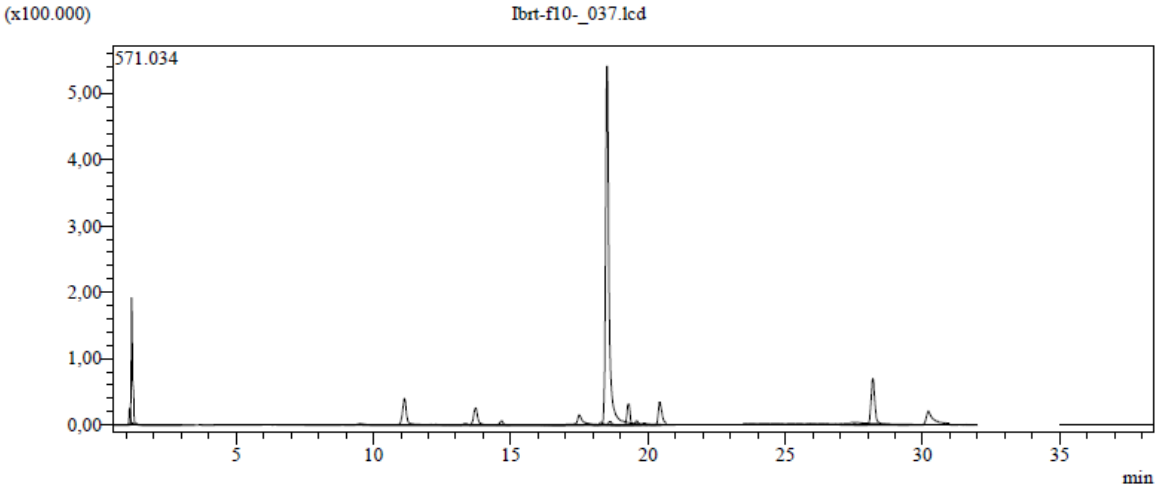
App. 2.8. The chromatograms of kampus



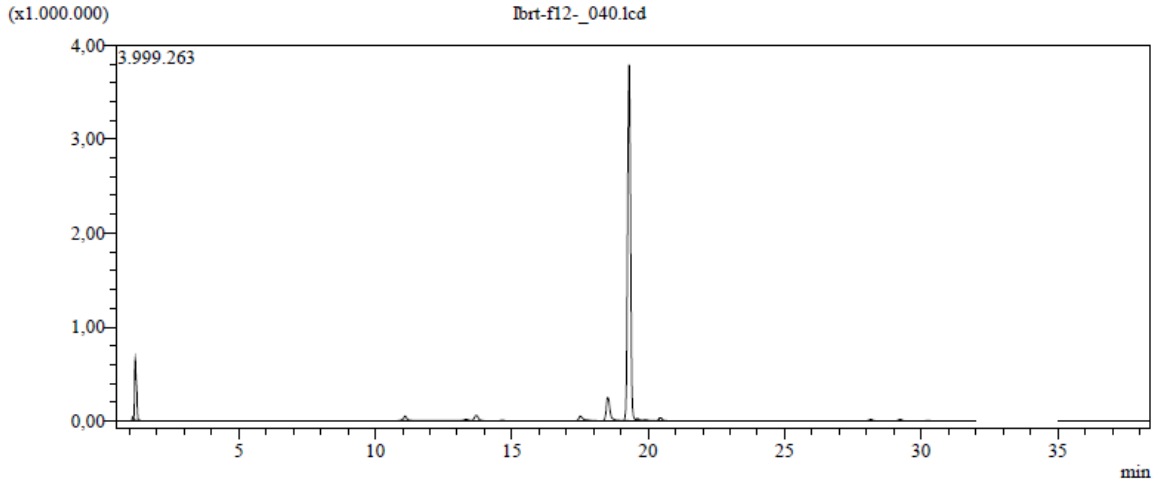
App. 2.9. The chromatograms of Koyu(Akdagmus)



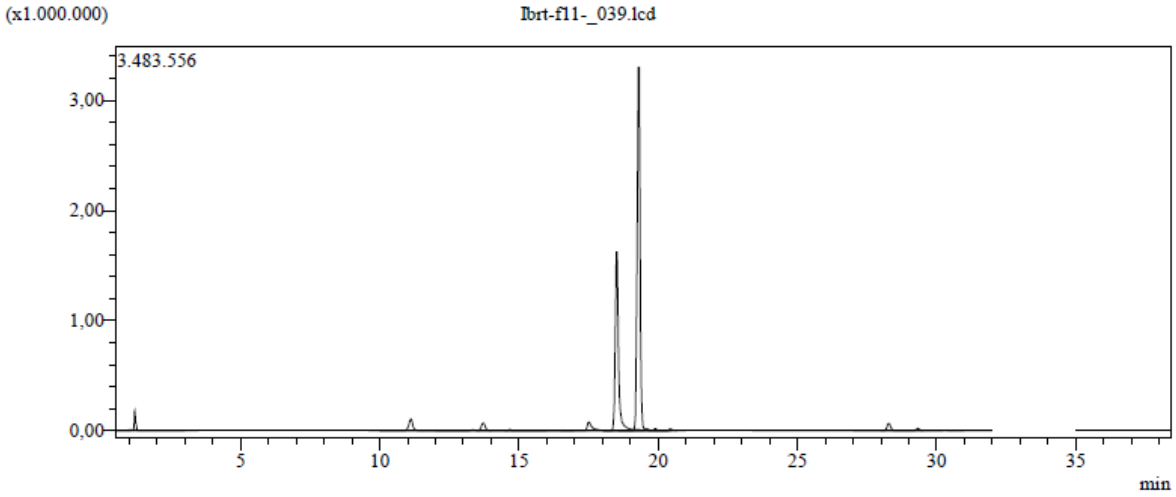
App. 2.10. The chromatograms of Tillo



App. 2.11. The chromatograms of Kurtalan(uyol)



App. 2.12. The chromatograms of Baykan (Girisi)



App. 2.13. The chromatograms of Kurtalan(zokayt,selala)

CURRICULUM-VITAE

PERSONAL INFORMATION

Name Surname	Karwan Mohammad Amin Mahmood
birth place and date	7.8.1987 sulaimany/ranya
Telephone	0096 750 1583435
E-mail	mahamad.karwan@yahoo.com

EDUCATİON

Degree		**Adı, İlçe, İl**	**Bitirme Yılı**
High school	:	Arasn , ranya, sulaimany, Iraq	**2007**
Üniversite	:	Sulaimany, University college of science	**2011**

WORK EXPERIENCES

Year	**institution**	**Duty**
2011	Hospital	Labortory
2012	High school	Teacher
2013	The independent high Electoral commission	Employer
2014	The independent high Electoral commission	Manger

SPECIALTY AREA: -

FOREIGN LANGUAGES: Turkish, Arabic, English, Kurdish

OTHER FEATURES YOU WANT TO SPECIFY: