

Original article

Using GC –Mass Method for Determination the Hydrocarbon Compounds in Some Vegetable Samples at Derna City, Libya

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Vegetables, Aromatic.**ABSTRACT**

The purpose of this study was to identify and quantify the polycyclic aromatic hydrocarbons (PAHs) in a variety of vegetables from various places in Derna City, Libya. The GC-MS equipment was used to identify and determine the types of aromatic hydrocarbon molecules. The findings demonstrated that polycyclic aromatic hydrocarbons were present in every vegetable sample that was analyzed. The findings indicated that the vegetables' total amounts of polycyclic aromatic hydrocarbons varied as follows: 8.257 µg/g for tomato, 8.258 µg/g for pepper, 7.841 µg/g for eggplant, and 8.216 µg/g for zucchini. The origin and sources of the investigated polycyclic aromatic hydrocarbons in vegetable samples were estimated using a few computed ratios, as the ratio (LMW/HMW) of low molecular weight (LMW) to high molecular weight (HMW) chemical composition. Diagnostic ratios of the concentrations of specific pairs of aromatic hydrocarbons, such as (Σ COMB/ Σ PAHs), Phe/Ant, Flu/Pyr, Inp/(Inp + Bghip), Bap/(Bap + Chr), and Ant/(Ant + Phe), were also employed to investigate the origin of the polycyclic aromatic hydrocarbons. The findings showed that petrogenic and pyrolytic sources were the primary sources of PAHs in vegetables. The investigation came to the conclusion that the majority of the PAHs in the study samples came from either pyrolytic or direct petroleum sources.

Introduction

Because of its fertile soil, temperate climate, and labor, Derna is an agriculturally oriented region that is heavily dependent on growing vegetables. This is especially true in some areas, like Wadi Derna and El-fatiah, where over half of households reported being involved in agricultural activities [1]. The size of this plant is 31,511 square kilometers, and it grows in two seasons: winter and summer. It is situated in the northeastern region of Libya, where the average domestic production of vegetables is estimated to be 1550,000 tons, while Libyan agricultural fields make up less than 3% of the country's total territory. Derna's primary crops are vegetables such as tomatoes, cucumbers, peppers, beans, peas, onions, lettuce, cilantro, and parsley [2]. Naturally, vegetables are considered the most vital component for human health and protection; they are essential for balanced meals because they contain dietary fiber, antioxidants, minerals (such as Fe, K, Mg, Zn, Ca, etc.), nitric oxide, and dietary fiber. Consuming vegetables on a daily basis has been closely associated with better gastrointestinal health, clear vision, and a lower risk of heart disease, stroke, and chronic conditions like diabetes, obesity, and some types of cancer. Furthermore, vegetables make up a sizable amount of the global human diet and are essential for human nutrition, especially as sources of phytonutriceuticals, which include vitamins A, B1, B6, B9, C, and E, as well as carbohydrates, protein, and phytochemicals like flavonoids, phenolic compounds, carotenoids, and bioactive peptides. Vegetables come in a variety of species and might be edible seeds, fruits, roots, or leaves. Every species has a unique contribution to the food [4]. However, the environment, which is heavily contaminated, has an impact on the veggies. The concentration of hazardous gases and compounds has increased to unsafe levels due to urbanization, industry, and vehicle pollution, harming people, animals, and plants [5]. Among the most significant and hazardous pollutants found in soil are polycyclic aromatic hydrocarbons (PAHs), which can spread to crops and damage the ecosystem. PAHs are found in soil, water, and the atmosphere [6]. The type of PAH compounds, their physicochemical characteristics, and the physiological state of the plant all have a direct impact on how well they are absorbed by vegetables, which can do so by foliar or root uptake. While high molecular mass PAHs can contaminate the surface by air fallout, low molecular mass PAHs can be absorbed by surface adsorption in plants. A potential route from polluted soil to plant roots was identified by the direct correlation between soil and plant PAH contents [7]. Both direct contact with soil or ingestion of it, as well as indirect exposure through the eating of crops and grazing animal products, can expose

humans to soil pollution. There are three ways that the latter could occur: (1) ingestion (deliberate or unintentional, geophagy); (2) inhalation; and (3) penetration or absorption through the skin. One potentially significant route of exposure to environmental contaminants may be through the ingestion of dust and soil particles. Hand-to-mouth contact, which is very common in youngsters, airborne dust, and improperly cleaned fruits and vegetables can all result in accidental consumption. Moreover, the main cause of animal product contamination (meat, milk, or eggs) may be farm animals kept outdoors, accidentally consuming soil containing POP. According to recent studies, soil poses a serious risk matrix for POPs entering the food chain, especially in regions with high contamination levels [8]. This study aims to determine the types and contents of polyhydrocarbon compounds in different vegetables collected from Derna city, Libya.

Methods

Vegetable Samples

Between March and June 2023, the vegetable samples were gathered at random from a number of farms. About 2 kg of samples of each of the four vegetable species were collected; the examined samples are displayed in (Table 1). Samples of vegetables were brought to the lab in plastic bags for protection. After being cleaned with tap water and deionized water, fresh samples were allowed to air dry before being meticulously weighed. The samples were cut into little pieces and then dried. Then dried once more in an oven set at 75 °C for 36 hours. To obtain a representative sample, it was then ground into powder using a food processor (blender) and sieved through a 2 mm nylon sieve. and the sample was moved into specimen cans in the proper volume.

Table 1. The studied Vegetable samples

s. no.	Vegetables	Species	Sites
1	Tomato	Fruiting	El-Fatihah 1
2	Zucchini	Fruiting	Sidi khalid 2
3	Pepper	Fruiting	El-Fatihah 2
4	Eggplant	Fruiting	Ain mara

Polycyclic Aromatic Hydrocarbons Analysis

Analysis of PAHs was conducted using the methods provided by some authors [9 -12].GC/MS was used to examine the polycyclic aromatic hydrocarbons (PAHs) components. Each chemical in the vegetable extract was identified by comparing its chromatogram and retention durations with those from HP memory and the EPA standard at the Marine Science Institute's central lab (Alexandria, Egypt). Different procedures, such as extraction, cleaning, and fractionation, instrumental analysis, and analytical quality control, were used to examine the samples for PAHs. Excel was used to tabulate and statistically analyze the collected data.

Extraction Step of PAHs for Vegetables Sample

30 g of anhydrous sodium sulfate was added to 0.5 g of agricultural soil and 1 g of the wet weight of each vegetable species. The mixture was then mixed for five minutes at high speed. After that, the mixture was extracted for eight hours using a Soxhlet and 200 milliliters of methanol. To saponify the lipids, 20 milliliters of 0.7 M KOH and 30 milliliters of distilled water were added to the flask, and the reflux was maintained for two hours. Using 80 milliliters of hexane, the contents of the extraction flask were extracted three times in a separating funnel. Anhydrous sodium sulfate was used to dry the three extracts before they were filtered through glass wool. Prior to cleaning and fractionation, the hexane fraction was concentrated using a rotary evaporator down to approximately 15 ml at 300 °C. It was subsequently concentrated to a volume of 1 ml using a nitrogen gas stream [9-12].

Instrumentation and Analysis Conditions

There are two components to the GC-MS instrument: The mass spectrometer (MS) detects and measures the chemicals in the sample after the gas chromatograph (GC) separates them. The sample is injected into the GC inlet after being sufficiently cleaned. There, it is vaporized and transported by an inert gas (mobile phase), usually helium, through a column that has a particular polymer coating on the inner surface (stationary phase). In addition to adsorption to the stationary phase, the molecules separate according to their volatility, with smaller molecules moving more quickly than larger ones. Retention time is the amount of time that passes after the molecule leaves the column. Since the molecules must be charged in order to pass through the filter (mass analyzer) in the mass spectrometer, it is important that the molecules are ionized at the end of the column. The filter separates the ions according to properties related to the mass-to-charge ratio (m/z), which varies depending on the analyzer. Throughout the investigation, the mass-to-

charge ratio (m/z) is computed. The filter separates the ions according to properties related to the mass-to-charge ratio (m/z), which varies depending on the analyzer. Throughout the investigation, the mass-to-charge ratio (m/z) is computed. The most popular mass filter is the quadrupole mass filter, which was used in this study. However, other filters, including sector field (SF) and time of flight (ToF), are also employed. Following their separation, the ions are sent through a detector that determines how many ions of a particular mass there are. A computer receives this data, records it all, and shows the mass spectrum [13].

Calculated ratios

R1, R2, R3, R4, R5, and R6 are some of the ratios that are used to identify the sources of aromatic compounds. These include the following:

$$R1 = \text{Inp} / (\text{Inp} + \text{Bghip})$$

$$R2 = \text{Flu} / (\text{Flu} + \text{Pyr})$$

$$R3 = \text{Bap} / (\text{Bap} + \text{Chr})$$

$$R4 = \text{Phe} / (\text{Phe} + \text{Ant})$$

$$R5 = \text{BbF} / \text{BKF}$$

$$R6 = \text{Ant} / (\text{Ant} + \text{Phe})$$

An indication of the origin of the aromatic hydrocarbons found in the samples can be obtained by computing these ratios. The kind and source of the hydrocarbon compounds are estimated using the values of these ratios as follows:

If R1 is less than 0.4, it indicates petroleum contamination; if R1 is greater than 0.5, it indicates hydrocarbon pollution from burning grass and wood [14]. Petroleum-derived hydrocarbon particles are present if the R2 ratio is within the range of 0.4; coal and combusted fuel are indicated if the R2 ratio is more than 0.4. Although the aromatic components are products of petroleum combustion, if the R3 is less than 0.2, uncombusted petroleum pollution is indicated; if the R3 is between 0.4 and 0.6. Additionally, if the $R4 < 0.1$ ratio is a sign of aromatic chemicals that come from a petroleum chemical source. On the other hand, $R5 > 0.1$ indicates that fuel has burned. Additionally, diesel, core oil, and coal are present if $R6 > 0.1$.

Ratio of Low Molecular Weight / High Molecular Weight of hydrocarbons

LMW = Fluorene + Phenanthrene + Anthracene + Naphthalene + Acenaphthylene + Acenaphthene
 HMW = BbF+ BKF + BaP + DBA + B (ghi) perylene + indenol pyrene + fluoranthene + pyrene + BaA+ chrysene

Results

The Polycyclic Aromatic Hydrocarbons (PAHs): contents of the vegetable samples under investigation were given in (Table 2). Furthermore, (Figures 1– 4) show the chromatograms of the compounds.

Table 2. GC/MS measurements of the polyaromatic hydrocarbon concentrations ($\mu\text{g/g}$) in the fruiting vegetable samples

PAHs Compound	Vegetable Samples				Average
	Tomato	Pepper	Eggplant	Zucchini	
	EL-fatiah 1	EL-fatiah 2	Ain mara	Sidi Khalid 1	
Acenaphthylene	0.327	0.327	0.326	0.326	0.327
Anthracene	0.429	0.430	0.424	0.426	0.427
Phenanthrene	0.556	0.557	0.531	0.538	0.545
Fluorene	0.315	0.318	0.309	0.313	0.313
Pyrene	0.115	0.113	0.112	0.113	0.114
Chrysene	0.508	0.499	0.498	0.498	0.500
Benzo(a) anthracene	0.716	0.725	0.716	0.716	0.718
Benzo(a) Pyrene	0.702	0.699	0.699	0.702	0.702
Benzo(b) fluoranthene	0.364	0.364	0.364	0.364	0.364
Benzo(k) fluoranthene	0.357	0.357	0.357	0.357	0.357
Indeno(1,2,3cd) pyrene	3.663	3.664	3.664	3.664	3.664
Dibenzo(a,b) anthracene	0.205	0.205	0.205	0.205	0.205
Benzo(g,h,i) perylene	N/A	N/A	N/A	N/A	0
	8.257	8.258	7.841	8.216	8.236

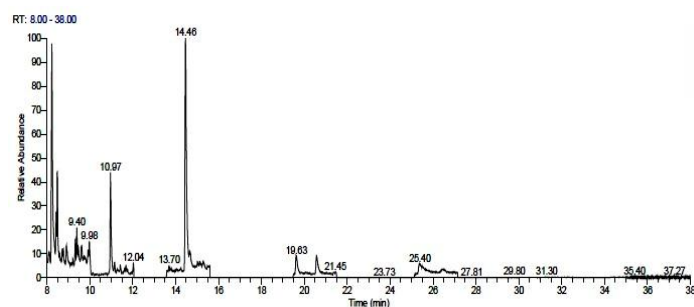


Figure 1. GC-MS chromatogram of PAH compounds fraction of tomato sample at EL-fatiah-1 location

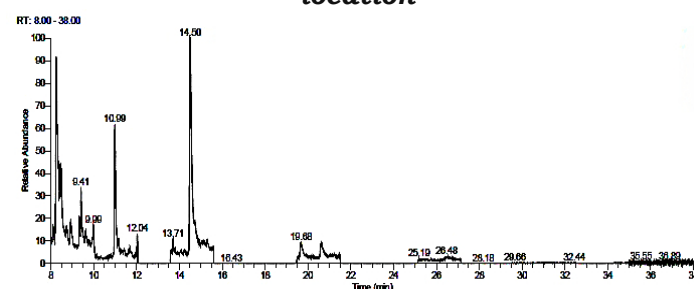


Figure 2. GC-MS chromatogram of PAH compounds fraction of pepper sample at EL-fatiah-2 location

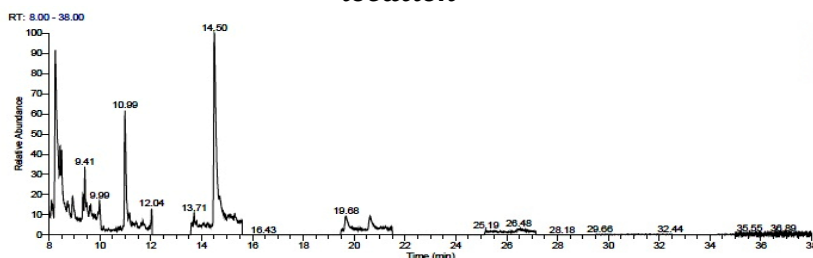


Figure 3. GC-MS chromatogram of PAH compounds fraction of eggplant sample at Ain Mara location

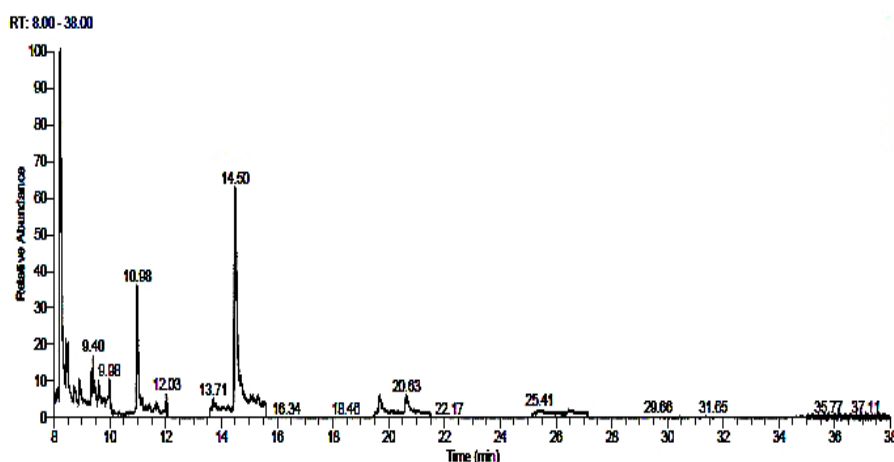


Figure 4. GC-MS chromatogram of PAH compounds fraction of zucchini sample at Sidi Khalid-1 location

Additionally, (Figure 5) showed the PAH values in the fruiting vegetable samples.

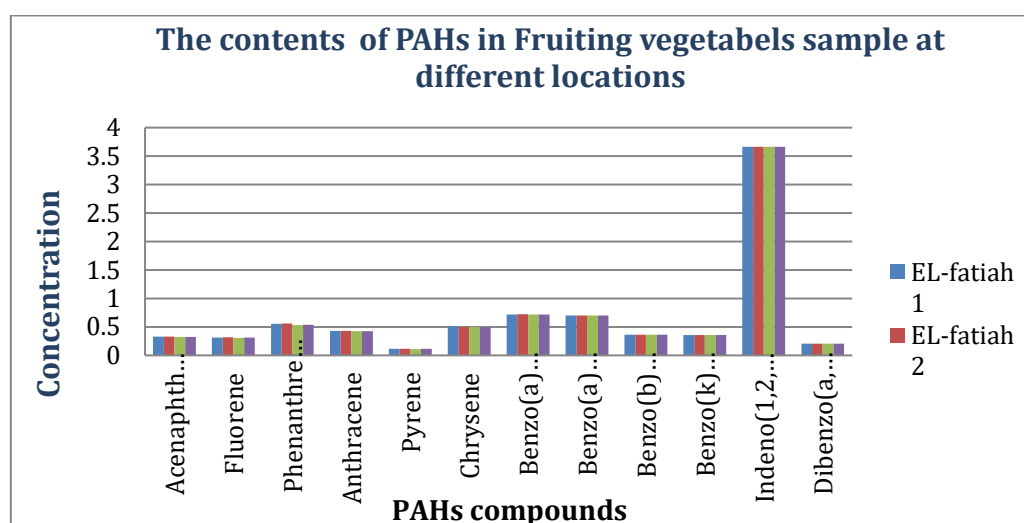


Figure 5. Distribution of the concentrations (µg/g) of poly aromatic hydrocarbons of fruiting vegetable samples at (EL-fatiah-1, EL-fatiah-2, Ain Mara, and Sidi Khalid 1) locations

Discussion

The main ways that PAHs are incorporated into plants are by foliar uptake, dry or humid deposition, with dry deposition being more common. Because PAHs are hydrophobic, root absorption is not thought to be an effective way for plants to incorporate them. The volatilization of PAHs from polluted soil can also result in foliar uptake. European Food Safety Authority (EFSA) maximum allowable limit. The European Food Safety Authority (EFSA) has established a maximum allowable limit of 0.54 to 1.5 µg/kg for the total of 15 PAH compounds found in vegetables (Dacal et al., 2021) [15]. With limited evidence of petrogenic origins, the current study concludes that incomplete combustion of recent and fossil organic matter (of pyrolytic origin) at high temperatures is the primary source of PAHs in the examined area. The primary causes of pyrolytic PAHs include industrial discharges, atmospheric deposition, and land runoff waters (Djomo et al., 2006) [16]. Generally speaking, the PAHs that have accumulated in agricultural soils and vegetables might originate from a variety of sources, including fuel combustion emissions, industrial emissions, and human activity. Additionally, the application of fertilizers and pesticides to boost productivity, as well as the burning of biomass and agricultural waste, contributed to a high level of contamination of agricultural fields. The growing demand for food has led to the expansion and intensification of agricultural systems. Environmental pollution loads have increased as a result of the overuse and abuse of agrochemicals, water, animal feed, and medications intended to boost productivity (Roozbahani et al., 2015) [17]. When compared to other agricultural systems in other nations, the level of PAH contamination in the vegetables in this study is moderate. It was discovered that the substantial aromatic proportion of the organic matter was primarily responsible for the high partitioning of PAHs to sedimentary organic matter. They viewed the organic matter as a naturally occurring "heterogeneous polymer" in which PAHs interact more positively with the aromatic areas [17].

The origin of PAHs was indicated by the Flu/Pyr ratio, one of several diagnostic ratios for PAH concentration that have been utilized to identify and quantify the contribution of each source of pollution to the designated environmental zones.

Coal and wood combustion produced (flu/pyr) ratios of 0.013 and 0.042, respectively. If the ratio of (flu/pyr) < 1 was ascribed to petrogenic sources, and if the ratio of (flu/pyr) > 1 is associated with pyrolytic origin. In the current investigation, all sites showed a Flu/Pyr ratio < 1, indicating a petrogenic origin, whereas crude oil and fuel oil had values between 0.6 and 0.9. In the study areas (EL-fatiah, EL-fatiah 2, Ain mara, and Sidi Khalid 1), the ratio of major combustion specific compounds ($\Sigma\text{COMB} = \text{Flu, Pyr, BaA, Chr, BbF, BkF, BaP, InP, and BghiP}$) to the total PAHs ($\Sigma\text{COMB} / \Sigma\text{PAHs}$) was 0.73, 0.82, 0.86, and 0.81 for the vegetables (tomato, pepper, eggplant, and zucchini), respectively [10].

The most powerful carcinogenic PAH, benzo(a)pyrene (BaP), and the total of six carcinogenic PAHs ($\Sigma\text{PAHsCARC} = \text{Flu, Pyr, BbF, BkF, BaP, and BghiP}$) for the species under study were listed in Table 5. Because physical and biogeochemical processes interact to change PAH profiles during transport and flux processes, it is important to utilize the predicted diagnostic isomeric ratios of PAHs with caution. Molecular indices based on ratios of certain isomers of PAH concentrations can be used to distinguish between the fingerprints of PAHs from petrogenic or pyrolytic origins [11].

Analytical Characteristics of the Method Used in PAHs Analysis

The GC-MS equipment was employed for the study of the prioritized PAHs. Prior to evaluating the samples, this technique is based on various stages, i.e., sampling, drying, isolating from the matrix, purification, concentration, and quantitative and qualitative analysis. With the exception of the lettuce sample, the chromatogram peaks of PAHs in all samples indicate that anthracene is the greatest peak (Table 3).

Table 3. peaks of the target compounds' analytical chromatography in every sample.

Compound	Concentration
Acenaphthylene	8.87
Fluorene	10.92
Phenanthrene	14.39
Anthracene	14.57
Pyrene	20.63
Benzo(a)anthracene	26.02
Chrysene	26.20
Benzo(b)fluoranthene	30.87
Benzo(k)fluoranthene	30.89
Benzo(a)Pyrene	32.12
Indeno(1,2,3-cd)pyrene	36.33
Dibenzo(a,b)anthracene	36.42

The potential for several contamination sources to coexist and the information processes that PAHs may undergo in the air, water, or wastes prior to deposition in the sediments under study make it challenging to identify the origins of PAHs. certain chemicals may have similar evolution kinetics that might be utilized to pinpoint the source of organic matter in the environment. The sources of production, the chemical makeup, and the temperature at which the organic matter burns all affect the distributions of aromatic compounds. Fossil fuels contain pyrene, phenanthrene, and benzo(b)fluoranthene, some of which are produced after combustion. Both catalysts and non-catalysts, as well as cars, commonly emit benzo(a)pyrene. Natural gas and diesel combustion commonly produce both chrysene and benzo(a)anthracene [11-12].

The ratios of individual PAH compounds based on oil peculiarities in PAH composition and distribution pattern as a function of the emission source can also be used to identify the sources of PAHs (Gschwend and Bites, 1981; Colombo et al., 1989). Pyrolytic contamination is the term used if the source is fuel combustion, and petrogenic contamination is the term used if the source is crude oil. Previous workers had used ratio values like (Phe/Ant) and (Flu/Pyr). When these two ratios are studied simultaneously, two distinct groups of sediments can be identified: Flu/Pyr <1 for petrogenic inputs and Phe/Ant >10 for petrogenic inputs [10].

Since phenanthrene is a more thermodynamically stable tricyclic aromatic isomer than anthracene, it is frequently found in petroleum in greater concentrations than anthracene. Accordingly, a Phe/Ant ratio is found to be low in situations of pyrolytic contamination but extremely high in petrogenic pollution caused by PAHs.

The Phe/Ant ratio of crude oil was approximately 50, while that of motor vehicle exhaust was approximately four. Low Phe/Ant ratio values (less than 10) suggested that the primary source of PAHs was the burning of fossil fuels proposed that sediments with Phe/Ant > 10 were primarily contaminated by petrogenic inputs, while Phe/Ant < 10 were typical of pyrolytic sources. Low Phe/Ant ratios (< 10) were detected in all regions in vegetable samples in the current study, suggesting that the sources of PAH contamination may differ. The ratio of specific PAH components varied between vegetable samples (Table 4). indicating that they were PAHs produced from pyrolysis. Nevertheless, the various Phe/Ant ratio values may be associated with weathering processes like photo-, chemical-, or bio-degradation, as well as the composition (for vegetable samples). The Flu/pyr ratio can indicate the source of PAH since. The majority of the samples in this study had Flu/pyr ratio values below 1. The Phe/Ant and Flu/Pyr ratio data, however, showed a heterogeneous pattern of pyrolytic contamination for the majority of vegetable samples. (Table 4).

The ratio of low molecular weight to high molecular weight compounds indicates that polyaromatic hydrocarbon compounds with two to three rings represent low molecular weight, while those with four to five rings represent high molecular weight. In all stations for vegetable samples, the ratio of (LMW/HMW) (Naphthalene + Acenaphthylene + Acenaphthene + Fluorene + Phenanthrene + Anthracene / Fluoranthene + Pyrene + Benzo(a)anthracene + Chrysene + Benzo(b)fluoranthene + Benzo(k)fluoranthene +

Benzo(a)pyrene + Dibenzo(a,h)anthracene + Benzo(ghi)Perylene + Indeno(1,2,3-cd)Pyrene) was less than 1, indicating that the source of PAHs was of pyrolytic origin.

Table 4. Poly aromatic hydrocarbon diagnostic ratios in fruiting vegetable samples (zucchini, eggplant, pepper, and tomato)

Ratio Species	(Flu/Pyr)	Σ COMB	Σ COMB /PAHs	(Phe/Ant)	Σ PAH _{CARC}	(LMW/HMW)
Tomato	2.739	6.038	0.73	1.29	1.484	0.25
Pepper	2.814	6.739	0.82	1.30	1.487	0.26
Eggplant	2.760	6.719	0.86	1.25	1.477	0.24
Zucchini	2.769	6.727	0.81	1.26	1.485	0.24

Table 5. The ratios of vegetable samples (R1–R6).

Ratio species	R1	R2	R3	R4	R5	R6
Tomato	1	0.732	0.580	0.564	1.02	0.469
Pepper	1	0.738	0.592	0.564	1.02	0.435
Eggplant	1	0.733	0.589	0.556	1.02	0.531
Zucchini	1	0.734	0.585	0.558	1.02	0.441

Generally, many studies on environmental impacts on the aquatic, soil, air, food, plants, and other samples were given importance in Libya. Where the determination of heavy metals, petroleum compounds, pesticides, radioactive, and toxic compounds was carried out [18 -45]. Most of these studies concluded that the human activities during 20 and 21st centuries were directly affecting the pollution of these samples [46 -86].

Conclusion

Based on the findings of this investigation, many kinds and compounds of polycyclic aromatic hydrocarbons were found in the chosen samples, indicating the origin and dispersion of these hydrocarbons. Numerous factors, such as the compounds' production and environmental dispersal, may influence the measured values when identifying the source of PAHs in agricultural fields. Because the uptake of toxins depends on the pollution source to which it is exposed, environmental factors might have an impact on the pattern of accumulation in the soil. The findings demonstrated that, mostly based on their place of origin, there are relative differences in the hydrocarbon molecules between locations. In general, pyrolytic and petrogenic sources account for the majority of the hydrocarbon compounds found in the samples from the examined area. Although the general public is most commonly exposed to polycyclic aromatic hydrocarbons (PAHs) through food, information on possible exposure to PAHs is of particular interest due to the highly immunotoxic and carcinogenic effects of these compounds on human health, regardless of the pathways of such accumulation.

Acknowledgment

Conflict

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