

Pharmacognostic evaluation of *Terminalia chebula* standard extracts and finished products

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Abstract: The different commercial samples of *Terminalia chebula* (*T. chebula*) standard extracts and finished products were taken for elemental analysis studies and were evaluated by ICP-OES and compared with standard reference data. Trace elements are naturally occurring and its daily requirement as dietary supplement form is only a few milligrams and even micrograms. They are categorized as essential and non-essential, and that is based upon their biological action, diseases that occur due to their deficiency and toxicity due to overdose. Use of herbal medicines is now increasing worldwide because of their minimal side effects. In general, *Haritaki* (*T. chebula*) acts as a blood purifier and prevents haemorrhage. It is also used as an internal cleanser by removing harmful toxins and excess fats out of the body. It can strengthen the hair roots and enriches hair color. The elements were determined quantitatively using the modern technique of ICP-OES. The extracts undertaken for this study contains Ca as a primary elemental content, and its concentration is found to be as high as 144 mg/kg in comparison with other macro elements. Ca helps to overcome the problems of high blood pressure, heart attack, premenstrual syndrome, and colon cancer, besides its ability to keep the bones healthy and to reduce the risk of osteoporosis. Other essential elements such as Fe (2.0732 mg/kg), Zn (1.0235 mg/kg), and Mn (0.2500 mg/kg) were also present but not to an appreciable amount in the extracts of *T. chebula*. It was concluded that due to soil nature, climatic condition, time of harvest could be a detrimental factor while reviewing the elemental profile.

Keywords: *T. chebula*, microwave digestion, ICP-OES, trace elements.

Introduction

Ayurvedic is an ancient tradition for about 5,000 years old practice and deep-rooted in the earlier civilization of Indian culture¹. According to the World Health Organization (WHO), about 80% of the world population depends mainly on plant-based traditional medicine for their primary healthcare requirement². When screening a number of herbal medicinal plants, scientists were discovered that *T. chebula* (Combretaceae) is one of the most revered medicinal plants, possesses several medicinal values due to the presence of many kinds of phytochemical constituents. *T. chebula* has been extensively used in ayurvedic, Unani & homoeopathic medicine and has become the cynosure of modern medicine³. *T. chebula* possesses a wide variety of activities like anti-microbial⁴, antioxidant⁵, anti-viral⁶, anti-carcinogenic⁷, hypocholesterolemic⁸, radio-protective⁹, anti-spasmodic and anti-purgative¹⁰.

The phytochemicals present in *T. chebula* are polyphenols, terpene, anthocyanins, flavonoids,

alkaloids, and glycosides. It is used as a natural cleanser of the digestive system. It improves the functioning of liver, spleen and colon and hence it is widely used as a digestive tonic. From some research studies reveals that the oil in the kernel of *T. chebula* contain certain substances that induce the motility of the gastrointestinal tract¹¹. This action was similar to that of castor oil. It is also used in combination with two more herbal extracts to prepare a formulation that is popularly known as Triphala. This formulation is used for enhancing body immunity. *T. chebula* contains appreciable amounts of tannins, which possess antiseptic and healing properties and hence, it is widely used externally for curing chronic ulcers, wounds, and piles¹²⁻¹⁵.

It has purgative properties¹⁶. Fine powder of *T. chebula* extracts is used in dental preparations^{17,18}. *T. chebula* possesses anti-bacterial¹⁹, anti-cancer²⁰, anti-caries^{21,22}, antimutagenic potential²³, and it inhibits local anaphylaxis²⁴. The fruit is used for its mild laxative, alterative, and anti-spasmodic effects.

T. chebula is useful for inducing appetite, as a digestive aid, liver stimulant and as gastrointestinal pro-kinetic agent²⁵⁻²⁷. It is useful in treating skin disorders with discharges like allergies, urticaria and other erythematous disorders²⁸⁻³⁰. *T. chebula* Retz (Combretaceae) has a legendary reputation as a male contraceptive³¹.

The present study has been undertaken with four samples of standard extracts procured from different geographical locations of India, and three samples of domestic market products containing *T. chebula* as formulated medicine were evaluated for their essential and trace elements contents and compared to NIST standard.

Material and Methods

Collection of Samples

Well-matured, healthy and dried fruits of *T. chebula* were collected from different locations in India (April to June).

Site I: Tambaram, India (12° 55' 22.4940" N and 80° 7' 38.8452" E)

Site II: Bangalore, India (12.9716° N and 77.5946° E)

Site III: Coimbatore, India (11° 0' 16.4016" N and 76° 57' 41.8752" E)

Site IV: Nagpur, India (21° 8' 47.8788" N and 79° 5' 19.8960" E)

Standard *T. chebula* extracts (TCEs):

Code	Source	Mass	Volume
TCE-1	Tambaram	1.0170 g	50 ml
TCE-2	Bangalore	1.0276 g	50 ml
TCE-3	Coimbatore	1.0143 g	50 ml
TCE-4	Nagpur	1.0198 g	50 ml

Finished products consisting of *T. chebula* as a major ingredient (TCFs):

Code	Source	Mass	Volume
TCF-1	Chennai	1.0303 g	50 ml
TCF-2	Coimbatore	1.0763 g	50 ml
TCF-3	Bangalore	1.0414 g	50 ml

Solutions and Reagents

Standard solutions were prepared by diluting individual metal standard solution (1,000 µg/ml) procured from BDH Middle East L.L.C. with 1 M HNO₃. The plant extract was incinerated in a muffle furnace and made into ash, which was then digested with HNO₃ + 30% H₂O₂ and prepared the solutions.

Preparation of Sample by Microwave Digestion Method

Microwave digestion involves the application of electromagnetic radiation with the frequency of 2,450 MHz to dissolve the samples³². The microwaves

interact with polar molecules and induce alignment of the molecular dipole moment with the microwave field. While field constantly changes and causing the molecular rotation and make intermolecular collisions occurs among themselves and heat evolves. Consequently, the rate of microwave digestion is dependent on the coupling efficiency of microwaves with the digestion acids. Microwave technology is often recommended for safety considerations. They are also programmable and can accommodate large numbers of samples. *T. chebula* extract was weighed accurately and transferred into seven fluorocarbon microwave vessels and added with 10 ml concentrated HNO₃ to each of it. Sealed the vessel and placed correctly in the microwave system (Anton par Microwave digester). A blank sample was also prepared simultaneously by placing 10 ml of concentrated HNO₃ acid in one of the empty fluorocarbon microwave vessels and placed it in the microwave system after sealing it tightly. All the samples and blank were digested at 175°C for 10 minutes. After cooling it to a sufficient time, carefully uncapped and vented each vessel in a fume cupboard. Contents of each vessel then transferred to 50 ml volumetric flask and made up to the mark with Mille-Q water.

Preparation of Reference Standard Solution

Standard solutions of each element were separately prepared using reference standard metal solutions. The procedure followed was as per the Analysis Guidebook of Shimadzu Solutions for Science. The solutions prepared were placed in tightly capped plastic bottles and used within a day of their preparation.

Preparation of Reagent Blank Solution

About 10 ml of concentrated Nitric acid and 2 ml of 30% hydrogen peroxide were quantitatively added into 50 ml volumetric flask and diluted with de-ionized water to the mark. These solutions were served as blank solution.

Determination of Elements Concentration

Elemental profile of *T. chebula* was determined quantitatively by Inductively Coupled Plasma – Optical Emission Spectroscopy (ICP-OES) (VARIAN INC. – liberty series, axially viewed plasma, following type ICP-OES spectrometer, wavelength range 189-940 nm). Using standard references, the calibration curves were obtained for all the elements. The sample solutions were adequately diluted to keep the absorbance in the linear range of measurement.

Statistical Analysis

All assays were carried out in triplicates and values were obtained by calculating the average of five experiments. Data are presented as mean ± SEM.

Standard Error of the Mean (SEM): if the parameter or the statistic is the mean, it is called the standard error of the mean (SEM).

The SEM is calculated by dividing the SD (σ) by the square root of n (number of samples).

The formula to calculate the standard deviation (σ):

$$\sigma = \sqrt{[\sum (x - \text{mean})^2 / n]}$$

$x = \text{data point}$

$n = \text{number of samples}$

The formula to calculate SEM:

$$\text{SEM} = \sigma/\sqrt{n}$$

Results and Discussion

Many ayurvedic preparations were successful in treating various diseases, but still, there is no concrete evidence in understanding the precise molecular mechanism. The pharmacological action of most of the plants could be usually focused on its organic, but not its inorganic, content. New research has now focused on the inorganic contents of herbal plants due to the presence of certain trace elements in a bioavailable form. A direct link between the healing ability and the role of trace elements in herbal plants are yet to be proven, and such studies are critical in understanding their pharmacological activities. To determine the trace elements more accurately, techniques like ICP-OES and Atomic Absorption Spectrophotometer (AAS) are a better choice because of their multi-elemental detection capability over a wide range of lower concentration, their blank-free nature, and minimum sample preparation³³.

The preparation procedure must ensure that simultaneous quantitative mineralization and dissolution by the use of appropriate chemical reagents and by physical means. Incomplete dissolution could cause poor recoveries for many elements due to the presence of silicates, especially those relatively abundant in plant body and partly associated with silicates. Based on the data in the literature, from this study, we found that the recovery of Al, Fe, and B is feeble in detection. After the preparation procedure applied in this work, the sample matrix was largely simplified, and the resulting solutions were clear, colorless and odorless with no observed residue with complete dissolution was achieved in most cases.

Concerning Atomic Emission Spectroscopic (AES) analysis, no particular precautions have to be taken if the measured concentrations satisfy the principal criteria (sensitivity, detection limits, working range) and if possible interfering substances should be under control. At optimal plasma condition, all the measurements were made. ICP-AES is mainly used for quantitative measurement of various elements in ppm level concentration.

The entire procedure starting from mineralization, dissolution and measurements were all validated by using AOAS method and compared with CRM (NIST 1573a - Tomato Leaves). It is found that the values obtained from the analysis are very close to the CRM values. % of recovery is also given in Table 1.

Table 1. Results of CRM (NIST 1573a - Tomato Leaves) in ppm (mg/kg). Values are in % for Ca.

Element	Found	Certified Value	Recovery Values (%)
Cu	7.55	4.70 ± 0.14	160.6
Zn	36.43	30.9 ± 0.70	117.9
Mn	225.8	246 ± 8.00	91.8
Fe	304.00	368 ± 7.00	82.6
Co	0.64	0.57 ± 0.04	112.3
Cr	2.23	1.99 ± 0.06	112.2
Ni	0.90	1.59 ± 0.04	56.60
Pb	1.04	Not available	Not available
Ca	5.1%	5.05 ± 0.09%	101
Al	526.4	598 ± 2.00	88.03
Cd	1.33	1.52 ± 0.03	87.5
K	2.46	2.70 ± 0.05%	91.1
Na	400.5	136 ± 4.00	294.5

For statistical analysis, regression analysis and SEM analysis were performed. The correlation coefficients (R) were determined, and an external calibration was carried out. The correlation coefficients were found in the range of 0.9981-0.9999. Table 2 and Figures 1-4 summarize the results of the

elemental analysis of *T. chebula* aqueous extracts (TCE-1, TCE-2, TCE-3, and TCE-4). Table 3 and Figures 5-8 summarize the results of the elemental analysis of *T. chebula*-containing finished products (TCF-1, TCF-2, and TCF-3).

The data in Table 2 indicate that the medicinal plant extract contains macro, micro, trace and toxic elements in various levels of concentrations. All the elements were analyzed by ICP-OES technique by measuring the emission of each element at its resonance wavelength. The nature of soil, the place where the plants were cultivated, and the complex nature of botanical structures are the essential factors that contributed to variation in concentration of each element. Other considerations leading to the variations in the elemental composition include environmental factors such as water and climatic

conditions, as well as the use of fertilizers and irrigation water.

The active constituents of the medicinal plants are the metabolic products of the plant cells. Several trace elements play an essential role in metabolism. These elements are called essential.

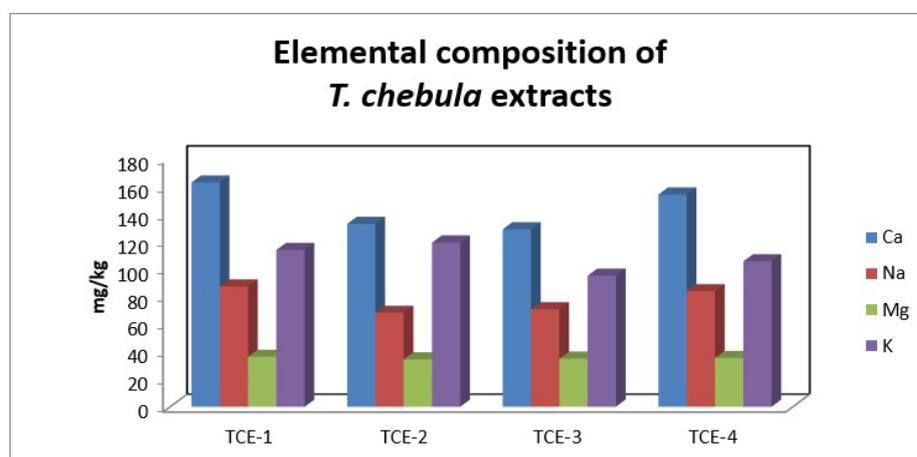
The elements play both curative and preventive roles in combating diseases³⁴. There is a vast scope to exploit the preventive medicinal aspects of various elements.

Table 2. Elemental composition of *T. chebula* aqueous extracts (TCE-1, TCE-2, TCE-3, and TCE-4).

Element	TCE-1 (mg/kg)	TCE-2 (mg/kg)	TCE- 3 (mg/kg)	TCE- 4 (mg/kg)
		Macro Elements		
Ca	162.50 ± 0.20	132.500 ± 0.100	128.560 ± 0.080	153.740 ± 0.180
Na	87.106 ± 0.04	68.000 ± 0.070	70.605 ± 0.070	83.675 ± 0.110
Mg	36.200 ± 0.324	34.300 ± 0.280	34.821 ± 0.292	35.345 ± 0.312
K	113.60 ± 0.25	119.000 ± 0.370	94.960 ± 0.790	105.360 ± 0.540
		Micro Elements		
Al	11.713 ± 0.008	8.6000 ± 0.003	14.097 ± 0.015	12.874 ± 0.0110
B	0.4830 ± 0.0096	0.5789 ± 0.0071	0.3534 ± 0.0082	0.4267 ± 0.0105
Ba	3.7100 ± 0.0053	3.6330 ± 0.0047	3.6759 ± 0.0051	3.6528 ± 0.0032
Bi	0.1150 ± 0.0123	0.9000 ± 0.0095	0.1074 ± 0.0104	0.1123 ± 0.0117
Cd	0.0060 ± 0.0015	0.0030 ± 0.0008	0.0030 ± 0.0008	0.0043 ± 0.0012
Co	0.0220 ± 0	0.0567 ± 0.010	0.0200 ± 0.008	0.0214 ± 0.0050
Cr	0.2000 ± 0.0073	0.7076 ± 0.0243	0.3478 ± 0.0115	0.2543 ± 0.0083
Cu	1.8202 ± 0.0243	1.7454 ± 0.0191	1.696 ± 0.0153	1.7453 ± 0.0189
Fe	2.0205 ± 0.0073	2.3300 ± 0.0094	1.9110 ± 0.0061	2.0438 ± 0.0102
Li	0.0400 ± 0.0081	0.0395 ± 0.0077	0.0433 ± 0.0118	0.0424 ± 0.0109
Mn	0.2850 ± 0.0123	0.2400 ± 0.0111	0.2390 ± 0.0107	0.2543 ± 0.0118
Mo	0.0023 ± 0.0005	0.0046 ± 0.0012	0.0018 ± 0.0003	0.0034 ± 0.0008
Ni	0.4238 ± 0.0031	0.4960 ± 0.0052	0.3578 ± 0.0017	0.4327 ± 0.0036
P	3.1064 ± 0.0097	2.9412 ± 0.0073	3.0435 ± 0.0081	3.1230 ± 0.0103
Pb	0.6874 ± 0.0069	0.6754 ± 0.0052	0.6323 ± 0.0029	0.6824 ± 0.0061
Rb	0.0305 ± 0.009	0.0976 ± 0.019	0.0876 ± 0.017	0.0326 ± 0.0110
Se	0.2130 ± 0.007	0.2436 ± 0.016	0.2324 ± 0.012	0.2254 ± 0.0090
Si	3.0524 ± 0.0054	2.9348 ± 0.0048	3.0754 ± 0.0059	3.1463 ± 0.0065
Sn	0.0193 ± 0.0007	0.0204 ± 0.0011	0.0198 ± 0.0009	0.0197 ± 0.0009
Sr	3.5102 ± 0.0071	3.5234 ± 0.0078	3.5765 ± 0.0089	3.5268 ± 0.0079
Te	0.9514 ± 0.0049	0.9668 ± 0.0067	0.9576 ± 0.0054	0.9534 ± 0.0051
Ti	0.0773 ± 0.0009	0.0745 ± 0.0006	0.0721 ± 0.0004	0.0769 ± 0.0008
V	0.0269 ± 0.0007	0.0276 ± 0.0009	0.0265 ± 0.0005	0.0263 ± 0.0004
Zn	1.0239 ± 0.0032	1.0275 ± 0.0049	1.0269 ± 0.0045	1.0235 ± 0.0033

Table 3. Elemental composition of *T. chebula* finished products (TCF-1, TCF-2, and TCF-3).

Elements	TCF-1 (mg/kg)	TCF-2 (mg/kg)	TCF-3 (mg/kg)
Macro Elements			
Ca	543.35 ± 0.13	549.45 ± 0.17	587.55 ± 0.570
Na	40.480 ± 0.110	45.300 ± 0.095	42.390 ± 0.065
Mg	57.106 ± 0.040	53.560 ± 0.018	55.879 ± 0.028
K	634.19 ± 0.06	637.07 ± 0.36	647.28 ± 0.53
Micro Elements			
Al	1.7135 ± 0.0038	1.6000 ± 0.0023	1.4097 ± 0.0017
B	0.2830 ± 0.0023	0.1789 ± 0.0009	0.1534 ± 0.0005
Ba	1.7100 ± 0.0046	1.6330 ± 0.0027	1.6759 ± 0.0033
Bi	0.0150 ± 0.0006	0.0400 ± 0.0014	0.0174 ± 0.0008
Cd	0.0010 ± 0.0002	0.0020 ± 0.0004	0.0020 ± 0.0004
Co	0.0220 ± 0.0013	0.0567 ± 0.0032	0.0200 ± 0.0010
Cr	0.8000 ± 0.0021	0.7076 ± 0.0008	0.7478 ± 0.0013
Cu	1.4202 ± 0.0043	1.3654 ± 0.0037	1.4960 ± 0.0052
Fe	4.0205 ± 0.0011	4.3300 ± 0.0036	4.9110 ± 0.0087
Li	0.0200 ± 0.0007	0.0195 ± 0.0006	0.0233 ± 0.0009
Mn	0.5850 ± 0.0034	0.6400 ± 0.0046	0.5390 ± 0.0023
Mo	0.0012 ± 0.0002	0.0036 ± 0.0008	0.0018 ± 0.0005
Ni	2.1064 ± 0.0023	1.9412 ± 0.0019	3.0435 ± 0.0041
P	0.6328 ± 0.0104	0.5259 ± 0.0065	0.4532 ± 0.0032
Pb	0.8765 ± 0.0027	0.7676 ± 0.0013	0.8524 ± 0.0021
Si	0.1436 ± 0.0018	0.1324 ± 0.0015	0.0993 ± 0.0008
Sn	0.0114 ± 0.0004	0.0146 ± 0.0011	0.0124 ± 0.0006
Sr	1.9348 ± 0.0045	2.0754 ± 0.0054	1.9510 ± 0.0048
Te	0.7432 ± 0.0026	0.6765 ± 0.0013	0.7773 ± 0.0033
Ti	0.0345 ± 0.0007	0.0344 ± 0.0007	0.039 ± 0.0008
V	0.0145 ± 0.0009	0.0121 ± 0.0004	0.0139 ± 0.0006
Zn	3.0276 ± 0.0011	3.5265 ± 0.0035	3.9765 ± 0.0043

**Figure 1.** Elemental composition of *T. chebula* extracts (TCE-1, TCE-2, TCE-3, and TCE-4) [Ca, Na, Mg, and K].

Calcium (Ca) is a macro element and found in abundant quantities in the *T. chebula* extract. The average concentration Ca found to be 144.3 mg/kg in *T. chebula* aqueous extract. The values are ranging from 128.56 to 162.5 mg/kg for samples TCE-1 to TCE-4. To overcome the problems of high blood pressure, heart attack, premenstrual syndrome, colon cancer and keeping the bones healthy and reduces the risks of osteoporosis in old age calcium element plays a vital role in those diseases ^{35,36}.

Magnesium (Mg) improves insulin sensitivity, protects against diabetes and its complications and reduces the blood pressure ^{37,38}. The average concentration Mg was 35.2 mg/kg in *T. chebula* aqueous extract. The range of values for Mg is from 34.3 to 36.2 mg/kg for the samples TCE-1 to TCE-4.

Sodium (Na) imparts production of energy, transport of amino acids and glucose into the body cells, the average concentration Na was found to be 77.3 mg/kg in *T. chebula* aqueous extract ³⁹.

Potassium (K) is helpful to reduce hypertension and to maintain smooth, cardiac function. In the human body, potassium plays vital roles in many physiological functions, and their deficiency or excess could affect human health ^{40,41}. The average concentration of K was 108.2 mg/kg in *T. chebula* aqueous extract. The range of value obtained for TCE-1 to TCE-4 is 94.960 mg/kg (TCE-3) to 119.00 mg/kg (TCE-2).

Copper (Cu) plays a vital role in the treatment of wounds and prevent inflammation in arthritis and

similar diseases ^{42,43}. The average concentration Cu was 1.752 mg/kg in *T. chebula* aqueous extract. The range of values obtained for samples TCE-1 to TCE-4 are 1.6960 mg/kg (TCE-3) to 1.8202 mg/kg (TCE-1).

Iron (Fe) is an essential element in the human body for the production of haemoglobin and oxygenation of red blood cells. Fe is needed for a healthy immune system and energy production ^{44,45}. The average concentration of Fe was found to be 2.0763 mg/kg in *T. chebula* aqueous extract. The range of value is from 1.9110 mg/kg (TCE-3) to 2.3300 mg/kg (TCE-2).

Manganese (Mn) can help to assist the human body in metabolizing protein, helps in metabolize carbohydrates and treating diabetes ^{47,48}. The average concentration Mn was 0.255 mg/kg in *T. chebula* aqueous extract. The range is between 0.2390 mg/kg (TCE-3) and 0.2850 mg/kg for (TCE-1).

Zinc (Zn) deficiency would contribute to arrested sexual maturation, growth retardation and hair loss, delayed wound healing, and emotional disturbance ⁴⁹. The average concentration of Zn was 1.025 mg/kg in *T. chebula* aqueous extract. The range of Zn was found to be between 1.0235 mg/kg (TCE-4) and 1.0275 mg/kg (TCE-2).

Based on the above details the essential elements such as Fe/Mn/Zn/Cu are not present in detectable levels, and hence formulators aiming for fortifying their products with these elements from *T. chebula* are not a preferable choice.

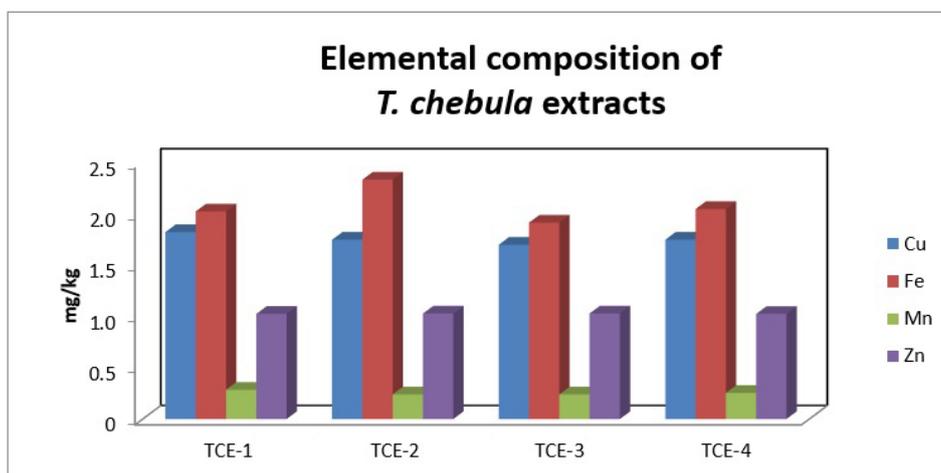


Figure 2. Elemental composition of *T. chebula* extracts (TCE-1, TCE-2, TCE-3, and TCE-4) [Cu, Fe, Mn, and Zn].

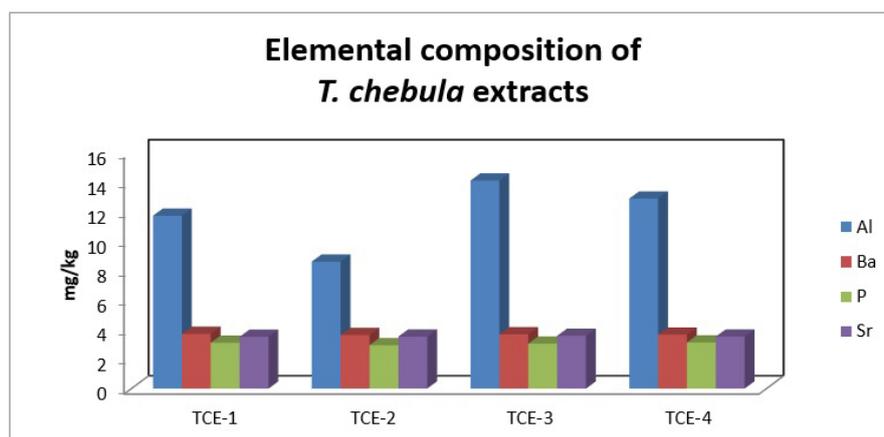


Figure 3. Elemental composition of *T. chebula* extracts (TCE-1, TCE-2, TCE-3, and TCE-4) [Al, Ba, P, and Sr].

Aluminum (Al) is used in the preparation of antacids and food additives. It is used as a component of dental cement and in the preparation of aluminum acetate ear drops^{50,51}. It is also used to treat ulcers and arrest foul discharges from mucous surfaces. The mean value of Al is 11.821 mg/kg. The range is from 8.6000 mg/kg (TCE-2) to 14.097 mg/kg (TCE-3).

Barium (Ba) is not extensively used element in practice. Of note, “barium meal” and “barium enemas” (barium sulfate) are given to patients suffering from digestive disorders⁵². The action of the stomach and intestines can be seen as the metal’s progress through the body is revealed by X-ray imaging. The average concentration of Ba from samples TCE-1 to TCE-4 is 3.6679 mg/kg, and range is between 3.6330 mg/kg (TCE-2) and 3.7100 mg/kg (TCE-1).

Phosphorous (P) is an essential mineral and is required for the healthy formation of bones and teeth,

and it is necessary to process many of the foods that we eat⁵³. It is also a part of the body’s energy storage system and helps to maintain healthy blood sugar levels. The proper heart functioning, cell growth and cell repair require an adequate amount of P in the body. The average concentration of P was found to be 3.0537 mg/kg and the range of values found to be 2.9412 mg/kg (TCE-2) and 3.1235 mg/kg (TCE-4).

Strontium (Sr) is used in scientific research to measure the release of neurotransmitters by neurons⁵⁴. Ca is replaced by Sr, which makes it easier to observe the neuron’s response. Radioactive Sr is used in radio-pharmaceuticals to treat metastatic bone cancer. Human body parts that are experiencing increased bone growth will absorb Sr instead of Ca. The average concentration of Sr is 3.5342 mg/kg and the range of values between 3.5102 mg/kg (TCE-1) and 3.5765 mg/kg (TCE-3).

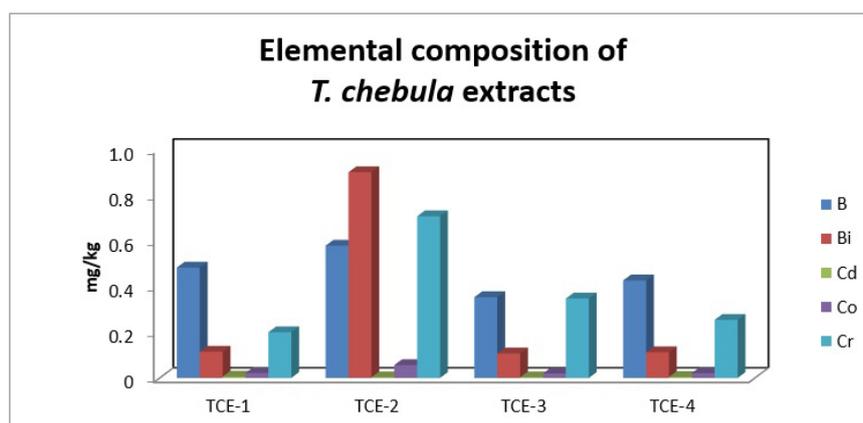


Figure 4. Elemental composition of *T. chebula* extracts (TCE-1, TCE-2, TCE-3, and TCE-4) [B, Bi, Cd, Co, and Cr].

Boron (B) is necessary to allow the brain to function properly and can increase mental alertness⁵⁵. Without sufficient amounts of B, bones would slowly weaken and become brittle⁵⁶. The average value of B was found to be 0.4605 mg/kg. The concentration

range was found to be 0.3534 mg/kg (TCE-3) and 0.5789 mg/kg (TCE-2).

Bismuth (Bi) is essentially a non-toxic in nature, but prolonged exposure or excessive use may lead to toxicity⁵⁷. This could cause mental confusion, memory loss, stammering during a speech, joint pain,

or muscle twitching and spasm. It is present in almost a trace amount in the plant material ⁵⁸. The average concentration of Bi is 0.3087 mg/kg, and the range was found to be 0.1074 mg/kg (TCE-3) and 0.9000 mg/kg (TCE-2).

Cadmium (Cd) is an element on which scientists have mixed opinions. While it is undoubtedly believed to be non-essential for plant and animal life processes, some believe Cd is a trace element with some important role in life processes ^{59,60}. The average concentration was found to be 0.0041 mg/kg, and the range is between 0.0030 mg/kg (TCE-2) and 0.0060 mg/kg (TCE-1).

Cobalt (Co) is necessary for minimal amounts in all mammals and used to treat several different types of cancer in humans and treat anaemia but the intake of the high amount can cause heart problems ⁶¹. The average concentration Co was 0.030mg/kg in

T. chebula aqueous extract. The concentration range of Co was between 0.0200 mg/kg (TCE-3) and 0.0567 mg/kg (TCE-2).

Chromium (Cr) level is low-moderate in all the analyzed samples because the soil where plants were grown has low amounts of Cr. Cr plays a vital role in carbohydrate metabolism, and its deficiency leads to diabetes in human body ⁶². Cr is a power-providing factor for many enzymatic activities. Cr deficiency imparts directly to decrease the efficiency of insulin secretion and increases sugar and cholesterol level in the blood ⁶³. Cr deficiency causes impairment in glucose tolerance factor, leading to insulin resistance. The average concentration of Cr in *T. chebula* aqueous extract is 0.3774 mg/kg, and the concentration range is found to be 0.2000 mg/kg (TCE-1) and 0.7076 mg/kg (TCE-2).

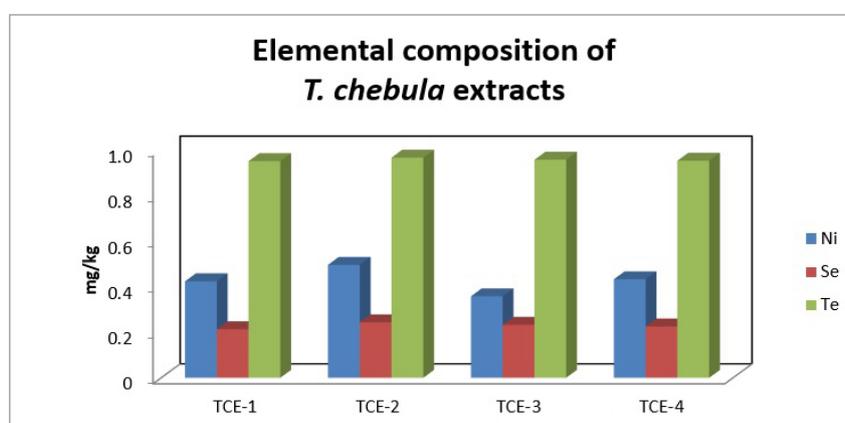


Figure 5. Elemental composition of *T. chebula* extracts (TCE-1, TCE-2, TCE-3, and TCE-4) [Ni, Se, and Te].

Nickel (Ni) is used in the formation of enzymes from bacteria ^{64,65}. Although many scientists believe that Ni is necessary for good health, this is not proven yet. People with certain liver and kidney defects are known to have low levels of Ni in their bodies. Also, excess Ni in the body is associated with a high incidence of heart disease, thyroid disease and cancer. In both of these cases, the significance of the amount of Ni needed for the body is unknown. Some scientist also believes that Ni could also affect hormones, cell membranes, and enzymes. The average concentration of Ni is 0.4276 mg/kg and the concentration range is 0.3578 mg/kg (TCE-3) and 0.4960 mg/kg (TCE-2).

Selenium (Se) is an important element, and its presence in the human body will protect the blood cells from the attack of toxic chemicals ⁶⁶. For the production of antibodies in the human body, Se will combine with vitamin E and synthesis antibodies. Selenium helps to improve the proper functioning of the heart and pancreas. It is also found useful in keeping the human tissues with elastic nature. Its deficiency in the body could be linked to the cause of leukaemia, arthritis, and other diseases. Many research data reveals that a deficient concentration of

Selenium lesser than the recommended minimum level in the bloodstream could cause a high risk of developing many types of carcinogenic problems. The mean concentration value of Se in *T. chebula* aqueous extract is 0.2286 mg/kg. The concentration of Se in the extract is between 0.2130 mg/kg (TCE-1) and 0.2436 mg/kg (TCE-2).

Tellurium (Te) has no known biological function. Te and Te-based compounds are considered to be mildly toxic and need to be handled with care, although acute poisoning is rare. Te is not reported to be carcinogenic. The average concentration of Te is 0.9573 mg/kg and the range is between 0.9514 mg/kg (TCE-1) and 0.9668 mg/kg (TCE-2).

Lead (Pb) is highly toxic metal and non-essential element for the human body due to it causes high blood pressure, kidney damage, miscarriages and subtle abortion, brain damage, declined fertility of men through sperm damage, diminished learning abilities of children and disruption of nervous systems ⁶⁷. The average concentration Pb was 0.670 mg/kg in *T. chebula* aqueous extract. The concentration range of Pb in all four samples is between 0.6323 mg/kg (TCE-3) and 0.6874 mg/kg (TCE-1).

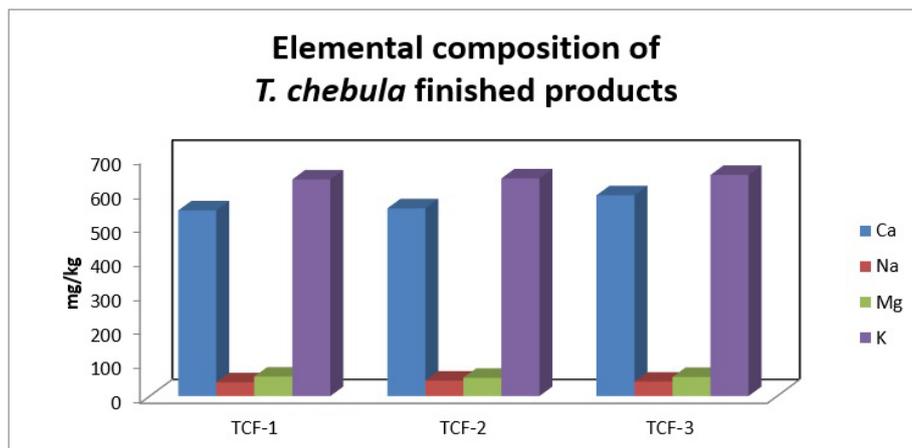


Figure 6. Elemental composition of *T. chebula* finished products (TCF-1, TCF-2, and TCF-3) [Ca, Na, Mg, and K].

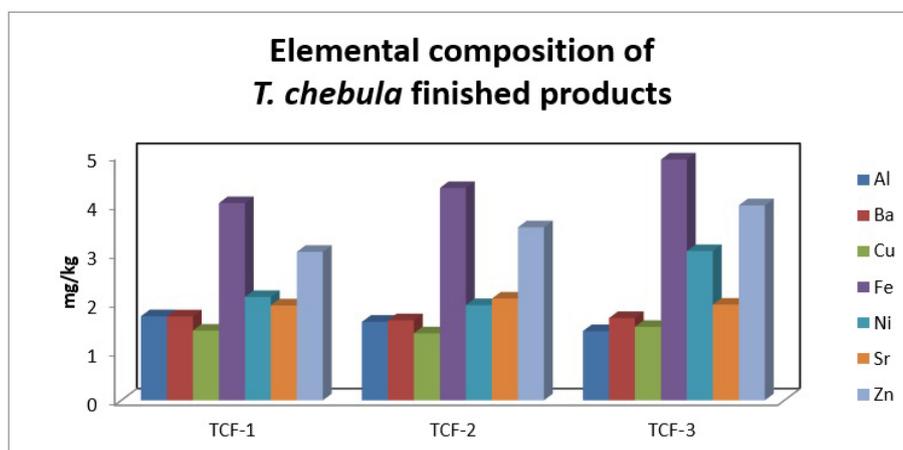


Figure 7. Elemental composition of *T. chebula* finished products (TCF-1, TCF-2, and TCF-3) [Al, Ba, Cu, Fe, Ni, Sr, and Zn].

The finished products of *T. chebula* consist of slightly different elemental composition in comparison with the aqueous extract of *T. chebula* because some of the elements are absent or shallow concentration in the finished products. Atmospheric

conditions, pollution, the season of sample collection, age of the plant, and soil conditions are the major environmental factors that could affect the concentration of elements as it varies from plant to plant and region to region.

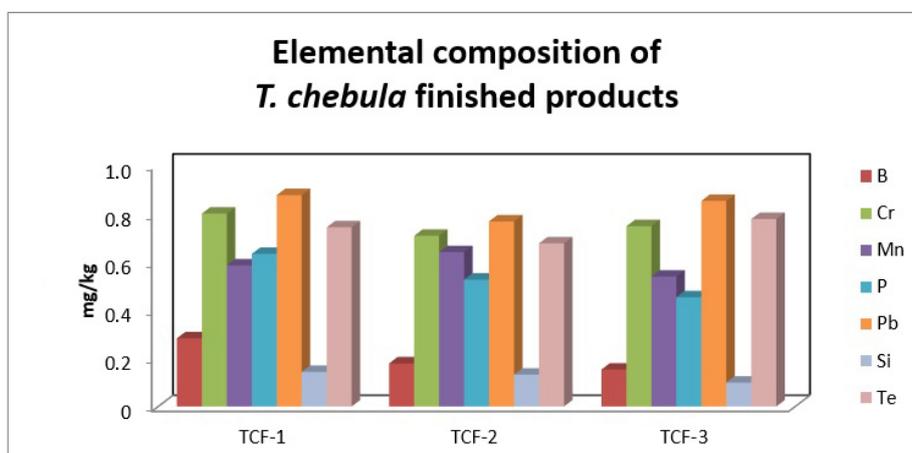


Figure 8. Elemental composition of *T. chebula* finished products (TCF-1, TCF-2, and TCF-3) [B, Cr, Mn, P, Pb, Si, and Te].

Trace elements with less toxicity in nature would play both restorative and protective roles in skirmishing diseases. Many trace elements could be well exploited for the development of preventive medicinal aspects and combating many diseases.

Conclusion

Determination of macro and microelements in *T. chebula* aqueous extracts and finished products was performed using ICP-OES. The macro elements (Ca, Mg, Na, and K) were found to be inappreciable concentration and low or fewer concentrations of toxic elements of Lead and cadmium in the extracts of *T. chebula*. Hence, the use of these extracts is considered to be safe for the finished products. The details of essential elements, toxic elements would be also helpful for the product manufacturers to specify the levels of these elements in the quality specifications. The different concentration of same elements with different *T. chebula* extracts is due to the attribution of plant collection, which in turn depends on the soil in which the plant was grown in, age of the plant, climatic conditions, the season of harvesting, etc. However, in order to develop a stronger basis for appreciating the curative effects of medicinal plants, there is a need to study the effect of soil and climatic conditions on the elemental contents of these medicinal plants. *T. chebula* extracts and finished products can be employed in the design of new ayurvedic drugs either alone or in combination with other herbal extracts for the treatment and control of many diseases.

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