PHYTOCHEMICAL, PHYSICO-CHEMICAL AND ELEMENTAL ANALYSIS ON THE ROOTS OF SHAN KOKKAYA AND INDIA KOKKAYA

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1. INTRODUCTION

- Medicinal plants -the richest bio-resource of traditional systems of medicine, modern medicines, food supplements[12].

- Organoleptic
  Phytochemical
  Physicochemical

→ **for correct identity, quality and purity of plant materials to the maintenance of health in human[14,16].**

- In Myanmar, over 80% of MTM of plant origins
- Many substituted plant materials in market

because of rareness, exploitation and high price

- At present, the root of Shan Kokkaya - as a substituted product for India Kokkaya in the market.

India Kokkaya

- cough, diarrhea and toothache[2,4].
- immunostimulating effect, antidepressant activity, aphrodisiacs, antimicrobial activity, local anaesthetic effect, insecticidal and molluscicidal effect[1].

Shan Kokkaya

- Leaves - vegetable and roots - tonic by local people in Southern Shan State in Myanmar.
- Triterpene, flavones and sterols involved in the aerial parts α-Glucosidase inhibitory and antioxidant activity[3].

- However, there was **no scientific evidence to compare** for the evaluation of authenticities, quality and purity of these two products.

Therefore, this study was carried out for the evaluation of authenticities, quality and purity of these two plant materials in the Myanmar market.

2. OBJECTIVES

General Objective

- To compare phytochemical, physicochemical and elemental properties on the root of the Shan and India Kokkaya

Specific objectives

- To identify the authenticated characteristics of these two species
- To investigate the phytochemical and physico-chemical properties of these two species
- To determine relative concentration of elemental components of the medicinally used parts
- To compare the authenticity, quality and elemental components of the roots of Shan and India Kokkaya

3. MATERIALS AND METHODS

3.1 Plant identification of Shan Kokkaya

Plant specimens(Shan Kokkaya) - collected from natural habitat in Pindaya Township, Southern Shan State during flowering and fruiting time.

Identified and classified according to morphological characters referred in Hutchinson (1967), Hooker (1879), Menglan *et al.* (2005).

3.2 Sample preparation

The roots of Shan and India Kokkaya were purchased from Zayjo market in Mandalay and prepared into powdered sample.

3.3 Organoleptic and Color Reaction by some chemical treatment

3.4 Phytochemical screening

Research Lab, UTM by Harborne (1998) and Raaman (2006) by using chemical reagents

3.5 Physico-chemical properties - Research Lab, UTM and R & D, DTM, Nay Pyi Taw by WHO (2011)

(1) Moisture content (Automatic Moisture Analyzer MAC series 110)

(2) 1% and 10% pH solution (pH meter, Horida F-51)

(3) Total ash (Digital Muffle Furnance)

(4) Acid insoluble ash

(5) Water soluble ash

(6) Solubility in water, ethanol, methanol, ethyl acetate and pet ether (Root powder + 100ml of solvent)

3.6 Elemental compositions - Wave length Dispersive X-ray Fluorescence (WDXRF, Super Mini 200, Rigaku, Japan) in **Department of Geology, University of Mandalay**

4. FINDINGS AND DISCUSSION

Shan Kokkaya

Scientific name - ***Pimpinella candolleana* Wight &Arnott**

Family - Apiaceae

India Kokkaya

Scientific name - ***Anacyclus pyrethrum* DC.**

Family - Asteraceae[8].

4. FINDINGS AND DISCUSSION

Shan Kokkaya

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India Kokkaya

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Family - Asteraceae[8].

P. candolleana Wight &Arnott from Pindaya Township

***A. pyrethrum* DC.**

Fig 4.1 Plant habitat

Fig 4.2 Dried roots of Shan and India Kokkaya from the market

Table 4.1 Organoleptic characters of two kinds of root powder

Characters	Shan Kokkaya	India Kokkaya
Color	Brownish white	Brown
Odour	Slightly aromatic	no characteristic
Taste	Hot, sweet, bitter, little pungent	Hot, pungent

In the concept of *Desana* system of Myanmar traditional medicine based on taste,

- Shan Kokkaya - '*2nd Byuhana*' (impaired internal *Pathavi* and aggreviated external *Pathavi*)
- India Kokkaya - '*Sangahita*' (aggreviated both internal and external *Pathavi*) according to *Desana* medicine

In *Ayurvedic* medicine according to *Rasaphachaka* theory[13],

Shan Kokkaya

***Rasa*(taste)** - Hot, sweet, bitter, little pungent

***Vipaka* (after effect)** - Madhura

***Guna*(properties)** - S*nigdha*(oily), *Guru*(heavy), *Mrudu*(soft);

***Virya*(potency)** - *Usna*(hot)

Dosakarma - *Tridosasamaka*

India Kokkaya [13],

***Rasa*(taste)** - Hot, pungent

***Vipaka* (after effect)** - Pungent.

***Guna*(properties)** - *Ruksa*(Dry), *Laghu*(light), *Tikshna*(sharp)

***Virya*(potency)** - *Usna*(hot)

Dosakarma - *Kaphavatasamaka*

Table 4.2 Color reaction of the sample powder after chemical treatment

S/N	Reagents	Color Reaction	
		Shan Kokkaya	India Kokkaya
1.	Treated with conc: H_2SO_4	Brown	Black
2.	Acetic acid	Golden yellow	Brown
3.	Acetic acid and conc: H_2SO_4	Black with yellow tinge	Dark brown
4.	5% iodine	Light brown	Black
5.	5% ferric chloride	Greenish yellow	Brown with yellow tinge
6.	10% Na OH	Orange	Dark Brown
7.	10% NaOH followed by a drop of 5% $CuSO_4$	Greenish blue	Greenish brown
8.	10% NaOH and a few drop of 10% lead acetate	Dark Yellow	Brown
9.	Conc: HNO_3	Reddish Brown	Orange
10	Conc: HNO_3 and NH_3	Orange	Yellow

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Table 4.3 Phytochemical constituents of the root samples

No.	Test	Extract	Test reagents	Results		References (Raaman 2006)
				Shan	India	
1.	**Alkaloids**	1% HCl	-Wagner's	-	+	Brown ppt
			-Dragendorff 's	-	+	Yellow ppt
			-Mayer's reagent	-	+	Creamy ppt
2.	Flavonoids	EtOH	Conc: HCl + Mg	+	+	Fluorescence Pink
3.	**Glycosides**	H_2O	Chloroform+10% ammonia	+	-	Pink
4.	Phenolics	H_2O	5% $FeCl_3$	+	+	Dark green
5.	Polyphenols	EtOH	10% $FeCl_3$ + 1%$K_3[Fe(CN)_6]$	+	+	Dark blue

Table 4.3 cont

No.	Test	Extract	Test reagents	Shan	India	References (Raaman 2006)
6.	**Phytosterols**	EtOH	Acetic anhydride + Conc: H_2S0_4	+	-	Color change (green)
7.	Saponins	H_2O	Distilled water	-	-	Foaming
8.	**Reducing sugar**	H_2O	Fehling A + B	+	-	Red
9.	Amino acid	H_2O	Ninhydrin	+	+	Purple
10.	Carbohydrate	H_2O	Naphthol + Conc: H_2S0_4	+	+	Red ring

Table 4.3 cont

No.	Test	Extract	Test reagents	Shan	India	References (Raaman 2006)
11.	Tannins	H_2O	Lead acetate	+	+	White ppt
12.	**Acid / Base / Neutral**	H_2O	Bromocresol green	**Base**	**Acid**	Yellow, green red–Acid Blue -Base
13.	Cyanogenetic substance	H_2O	Na pictrate paper + conc: H_2SO_4	-	-	Color change (Harborne 1998)

(+) Present (-) Absent

Therefore, it was indicated that it may be contributed to therapeutic activities with respect to phytochemical compounds involved in these two materials.

Table 4.4 Physico-chemical properties of the root samples

No.	Physico-chemical parameters	Quantity determined percentage	
		Shan Kokkaya	India Kokkaya
1.	Moisture content	6.54	7.86
2.	1% pH solution	5.68	5.17
3.	10% pH solution	5.63	5.20
4.	Total ash	18.23	8.31
5.	Acid insoluble ash	5.83	12.82
6.	Water soluble ash	93.86	40.66

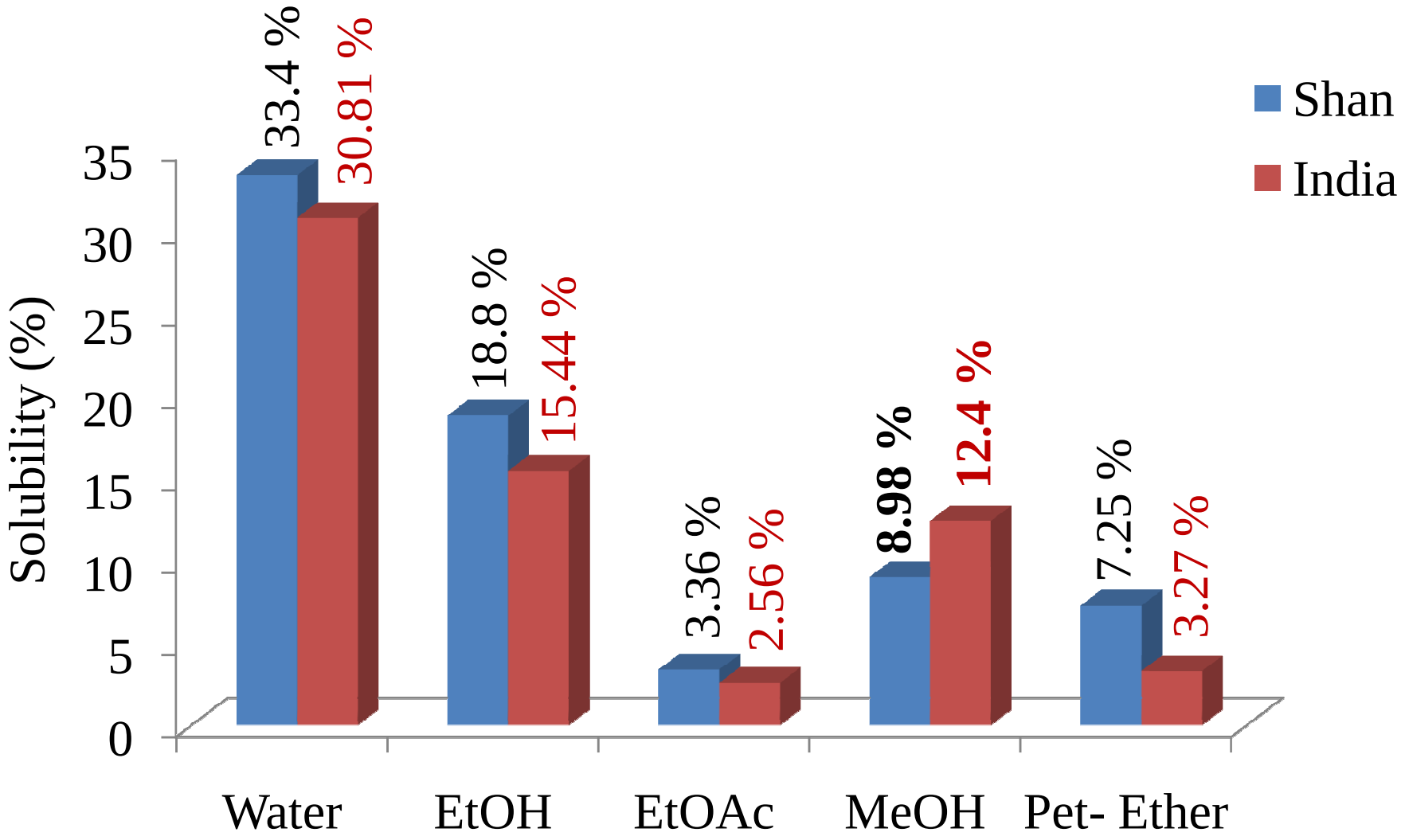

Fig 4.3 Comparison on solubility of these two powdered roots in various solvents

Physico-chemical properties were determined for the quality control parameter of medicinal purposes.

- Moisture content of Shan Kokkaya was less than India Kokkaya
- Ash values were not the same
- Solubility Shan Kokkaya is more than India Kokkaya in various solvents except methanol

Elemental components of two powdered roots

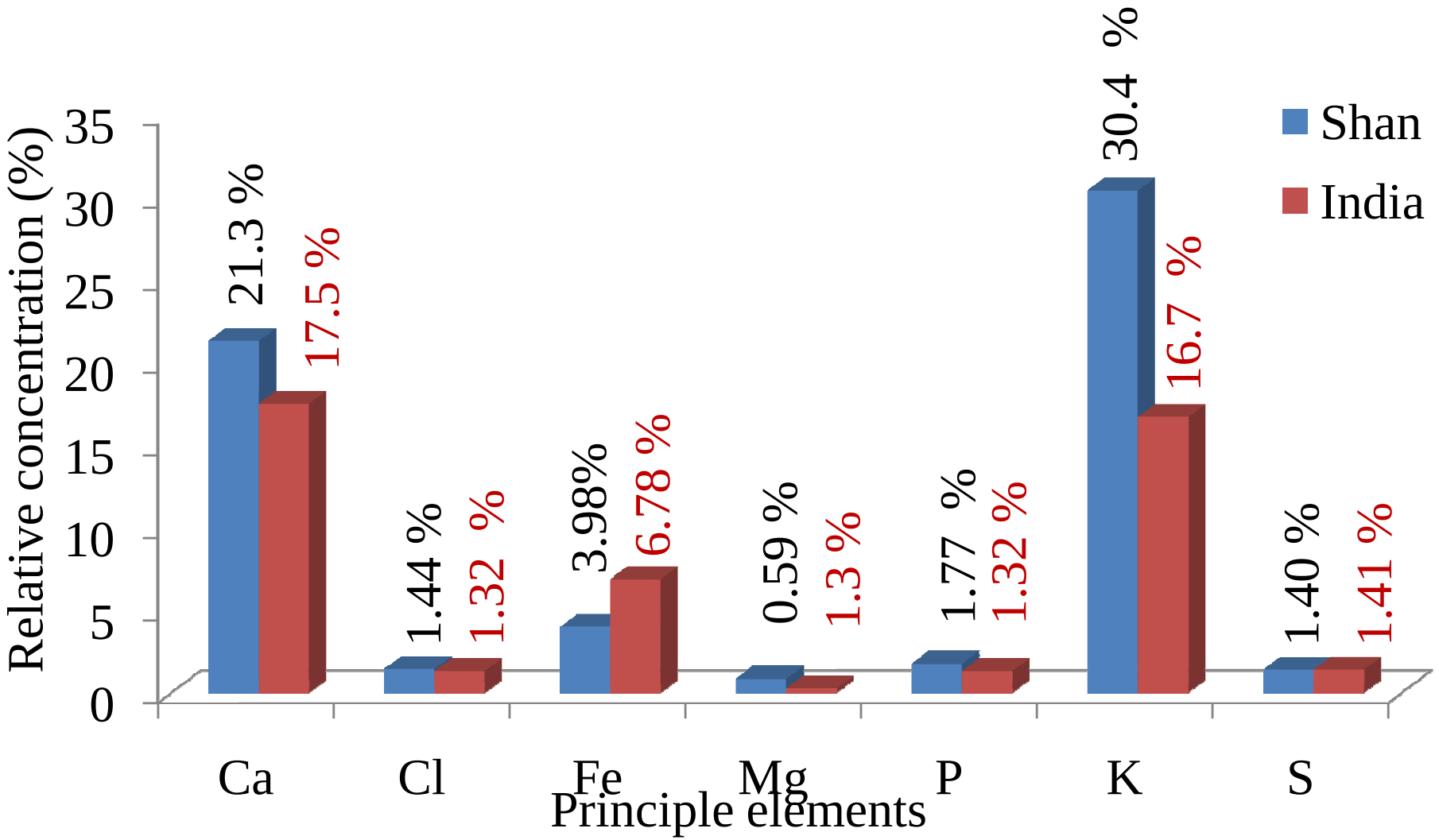

Fig 4.4 Comparison on principle elemental components of these two powdered roots

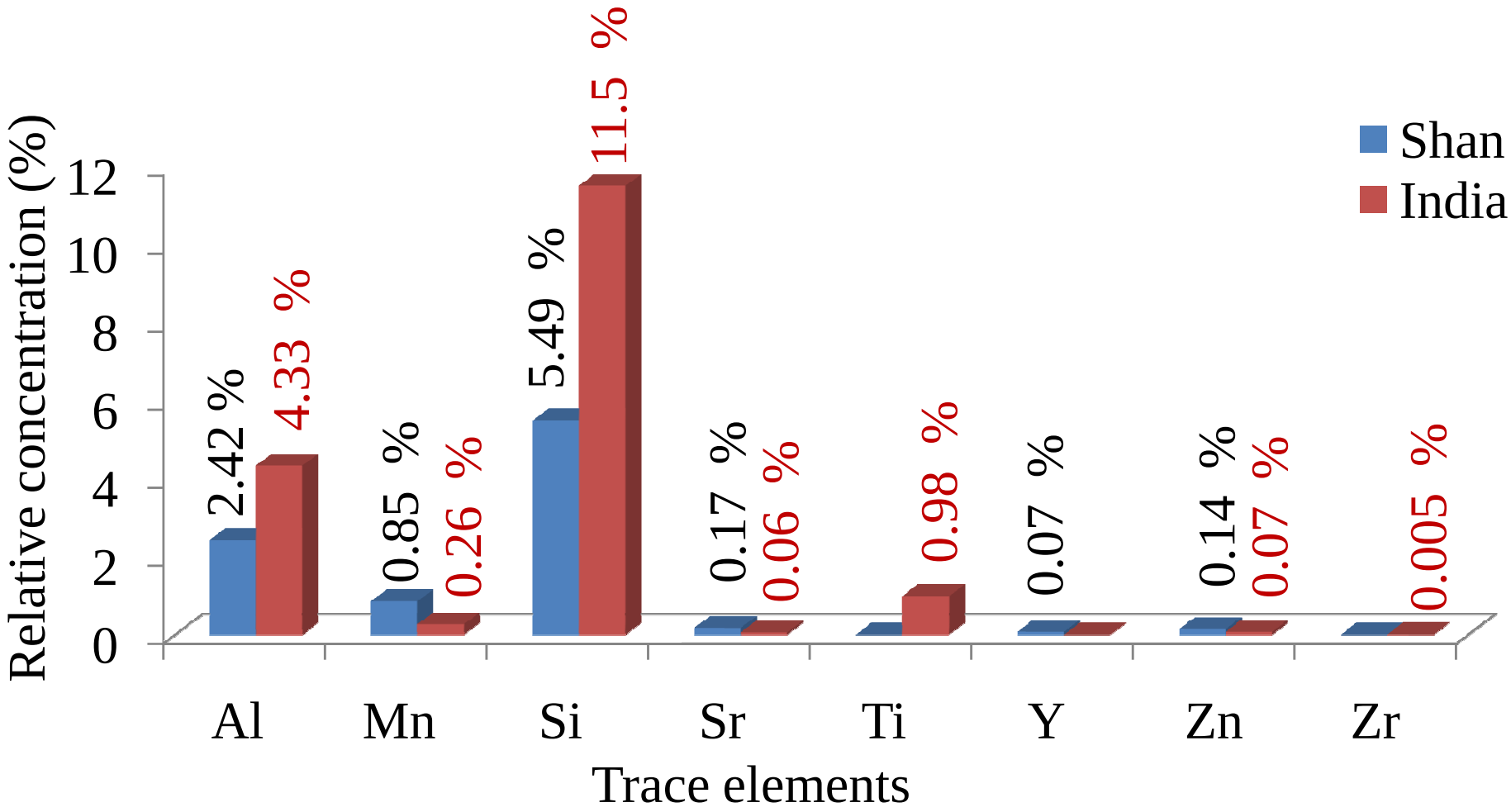

Fig 4.5 Comparison on trace elemental components of these two powdered roots

Table 4.5 Toxic element and radioisotopes content in 2 g

Element	Content in 2 g		Daily Intakes (WHO)[16]
	Shan	India	
Al	**48.4 mg**	**86.6 mg**	3-9 mg/day (UK) 2.1-8.2 mg/day(USA)
Sr	**3.44 mg**	**1.15 mg**	3 mg/day
Y	**1.4 mg**	**-**	< 1 µg/day
Ti	**-**	**19.62 mg**	8.5 mg/day
Zr	**-**	**0.108 mg**	53µg /day

- **Al is a toxic element** and the intake of large amount of Al can cause anaemia, glucose intolerance, cardiac arrest, Parkinson's and Alzheimer's diseases in human[6].
- The trace amount of radioisotopes,

  Sr and Y - Shan Kokkaya

  Sr, Ti and Zr - India Kokkaya,

The high concentration of these elements in the human body can cause toxic effect[10].

These results were pointed out that the medicinal plants for toxic metals concentration should be monitored in protecting the public from the adverse effects of these toxic metals.

5. CONCLUSION AND SUGGESTION

Desana system of Myanmar traditional medicine,

Shan Kokkaya - *'2nd Byuhana'*

India Kokkaya - *'Sangahita'*

In *Ayurvedic* medicine,

Shan Kokkaya - *Tridosasamaka*

India Kokkaya - *Kaphavatasamaka*

The pharmacological actions of Shan Kokkaya and India Kokkaya could be different due to the organoleptic characteristics and the physico-chemical properties, secondary metabolites as well as elemental compositions and relative concentrations of elements are different.

SUGGESTION

The quality, purity and adulteration of other commercial substituted plant materials should be scientifically evaluated for the effective pharmacological actions.

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