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# Fatty Acids Composition And Elements Analysis Of *Cassia Siamea Lam* Seed Oil

[1]Neetu, [2]Roli Agrawal
[1]Research Scholar, [2]Associate Professor
[1,2]Department of Chemistry,
[1,2] Shri Varshney College Aligarh- 202001,
[1,2]Dr. Bhim Rao Ambedkar University, Paliwal Park, Agra,(U.P.), 282004. India
neetujhansi@gmail.com

***Abstract:*** *Cassia Siamea Lam* is well known Ayurvedic medicinal tree. *Cassia Siamea Lam* is a genus of family Fabaceae and sub-family Caesalpinioideae which is selected for investigation. In this study, we determined the fatty acid composition and elements analysis of *Cassia Siamea Lam* seed oil. The oil was extracted from *Cassia Siamea Lam* seed using a Soxhlet extractor with petroleum ether as solvent. The fatty acid composition was analyzed by using the GC-FID technique and elements were analyzed by the ICP-MS instrument. Unsaturated fatty acids and saturated fatty acids are very important for pharmacological investigation. Linoleic and gamma linoleic acids are important unsaturated fatty acids that can be used in the synthesis of tissue hormones. A little amount of Trans fatty acid (0.14), substantial amounts of polyunsaturated fatty acids (56.81%), low amounts of monounsaturated fatty acid (17.18), and saturated fatty acid (25.87%) were noted to be present in this oil. Elements Pb, Cu, As, and Sn levels in this oil were measured. Although this oil contains lead and copper, it does not contain As and Sn. Cu is required in the body to prevent anemia, heart diseases and nervous disorders is found in this oil. A very small amount of Pb is present in this oil, which is not harmful to our bodies. Fatty acids and Elements of *Cassia Siamea Lam* seed oil suggest that it may find application in cosmetic, food consumption and pharmaceutical natural product formulations.

***Index Terms -*** *Cassia Siamea Lam*, Petroleum ether, Fatty acid Composition, Linoleic acid, Element analysis.

## I. INTRODUCTION

Fatty acids are essential for human growth and development. They are structural components of lipids that make up cell membranes and play an important role in processes such as gene expression and cellular communication. [1] Edible oils and fats are plant-derived biological mixtures composed of ester mixtures derived from glycerol and fatty acid chains. [2] The type and proportion of fatty acids in triacylglycerol influence both the physical and chemical properties of oils and fats. [3, 4] Saturated, mono-unsaturated (MUFA), and polyunsaturated (PUFA) fatty acids are the three types of fatty acids. Unsaturated fatty acids, on the other hand, are classified into a series known as omega, with omega-9 considered non-essential for humans and omega-3 and -6 considered essential because mammals cannot synthesize the latter; thus, they must be obtained through diet. [5, 6] The high percentage of unsaturated fatty acids in triacylglycerol is a feature shared by most plant-origin oils and fats. The higher the degree of unsaturation of fatty acids in vegetable oils, the more vulnerable they are to the oxidative deterioration. [4],[7-8] As a result, it is critical to understand the composition of fatty acids in oil or fat, as well as their characteristics and potential adulteration, as well as the stability and physical-chemical properties of these products. [4], [9] Short-chain fatty acids have a lower melting point and are more soluble in water. Longer chain fatty acids have higher melting points. Unsaturated fatty acids have a lower melting point than saturated fatty acids of comparable chain length. [10] Vegetable oils play important functional and sensory roles in food products, as well as serving as carriers of fat-soluble vitamins (A, D, E, and K). They also provide essential linoleic and linolenic acids, which are necessary for growth. [11] For human nutrition, the ratio of unsaturated to saturated fatty acids in dietary oils and fats is crucial. While high levels of saturated fatty acids are preferred to increase oil stability, they become unfavorable nutritionally because they are frequently thought to increase the concentration of low-density lipoproteins (LDL), alter the ratio of LDL to HDL (high-density lipoproteins), and encourage the growth of vascular smooth muscle and clothing. [12], [13] A higher intake of oleic acid lowers LDL cholesterol but has little effect on HDL cholesterol levels, whereas increasing the intake of linoleic and linolenic acids increases HDL cholesterol and decreases LDL cholesterol in the diet. [14]

*Cassia Siamea Lam* is a genus of family Fabaceae and sub-family Caesalpinioideae. The plant grows to a height of 10 to 12 meters, occasionally 20 meters. Initially has a short, compact, and spherical crown; later, it develops an uneven and spreading shape. Early on, the bark is smooth and greyish; subsequently, longitudinal fissures appear. The compound, alternating leaves measure 15–30 cm long and have 6–14 leaflets, each of which has a small bristle at the end. [15] It features erect, pyramid-shaped panicles and a bright yellow, up to 60 cm long flower. The fruits are flat, 5–30 cm long, and constricted between the seeds. They have an indehiscent pod. Each pod contains roughly 20 seeds. The beans-shaped, greenish-brown seeds are 8–15 mm long. [16] The seeds are employed as a repellent for intestinal worms as well as a treatment for scorpion and snake bites. [17-18]. Typhoid fever is treated and prevented in children by drinking a decoction made from a combination of *Cassia Siamea Lam* and Ficus thonnigii fruits. [19] Flowers and young fruits are frequently used in Sri Lanka and Thailand as vegetables and as a seasoning for curries. Both a laxative and a sleeping aid are produced by it. Additionally, this dish has sedative and anti-diuretic properties. [20-21]The current study's objective was to measure the concentrations of various fatty acids and confirm those using gas chromatography with flame-ionization detection in various edible sections of commercially grown Indian cultivars of M. oleifera. To choose nutrient-rich cultivars for functional food formulation, this information would be helpful.

## II. MATERIALS AND METHOD

### 2.1. COLLECTION OF PLANT MATERIAL

In Dhanipur Mandi Aligarh, seeds of the *Cassia Siamea Lam* plant were gathered. The collected seeds were crushed into a fine powder and stored for chemical analysis after being dried in direct sunshine for two days.

### 2.2. OIL EXTRACTION

According to the procedure described by, petroleum ether was used as a solvent to extract the oil from the powdered *Cassia Siamea Lam* seeds [22, 23]. The Soxhlet extractor was used to extract 100 g of powdered *Cassia Siamea Lam* seeds using petroleum ether as the extraction solvent. The seeds were packaged in filter paper for protection. The solvent was extracted after 24 hours using a water condenser, and the remaining oil was put into a desiccator and kept in an airtight container until it was required for analysis.

### 2.3. DETERMINATION OF FATTY ACID ANALYSIS

#### 2.3.1. SOLVENTS AND REAGENTS

Using a Super Purity Water System, the water for sample preparation had been created. Guidelines for GC-FID analysis: Fatty acid methyl ester, 37 Component (FAME) purity of fame mix ingredients must be at least 98.7% and Methanol (99.9%), chloroform (99.9%), hexane (>95%), toluene (99.7%), and a 10% solution of boron tri fluoride in methanol (1.3 M).

#### 2.3.2. EXTRACTION OF FATS FROM OIL SAMPLE

Using 10 mL of a chloroform/methanol combination (1:1) and 1000 L of water, fats from (0.5 g) oil were extracted. The obtained samples were then sealed and left to stand overnight at 20 0C 20 C in a dark environment. A new tube was used to transfer the 1 mL of chloroform layer, and the solvent was then evaporated off. The fatty acid esters were hydrolyzed and methylated simultaneously in a glycerol bath for 90 minutes at 70 0C with a solution of 100 L of toluene and 0.5 mL of boron tri fluoride/methanol. After cooling, 800 L each of distilled water and hexane were added. Fatty acid methylated esters (FAME) were transferred from the upper hexane layer to gas chromatography (GC-FID) vials after shaking and settling, where they were stored at 4 0C until analysis.

#### 2.3.3. QUANTITATIVE AND QUALITATIVE ANALYSES

Fatty acid was analyzed by AOAC Official method 996.06 methods [24]. 37 Component FAME Mix standard solution was used in accordance with the certified methodology for GC/FID. On the Shimadzu GC-2010 Plus, gas chromatography with flame ionization detector (FID) analysis was carried out. The injection volume was 1 L, helium was used as the carrier gas at a flow rate of 1.26 mL/min, the injector temperature was 230 0C, the flame ionization detector temperature was 250 0C, the split ratio was 1:20, and the oven temperature ranged from 100 to 2400 C with a stepwise temperature program over a 71.67 minute run time. A 100 m Restek RT2560" column with a 0.25 m diameter and 0.20 m thickness was employed for the analysis.

### 2.4. DETERMINATION OF ELEMENTS ANALYSIS

#### 2.4.1. SOLVENTS AND REAGENTS

Calibration standards were prepared using a mix of 1% HNO3 and 0.5% HCl. Cu, As, Sn, and Pb were calibrated from 0.5 to 10 ppb. For sample preparation, H2O2, HNO3, HCL, and distal water had been used. Determination of element compositions of Cassia Siamea Lam seed oil had been done using the methods described by AOAC 2013.06.

#### 2.4.2. SAMPLE PREPARATION

10 ml of concentrated HNO3 was added to 0.5 g of samples in a cork flask, and the mixture was let to stand overnight. Heat was applied to the material until no more fumes were being produced. After cooling the flask, 2-4 ml of 70% H2O2 were added. The solution was heated continuously until it became colorless. The solution was poured into a 100 ml flask and filled to the proper level with deionized water. Using ICP-MS, the total mineral content was evaluated.

#### 2.4.3. QUANTIFICATION OF ELEMENTS

Little modification was made to the analysis methods specified in AOAC 2013.06 [25] in order to determine Cu, As, Sn, and Pb. The analysis used ICP-MS equipment. The typical sample introduction system for ICP-MS was set up, comprising a Micro Mist glass concentric nebulizer, a quartz spray chamber, and a quartz torch with a 2.5 mm id injector. Both a nickel skimmer cone and a nickel-plated copper sampling cone made up the interface. The autotuned HMI settings are suitable for the target sample types' matrix levels. The HMI settings are automatically modified to match the target sample types' matrix levels. The HMI dilution factor in this instance was 4x. The technique of acquisition for all analytes was helium (He) collision.

The samples were digested using the following system for microwave digestion: Milestone Ethos MicroSYNTH oven with programmable power control (10W increments, maximum power 1000 W) with segmented rotor MPR-600 (operating pressure up to 35 bar maximum; operating temperature 260 maximum) and 10 reaction vessels (Milestone Srl. Sorisole, Italy). The spectrum was checked on the computer system and then interpreted for the quantitative determination of elements by direct comparison of count rates.

## III. RESULT AND DISCUSSION

### 3.1. FATTY ACIDS PROFILE

Thirty-seven fatty acids were identified in the *Cassia Siamea Lam* seed oil in Table-1. The result reveals that total saturated fatty acids constitute 25.87 %, total monounsaturated fatty acids constitute 17.18 %, and total polyunsaturated fatty acids amount to 56.80 %, and total Trans fatty acids amount to 0.14%. Elaidic acid and Linolaidic acid are Trans fatty acids. The total value of the Trans fatty acid is 0.14 % which is not harmful to our bodies. Among the saturated fatty acids present, palmitic acid was the most abundant (19.41 %) and Stearic acid (5.34%). The polyunsaturated fatty acids were mainly linoleic acid (51.02 %), linolenic acid (0.54 %), gamma-linolenic acid (2.05%), Linolenic acid (0.54%), and Arachidonic acid (0.40%). Omega 6 Fatty Acid (56.07%), Omega 3 Fatty Acid (0.74%), and Omega 9 Fatty Acid (16.57%) are present in *Cassia Siamea Lam* seed oil. Linoleic and linolenic acids are essential polyunsaturated fatty acids that the body cannot synthesize and they have been reported to be crucial in the maintenance of some key physiological functions in the body. A deficiency of bioactive linoleic acid leads to poor growth, fatty liver; skin lesions, and reproductive failure, [26] Oleic acid (15.49%) is a mono-unsaturated omega-9 fatty acid it is most commonly used for preventing heart disease and reducing cholesterol. Saturated and monounsaturated fatty acid content is less than of the total polyunsaturated fatty acid content hence *Cassia Siamea Lam* could be a dietary source of this acid in ameliorating health-related diseases. Monounsaturated fatty acids and saturated fatty acids are very important for pharmacological investigation. Linoleic and gamma linoleic acids are important polyunsaturated fatty acids that can be used in the synthesis of tissue hormo nes, which regulate blood pressure take part in immunological response, and show any inflammatory and immunity ambulatory activity. (41, 45)

**TABLE -1: SATURATED, MONOUNSATURATED, POLYUNSATURATED FATTY ACIDS AND TRANS FATTY ACID PROFILE OF *CASSIA SIAMEA LAM SEED* OIL**

| Polyunsaturated fatty acids | Percent (%) |
|---|---|
| Linoleic acid (C18:2) | 51.02 |
| Gamma linolenic acid | 2.05 |
| Linolenic acid (C18:3) | 0.54 |
| Cis-8-11-14 eicosatrienoic | 0.07 |
| Cis-11-14-eicosadienoic | 2.53 |
| Cis-11-14-17-eicosadienic | 0.00 |
| Archidonic acid (METYHL CID)(C20:4) | 0.40 |
| Cis-13-16-docosadienic | 0.00 |
| Cis-5-8-11-14-17-eicosanoic | 0.20 |
| Cis-4-7-10-13-16-19 | 0.00 |
| **TOTAL** | **56.81** |

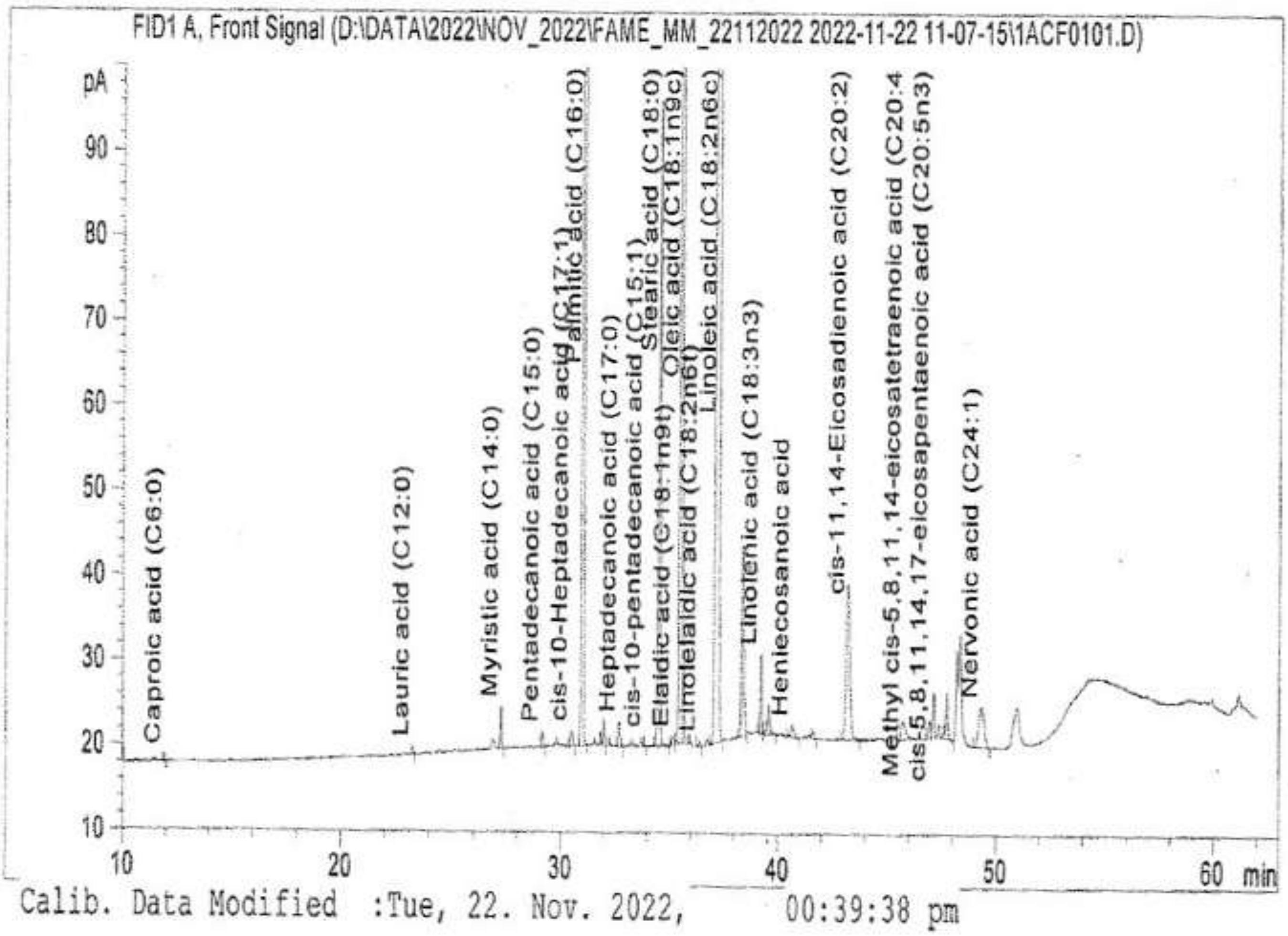
| Saturated Fatty acids | Percent (%) |
|---|---|
| Butyric acid (C4:0) | 0.00 |
| Caproic acid (C6:0) | 0.03 |
| Caprylic acid (C8:0) | 0.00 |
| Undecanoic acid (C11:0) | 0.00 |
| Lauric acid (C12:0) | 0.04 |
| Tridacanoic acid | 0.00 |
| Myristic acid (C14:0) | 0.21 |
| Pentadecanoic acid | 0.08 |
| Palmitic acid (C16:0) | 19.41 |
| Hepta decanoic acid | 0.20 |
| Stearic acid (C18:0) | 5.34 |
| Arachidic acid (C20:0) | 0.00 |
| Heniecosanoic acid | 0.18 |
| Behenic acid (C22:0) | 0.00 |
| Tricosanoic acid | 0.00 |
| Lignoceric acid (C24:0) | 0.38 |
| Capric acid | 0.00 |
| TOTAL | 25.87 |

| Monounsaturated fatty acids | Percent (%) |
|---|---|
| Myristoleic acid (C14: In 9c) | 0.00 |
| Cis 10 pentadecanoic acid | 0.18 |
| Palmitoleic acid (C16:1 In9c) | 0.14 |
| Cis 10 hepta decanoic acid | 0.05 |
| Oleic acid (C18: In9c) | 15.49 |
| Cis 11 eicosanoic acid | 0.33 |
| Erucic acid (C22: In9c) | 0.00 |
| Nervonic acid (C24:1) | 0.99 |
| **TOTAL** | **17.18** |

| Trans Fatty acids | Percent (%) |
|---|---|
| Elaidic acid | 0.09 |
| Linolaidic acid | 0.05 |
| **TOTAL** | **0.14** |

Fatty acids play an essential role in the human diet and health. A high amount of lipids may provide abundant fatty acids. The fatty acid composition of oil mostly depends on the botanical and geographical origin, as well as on the used methodology for the isolation and extraction of fatty acids. [27-28] The method developed in this study was employed for the determination of saturated/unsaturated fatty acids, and the ratio of total unsaturated/saturated fatty acids. All investigated fatty acids were identified by their retention times. The typical chromatogram of the *Cassia Siamea Lam* seed oil was shown in Figure -1, and fatty acids contents were summarized in table-1

**Figure-1. Chromatogram of the 37 Component FAME mix obtained using the Varian GC-FID equipment**




**3.2. Aliment analysis profile**

The levels of micro and macro elemental nutrients were presented in Table 2. Minerals are found to be important in human nutrition. [31] It is evident from Table 2 that there is a significant difference in elements (Lead, Copper, Arsenic, and Tin).

This sample contains copper (Cu), which the body needs to prevent anemia, heart disease, and nervous system illnesses. It is also in charge of producing hormones, enzymes, and vitamins. Cu deficiency causes aneurysm formation and skeletal mal development by reducing the tinsel strength of artery walls. [32] Lead is absorbed and retained by our bodies in our bones, blood, and tissues. It is stored there as a source of ongoing internal exposure; it does not remain there permanently. Because lead is poisonous, it should only be used and disposed of with extreme caution around people. [33-34] As shown in Table 2, this oil contains 0.68 mg/kg of Cu, 0.03 mg/kg of Sn, and 0.12 mg/kg of Pb.

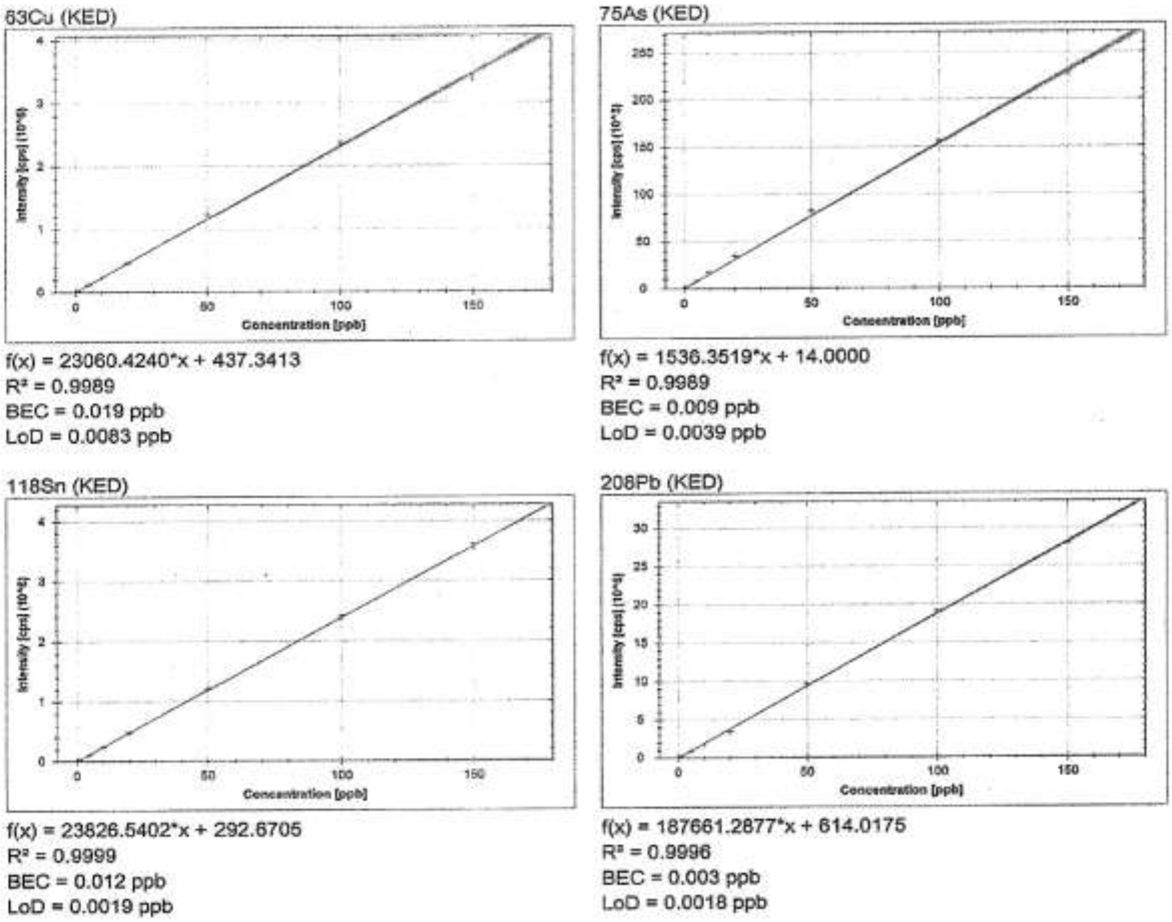
**Table -2. Element contents of *Cassia Siamea Lam* seed oil**

| Element | Sample wt. | Vol. | Dil. | Sample Conc.(µg/kg) | Reagent Blank(µg/kg) | Final Conc.(mg/kg) |
|---|---|---|---|---|---|---|
| Cu | 0.1310 | 50 | 1 | 3.054 | 1.263 | 0.68 |
| As | 0.1310 | 50 | 1 | 0.017 | 0.027 | 0.00 |
| Sn | 0.1310 | 50 | 1 | 0.09 | 0.021 | 0.03 |
| Pb | 0.1310 | 50 | 1 | 0.471 | 0.157 | 0.12 |

**Calibration curves and verification**

Trace elements Cu, As, Sn, and Pb are shown in Figure 2. All curves show excellent linearity across the calibration range.

**Figure - 2. Calibration curves for Cu, As, Sn, and Pb**




## IV. CONCLUSION

37 fatty acids were identified by GC-FID in *Cassia Siamea Lam* seed oil. In this oil palmitic acid (19.41 %), and linoleic acid (51.02 %). The oil was also recorded as containing low amounts of monounsaturated fatty acid (17.18), saturated fatty acid (25.87 %), high amounts of polyunsaturated fatty acids (56.81 %), and a small amount of Trans Fatty acid (0.14). The value of polyunsaturated fatty acids can increase the health advantages through functional food products. Omega 6 fatty acids make up 56.07% of *Cassia Siamea Lam* seed oil, Omega 3 fatty acids are 0.74%, and Omega 9 fatty acids are 16.57%.The findings of this study suggested that *Cassia Siamea Lam* seed oil is beneficial for both human and animal health. The findings of this study suggested that *Cassia Siamea Lam* seed oil is beneficial for both human and animal health. If appropriately ingested can act as a possible source of functional meals in line with the rise in world food consumption.

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