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Nutraceutical potential of North-West Himalayan spices *Allium stracheyi* and *Angelica glauca* and their comparison with commonly used spices

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Abstract

Proximate and mineral composition, phytochemical content, antinutrient content, and antioxidant activities of Himalayan spices have been analyzed for estimating their nutraceutical potential and compared with other commonly used spices. For understanding the closeness among the spices, factor analysis, and cluster analysis was carried out. The Himalayan spices were found rich in important nutraceutical components and equivalent to other commercially available spices. *Allium stracheyi* has shown far proximity, and *Angelica glauca* has shown proximity with the rest of the spices except *Syzygium aromaticum* and *Curcuma longa*. As the Himalayan spices have high nutraceutical potential, so can be used as nutraceutical as well as for fortification of food.

Keywords Himalaya · Spice · Phytochemical · Antioxidant activity · Mineral · Nutraceutical

Introduction

Spices are an integral part of the food system throughout the world, mainly used for enhancing the taste. Nowadays, people are more attracted towards the spices due to nutraceutical potential [1, 2]. These are generally used in minimal quantities in comparison to the main ingredients of food, but due to the presence of various types of phytochemicals, spices are capable of increasing the nutraceutical value of the prepared food [3, 4]. Several spices such as clove, cumin, cinnamon, curcumin, ajwain, fenugreek seeds, oregano, and thyme, etc. have the capability to treat infectious diseases due to the presence of various types of secondary metabolites. These properties have proposed a new way of the utilization of these spices as nutraceuticals.

According to Ayurveda, spices have the capacity to work as anti-inflammatory, antiarthritic, hypoglycaemic,

antithrombotic, and anti-atherosclerotic agents [5]. Several spices and their related products have been reported to have nutraceutical potential, e.g., aged garlic extract, Cumin seeds, turmeric, black pepper, clove oil, ajwain seeds, fenugreek seeds, ginger oil [6–9], etc. There are many more spices having such properties.

There are many Himalayan herbs, which are being used as spices by local people. *Allium stracheyi* and *Angelica glauca* Edgew. commonly known as Jambu and Gandhrayan, are two of those herbs [10]. Dried flowers of *Allium stracheyi* and dried roots of *Angelica glauca* have been used as spices. *Angelica glauca* Edgew. is the native and endemic species of the Indian Himalaya and distributed in the different Indian Himalayan states eg. Himachal Pradesh, Uttarakhand, Jammu & Kashmir and Sikkim with different altitudinal variation in its distribution [11]. Its roots can be harvested only after 2–3 years of cultivation. After harvesting, the roots have been dried in shade before packaging [12]. Distribution of *Allium stracheyi* is in an altitudinal range of 2500–3625 m in Jammu & Kashmir, Himachal Pradesh and Uttarakhand in Indian Himalayas along with neighbouring countries such as Pakistan and Nepal [13]. Aerial part, which is used as spice, is harvested twice/trice a year and generally shade dried before packaging [14]. According to IUCN and CAMP, 2003 *Allium stracheyi* and *Angelica glauca* are considered as vulnerable and critically endangered mainly due

Chemical compounds studied in this article Quercetin [PubChem CID: 5280343], Ascorbic acid [PubChem CID: 54670067], Tannic acid [PubChem CID: 16129778].

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to indiscriminate collection of the plants materials from wild [15–17], but their propagation protocols are being developed for making these herbs available through cultivation practices [18, 19]. Commercial production of these spices is also reported [11, 18].

These two herbs are also known for their medicinal properties like *Allium strecheyi* is useful as a stimulant, appetizer dysprosia, cordial, cardioactive, and diaphoretic, carminative, expectorant, curing stomach troubles and in treating constipation whereas *Angelica glauca* is known for healing wound, cold, cough, healing jaundice, and other stomach problems [20, 21].

The studies which have been carried out on phytochemical assessment of these species are mainly focussed on the estimation of their essential oils [22–24]. Only a few studies have been reported on the assessment of the nutritional potential and the antioxidant activities of these species [25, 26]. Present study is based on the analysis of the nutraceutical potential of *Angelica glauca* and *Allium strecheyi* in terms of their nutritional components, antinutrients, and antioxidant potential along with their comparison with other commonly used spices, readily available in the market, based on these properties.

Materials and methods

Chemicals and reagents

2,4,6-tripyridyl-1,3,5-triazine (TPTZ) (AR), methanol (HPLC), sodium carbonate (AR), ferrous chloride (AR), hexane (AR), sulphuric acid (AR), hydrochloric acid (AR), boric acid (AR), sodium Hydroxide (AR) were procured from Merck Co. (Germany) 1,1-diphenyl-2-picrylhydrazyl (DPPH) (AR), ascorbic acid (AR), gallic acid (AR), and quercetin (AR) were procured from Sigma-Aldrich (Germany).

Sample collection

The targeted spices, i.e., *Allium stracheyi* (*jambu*) and *Angelica glauca* (*gandhrain*), along with the spices used for comparison, eg. *Curcuma longa* (*haldi*), *Piper nigrum* (*kali mirch*), *Cuminum cyminum* (*jeera*), *Trigonella foenumgraecum* (*methi*), *Capsicum annum* (*mirch*), *Coriandrum sativum* (*dhania*), *Trachyspermum ammi* (*ajwain*), *Syzygium aromaticum* (*laung*) were purchased from the local market of Almora, Uttarakhand, India (FS.1). These spices were cleaned and ground to a fine powder by using a lab-based grinder and kept in airtight well-labeled boxes at room temperature until analyzed.

Extraction procedure

1.5 g grounded spice samples were macerated in an ambient conditions using 75 ml of 85% methanol (1:5 w/v) on the rotary shaker (Remi, India) until the clear extract is obtained, with an interval of 24 h. The extracts collected after each 24 h were filtered through filter paper and mixed subsequently. The extracts were stored at $-20\text{ }^{\circ}\text{C}$ in a refrigerator until analyzed.

Estimation of proximate and mineral composition

Moisture content, ash content, fat, crude fiber of the ground samples was determined following AOAC, 2016 method. Total carbohydrate content was determined using the Anthrone Method [27], and total protein was determined using the Lowry method [28]. For mineral estimation, spice samples were digested using Allen's method [29]. 0.3 g of ground sample was taken to which 4.4 mL of digestion mixture (0.42 g Se powder and 14 g Lithium sulphate monohydrate in 350 mL H_2O_2 and 420 mL H_2SO_4 ice cold) was added. Samples, as well as blank sample, were digested at $360\text{ }^{\circ}\text{C}$ for two hours. Further, the digested samples were used for mineral analysis. Actual mineral content was estimated by subtracting blank value.

Sodium and potassium were analyzed through flame photometer (Systronic flame photometer 128); phosphorus content was determined using the colorimetric method following Murphy and Riley [30], and iron content was determined colorimetrically following AOAC, 2016 phenanthroline method [31] using UV–Vis spectrophotometer (Shimadzu UV-2600).

Determination of total phenolic, flavonoid and antinutrient content

Total phenolic contents in the methanolic extract of spices were determined using Folin-Ciocalteu's colorimetric method [32]. Briefly, 250 μL of methanolic extract was diluted with 2250 μL of distilled water. Thereafter 250 μL Folin-Ciocalteu's reagent was added and allowed to stand for 5 min. The mixture was neutralized by sodium carbonate (7%) and kept in the dark at room temperature for 90 min. The reaction results in the appearance of a blue color, which was measured at 765 nm using a UV–Vis spectrophotometer. A standard curve was prepared using Tannic acid standard solution prepared in 85% methanol which was further used for the quantification of total phenolic content present the spice extracts. The results were presented in milligram tannic acid equivalent per gram dry weight (mg TAE/g DW).

Total Flavonoid Content in the methanolic extract of spices was determined following a method developed by

Zhishen et al. [33], with some modifications [34]. For the determination of total flavonoid content, 500 μL of methanolic extracts were diluted with 2000 μL of distilled water. Thereafter 150 μL of sodium nitrite was added and allowed to stand for 5 min. 150 μL of 10% aluminum chloride was added, and the mixture was shaken. After waiting for 6 min. 1000 μL sodium hydroxide was added. The mixture was again shaken and incubated for 15 min. The absorbance was measured at 510 nm. Quantification of total flavonoid content was done based on a calibration curve of quercetin standard solution prepared using 85% methanol, and results were expressed in milligram quercetin equivalent per gram dry weight (mg QE/g DW).

Antinutrients such as tannins, phytic acid, and oxalate content were determined for all the target spices using the modified method of Sadasivam and Manickam [35], originally described by Price et al. [36], Wheeler and Ferrel [37] and Abaza et al. [38] respectively. Calibration curve of catechin standard solution prepared in 85% methanol was used for quantification of tannin content and it was expressed as milligram catechin equivalent per gram dry weight (mg CE/g DW). For analyzing the effect of phytic acid on the bioavailability of iron, molar ratio of phytate to iron (Phy:Fe) was calculated using Eq. 1:

$$\text{Molar ratio} = \frac{\text{Moles of antinutrients}}{\text{Moles of minerals}}. \quad (1)$$

Molar mass of 660 g/mol was used for phytate.

Determination of vitamin C content

Vitamin C content of spices was determined by the method given by Roe and Oesterling [39] with some modifications. In this method, 75 μL of DNPH (2 g dinitrophenyl hydrazine, 230 mg thiourea and 270 mg $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ in 100 mL of 5 mol L^{-1} H_2SO_4) were added to 500 μL reaction mixture (300 μL of the methanolic extract with 100 μL 13.3% TCA and water). The reaction mixtures were then incubated for 3 h at 37 °C, and 500 μL of 65% H_2SO_4 (v/v) was subsequently added to the medium; absorbance was taken at 520 nm. Vitamin C content was quantified based on the standard curve of ascorbic acid solution prepared in 85% methanol, and results were expressed in milligram ascorbic acid equivalent per gram dry weight (mg AAE/g DW).

Determination of antioxidant activity

DPPH radical scavenging ability assay

DPPH free radical scavenging activity was measured following the method given by Blois with modifications [40]. For the measurement of free radical scavenging activity

of the sample, 2500 μL of methanolic extract was taken in test tubes to which 2500 μL of 0.3 mM DPPH was added. The mixture was incubated for 20 min at room temperature. Absorbance was measured at 517 nm. A standard curve was prepared with ascorbic acid. Final results were given as % IC (Inhibition Concentration).

FRAP antioxidant assay

Ferric reducing antioxidant power (FRAP) assay was performed following Benzie and Strain [41] with slight modifications. FRAP reagent was prepared by adding 10 volumes of 0.3 M acetate buffer, 1 volume of 10 mM TPTZ in 40 mM HCl, and 1 volume of 20 mM ferric chloride. The 3600 μL of FRAP reagent was added to 1000 μL of methanolic extract and incubated at 37 °C for 10 min. Absorbance was taken at 593 nm. A standard curve was prepared with ascorbic acid in 85% methanol. Final results were given as milligram ascorbic acid equivalent per gram dry weight (mg AAE/g DW).

RP-HPLC analysis

HPLC analysis was carried out using Waters 2695 separations module equipped with an inline degasser, a binary pump delivery system and a Waters 2996 photodiode array detector, connected to a reverse phase C-18 column (4.6 \times 250 mm Analytical Column Waters @ SPHERISORB @ 5 μm OD 52). Post chromatographic analysis was carried out using "Empower Software System" (Waters Corp., Milford, USA). All the solutions, including sample extract and solvents, were filtered through 0.45 μm membrane filter prior analysis. The amount of flavonoid and phenolic compounds was quantified using RP-HPLC at ambient temperature. The mobile phase consists of 0.05% orthophosphoric acid and acetonitrile (45:55 mix ratio). An injection volume of the sample used was 10 μL . The flow rate was kept at 1 mL/min during analysis with the total run time of 15 min. Quercetin, tannic acid and ascorbic acid were used as standards for quantification of phenolic and flavonoid compounds present in the samples under study. All these compounds were analyzed at their maximum wavelength (λ_{max}), i.e., 255, 269, 244 nm for quercetin, tannic acid, and ascorbic acid, respectively (Table 3). All the samples were analyzed at these λ_{max} for getting the exact concentration of these compounds.

Statistical analysis

All the experiments were performed in triplicates. Basic statistical analysis was carried out using MS excel, while Factor analysis (FA) using Principal components (PCs) and cluster analysis (CA) was carried out using STATISTICA 8 for understanding dominating factors affecting variability

among the targeted spices and understanding similarity of targeted Himalayan herbs with other commonly used spices. FA was done to extract information on the correlation among the variables analyzed for spice samples. Before multivariate analysis, data was normalized through log transformation and then z-score estimation for minimizing the effect of different variables used in the study and to adjust the disparity caused due to measurement units. Factor scores were obtained from standardized variable data. Varimax rotation was used to minimize the variance of squared loading. Kaiser's criterion was used for the estimation of a number of factors which need to be retained. For investigating the similarity among the spices, cluster analysis was done using normalized data, the ward's method was used for linkage study and Euclidian distances as a measure of similarity [42]. The result is presented as a dendrogram for presenting the clusters and their proximity [43].

Result and discussion

Proximate and mineral analysis

Moisture content, ash content, total fat, crude fiber, protein, the carbohydrate content of investigated spices is shown in Table 1. Spice compositional data obtained was compared with that of USDA (TS1) [44]. Except for *Curcuma longa* and *Syzygium aromaticum*, moisture content was found closer to recommended values of respective spices. The highest moisture content was found in *Syzygium aromaticum*, whereas the lowest was found in *Coriandrum sativum*. Low moisture content leads to a longer shelf life of spice/food samples as bacterial and mold growth is inhibited due to lower moisture availability. Because of this, spices can be stored for a longer period. The moisture content of

Himalayan herbs *Allium stracheyi* and *Angelica glauca* was found at the lower side.

Ash contents vary much among the spices, which was varying from $11.79 \pm 0.14\%$ for *Allium stracheyi* to $3.14 \pm 0.15\%$ for *Trigonella foenumgraecum*. The ash content of most of the spices were found closer to the USDA recommended value except in *Capsicum annum* and *Trachyspermum ammi* (Table 1 & TS 1). Total ash content depicts the presence of mineral constituents in the sample; if it is low in case of spices, then it is believed that these spices are previously extracted [45]. Along with this, differences can also be observed due to the variations in different climate factors such as soil and weather conditions where the plants are growing. There are no prescribed values of ash content yet for spices used out of *Allium stracheyi* and *Angelica glauca* plant parts, and the current study has found it at the level of 11.13 and 4.64% respectively.

Total fat content was found lowest in *Trigonella foenum graecum* ($5.45 \pm 0.24\%$), and the highest was found in *Syzygium aromaticum* ($23.39 \pm 0.17\%$). Values of fat contents for most of the spices were found different from standard values recommended by USDA (Table 1 and TS1), while for *Allium stracheyi* and *Angelica glauca*, it was found at the level of 13.2 and 9.5% respectively. Crude Fibre content was found, varying from $5.57 \pm 0.28\%$ (*Curcuma longa*) to $47.46 \pm 1.81\%$ (*Syzygium aromaticum*). Protein level was found on the lesser side in comparison to USDA rules. The highest protein content is $8.08 \pm 0.03\%$ was found in *Syzygium aromaticum*, and the lowest was in *Curcuma longa* ($0.11 \pm 0.00\%$). The difference in protein content can be due to the seasonal variation or time, geographical distribution, or soil fertility. Since low nitrogen-containing soils can influence protein levels reported by Blumenthal et al. [46]. Total carbohydrate contents of spices were highest among all the proximate parameters in all the selected spices and are found closer to USDA recommended values for total

Table 1 Proximate composition and crude fibre content in the spices

| Spice name | Total moisture (%) | Total ash (%) | Total fat (%) | Total crude fiber (%) | Total Protein (%) | Total carbohydrate (%) |
|---------------------------------|--------------------|------------------|------------------|-----------------------|-------------------|------------------------|
| <i>Allium stracheyi</i> | 11.13 ± 0.08 | 11.79 ± 0.14 | 4.64 ± 0.30 | 13.20 ± 0.29 | 6.29 ± 0.18 | 64.92 ± 7.07 |
| <i>Angelica glauca</i> | 9.86 ± 0.26 | 4.64 ± 0.10 | 6.60 ± 0.35 | 9.51 ± 0.31 | 1.48 ± 0.27 | 67.38 ± 2.54 |
| <i>Curcuma longa</i> | 18.03 ± 0.29 | 7.16 ± 0.12 | 13.76 ± 0.19 | 5.57 ± 0.28 | 0.11 ± 0.09 | 68.56 ± 1.94 |
| <i>Piper nigrum</i> | 12.84 ± 0.28 | 4.12 ± 0.10 | 9.33 ± 0.13 | 13.46 ± 0.15 | 0.44 ± 0.18 | 69.57 ± 0.53 |
| <i>Cuminum cyminum</i> | 11.02 ± 0.02 | 7.20 ± 0.15 | 11.59 ± 0.13 | 25.47 ± 0.42 | 3.5 ± 0.55 | 56.43 ± 1.76 |
| <i>Trigonella foenumgraecum</i> | 9.85 ± 0.23 | 3.14 ± 0.15 | 5.45 ± 0.24 | 9.02 ± 0.15 | 1.58 ± 0.42 | 61.25 ± 0.40 |
| <i>Capsicum annum</i> | 8.32 ± 0.26 | 4.71 ± 0.08 | 18.64 ± 0.15 | 42.18 ± 0.44 | 3.72 ± 0.24 | 59.82 ± 4.94 |
| <i>Coriandrum sativum</i> | 7.75 ± 0.37 | 6.19 ± 0.10 | 9.22 ± 0.15 | 37.85 ± 1.77 | 1.86 ± 0.37 | 67.78 ± 3.35 |
| <i>Trachyspermum ammi</i> | 11.47 ± 0.35 | 8.65 ± 0.07 | 13.71 ± 0.18 | 24.03 ± 0.74 | 4.17 ± 0.09 | 63.61 ± 3.53 |
| <i>Syzygium aromaticum</i> | 23.69 ± 0.59 | 6.78 ± 0.13 | 23.39 ± 0.17 | 47.46 ± 1.81 | 8.08 ± 0.58 | 63.46 ± 1.33 |

carbohydrate (Table 1 and TS1). In the case of *Allium stracheyi* and *Angelica glauca* it was also found in the range similar to other spices, i.e., 64.92 ± 7.07 and $67.38 \pm 2.54\%$ respectively (Table 1 and TS1).

Sodium, potassium, phosphorus, nitrogen and iron contents were analyzed in selected spices (Table 2). Phosphorus was found as the most abundant mineral in almost all the spices under study ranging from 109.49 ± 2.91 mg/100 g for *Syzygium aromaticum* to 687.78 ± 0.67 mg/100 g for *Allium stracheyi*. Sodium values ranged from 56.22 ± 6.61 mg/100 g (*Angelica glauca*) to 240.94 ± 6.34 mg/100 g (*Cuminum cyminum*). The values were found higher in comparison to USDA recommended values except for *Syzygium aromaticum*. Potassium content was found to vary between 872.11 ± 26.65 mg/100 g (*Trigonella foenum graecum*) to 4260 ± 106.80 mg/100 g (*Allium stracheyi*). Potassium content for most of the spices under study was found closer to USDA recommended values (Table 2 and TS1). Iron values varied between 19.46 ± 0.67 mg/100 g (*Syzygium aromaticum*) to 212.8 ± 2.31 mg/100 g (*Trachyspermum ammi*). Iron contents were found higher than the level reported in USDA in most of the studied spices. Results from this study showed that spices are a good source of phosphorus, sodium, potassium, nitrogen, iron; hence their consumption is beneficial for human health, and these spices increase the health quotient of the food.

Total phenolics, total flavonoids, and antinutrient content

Total phenol content in *Syzygium aromaticum* was found highest, i.e., 72 mg TAE/g DW and least in *Coriandrum sativum*, i.e., 4.95 mg TAE/g DW. Total Flavonoid content was found highest for *Curcuma longa* i.e., 47.11 mg QE/g DW, and lowest for *Capsicum annum* i.e., 0.16 mg

QE/g DW (Fig. 1a, b). Lower Quercetin value in red chilli has been reported by the US Department of Agriculture, i.e., 0.14 mg/g [47]. The values of phenolic and flavonoid contents in *Angelica glauca* found at higher side i.e., 5.92 ± 0.20 mg TAE/g DW and 6.22 ± 0.11 mg QE/g DW respectively (Fig. 1a, b) in comparison to the values reported by Rawat et al. [48] i.e., 1.03 ± 0.1 mg TAE/g DW and 4.02 ± 0.2 mg QE/g DW respectively measured in the control samples which were reported to be collected from Tungnath, Chopta, Rudraprayag district, Uttarakhand, India.

Antinutrient such as phytic acid was found varying between a range of 3.09 to 5.33%, tannic acid in the range of 0.09 to 2.58% and oxalate in the range of 0.06 to 0.94% among the spices (Fig. 2). Highest phytic acid, tannic acid, and oxalate content were present in *Coriandrum sativum*, *Cuminum cyminum*, and *Syzygium aromaticum*, respectively. Phy:Fe was found more than 1 in all the spices (Table 3), as recommended by Hallberg et al. [49], which means iron present in spices are in available form and is not affected by the presence of phytic acid.

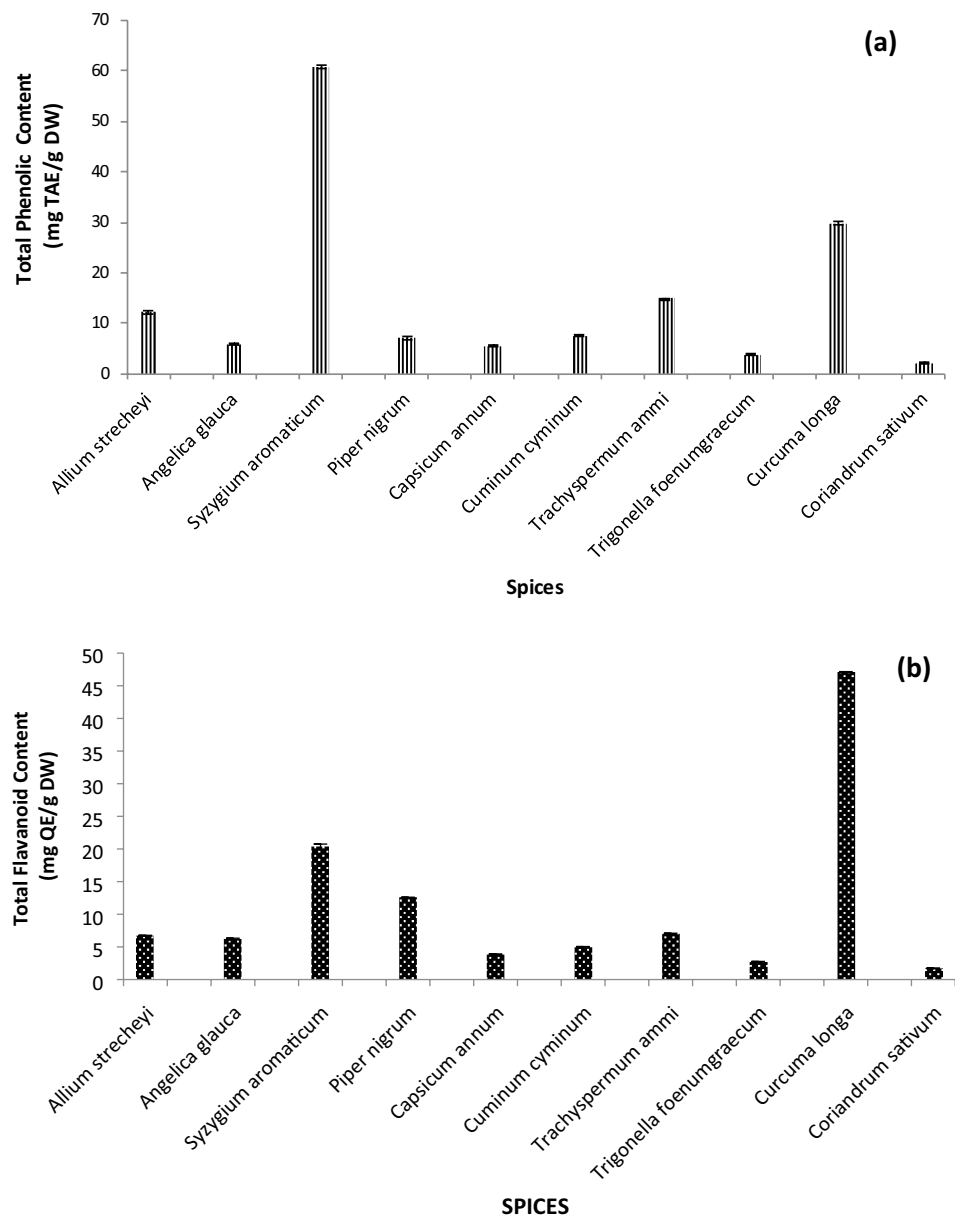
FRAP and DPPH assay

Antioxidant activities of the selected spices against FRAP and DPPH is shown in Fig. 3a, b respectively. Among FRAP activity of spices, it was found highest for *Curcuma longa* (6.13 mg AAE/g DW) and lowest for *Trigonella foenum graecum* (1.55 mg AAE/g DW). It was found positively correlated with phenolic and flavonoid content for all the spices, although the strong correlation was observed with flavonoids ($p < 0.05$) (TS 1). The DPPH scavenging activity was found, ranging from 0.3 mg AAE/g DW for *Curcuma longa* to 0.96 mg AAE/g DW for *Capsicum sativum* and *Trachyspermum ammi* (Fig. 3b). DPPH activity was not found correlated with flavonoid and phenolic content (TS 1

Table 2 Mineral composition (mg/100 g) of the spices

| Spice name | Sodium (Na) (mg/100 g) | Potassium (K) (mg/100 g) | Phosphorus (P) (mg/100 g) | Nitrogen (N) (mg/100 g) | Iron (Fe) (mg/100 g) |
|----------------------------------|---------------------------|-----------------------------|------------------------------|----------------------------|-------------------------|
| <i>Allium stracheyi</i> | 56.66 ± 5.16 | 4260 ± 106.80 | 687.78 ± 0.67 | 4.97 ± 0.06 | 93.46 ± 2.30 |
| <i>Angelica glauca</i> | 56.22 ± 6.61 | 1074.88 ± 66.29 | 258.73 ± 8.42 | 1.26 ± 0.06 | 86.13 ± 4.16 |
| <i>Curcuma longa</i> | 60.33 ± 0.86 | 2898.33 ± 31.22 | 560.25 ± 6.59 | 1.55 ± 0.08 | 28.13 ± 13.01 |
| <i>Piper nigrum</i> | 57.55 ± 7.42 | 1068.33 ± 3.48 | 269.6 ± 6.22 | 2.73 ± 0.21 | 72.8 ± 2 |
| <i>Cuminum cyminum</i> | 240.94 ± 6.34 | 1629.94 ± 11.92 | 416.06 ± 6.63 | 4.08 ± 0.05 | 76.13 ± 2.30 |
| <i>Trigonella foenum graecum</i> | 160.16 ± 5.26 | 872.11 ± 26.65 | 207.22 ± 13.96 | 5.69 ± 0.25 | 41.46 ± 3.05 |
| <i>Capsicum annum</i> | 60 ± 2.02 | 1616.61 ± 19.53 | 510 ± 13.82 | 3.30 ± 0.03 | 27.46 ± 1.15 |
| <i>Coriandrum sativum</i> | 63.55 ± 2.33 | 1215.72 ± 24.91 | 508.49 ± 18.37 | 3.01 ± 0.34 | 48.13 ± 1.15 |
| <i>Trachyspermum ammi</i> | 119.05 ± 6 | 1434.11 ± 12.22 | 374.14 ± 5.91 | 4.32 ± 0.25 | 212.8 ± 4 |
| <i>Syzygium aromaticum</i> | 189.5 ± 1.89 | 1473.72 ± 8.42 | 109.49 ± 2.91 | 1.54 ± 0.10 | 19.46 ± 1.15 |

Fig. 1 a Total phenolic, and **b** total flavanoid content in spices

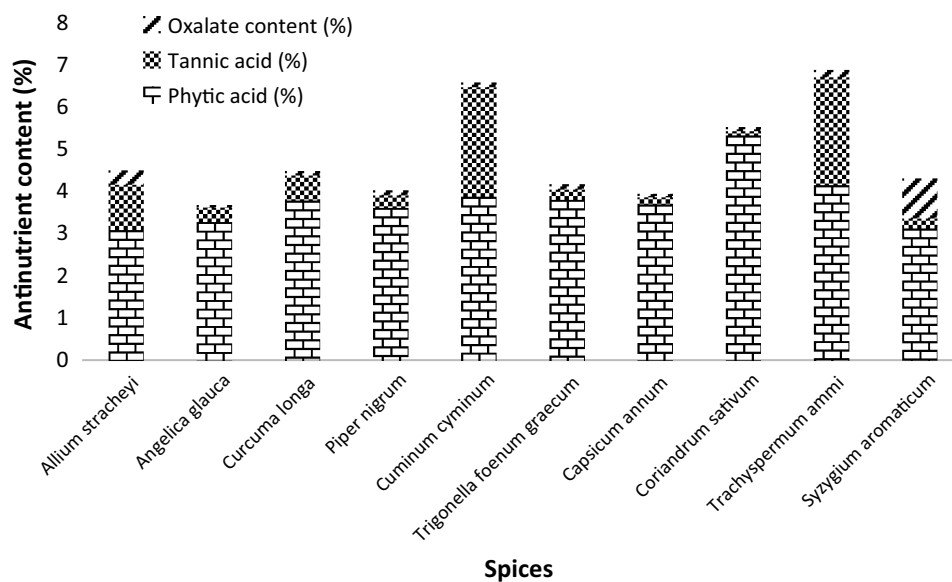


& FS 2). This can also be understood by order of percent (%) activity of spice extract volume which is *Syzygium aromaticum* > *Curcuma longa* > *Trachyspermum ammi* > *Angelica glauca* > *Cuminum cyminum* > *Allium strecheyi* > *Coriandrum sativum* > *Trigonella foenum graecum* > *Piper nigrum* ≥ *Capsicum sativum* (FS. 3(a) and (b)). Very low extract volume (45 and 13 μ L respectively) was found enough for 50% inhibition of antioxidants in the case of *Curcuma longa* and *Syzygium aromaticum* (TS 3). Goswami et al. [50] had also observed high DPPH radical scavenging activity for

Curcuma longa, *Trachyspermum ammi*, *Cuminum cyminum*, *Coriandrum sativum* and *Piper nigrum* among twenty spices purchased from Siliguri local market which mean these spices have high antioxidant potential.

RP-HPLC analysis

The contents of quercetin, tannic acid, and ascorbic acid in all the spices were also quantified using RP-HPLC (Table 4 and Fig. 4). Among all the selected phenolic and

Fig. 2 Antinutrient content in spices

flavonoids, ascorbic acid was recorded in higher concentrations, followed by tannic acid and quercetin. HPLC analysis reveals that most abundant ascorbic acid content was found in *Angelica glauca* (2822.84 ± 129.15 mg/100 g), whereas *Capsicum annum* contains second-highest amount of ascorbic acid, i.e., 364.48 ± 16.94 mg/100 g. These values of ascorbic acid are found higher than different varieties of *Capsicum annum* grown in the southeast Anatolia region [51]. *Cuminum cyminum* contains higher tannic acid, i.e., 9.77 ± 0.04 mg/100 g among all the spices. This value was found to be lower than the value of tannic acid reported by Allaithy [52] in methanolic extract of the cumin seeds, i.e., 5.20 mg/100 g (Table 4). Highest quercetin content was found in *Trigonella foenum graecum*, i.e., 1.91 ± 0.046 mg/100 g. Quercetin was not found in *Cuminum cyminum*; similar result was reported by Allaithy [52] with methanolic extracts of cumin seeds which were collected from the local market at Kerbala city. In *Curcuma longa*, 0.18 ± 0.02 mg/100 g quercetin was found, whereas Alejandro et al. [53] had not found any quercetin content in turmeric obtained from producers in the State of Sao Paulo, Brazil.

Multivariate analysis

Factor analysis-based correlation matrix was developed for understanding dominant chemical characteristics in the form of factors which are affecting the differences among the spices (Table 5). A scree plot was used to identify the

numbers of factors on the basis of eigen values (> 1) [54]. Five factors were obtained on with the total variance of 88.49% among all the analyzed parameters for the targeted spices. The factor 1 was responsible for 36.91% of total variance and showed high positive loading (> 0.70) for FRAP activity, TPC, TFC, and total moisture content which shows that these parameters are strongly affecting the variation among the spices. This also depicts that total phenolic and flavonoid contents are responsible for FRAP activity. High negative loading for iron content depicting its lower concentration with high correlation among all the spices. The flavonoid and phenolic contents are mainly responsible for FRAP activity in all spices. This also depicts that TPC and TFC are not responsible for DPPH activity, which is like the actual finding shown in TS 1. Factor 2 is accounting for 19.57% of total variance with negative loading for crude fiber and protein while positive loading for carbohydrate. Factor 3 accounts for 15.53% of total variance with positive loading for total ash, potassium content, and antinutrient tannic acid. It shows that potassium is the major component of ash, and these are dominant parameters. Factor 4 accounts for 9.82% of total variance with negative loading for phytic acid and positive loading for oxalate content. Factor 5 accounts for 6.65% of total variance with high loading for phosphorus content.

Through cluster analysis, it was observed that all the studied spices could be categorized into two main clusters with less difference in D_{link}/D_{max} value (Fig. 5). *Curcuma longa* and *Syzygium aromaticum* have made separate cluster (C1)

Fig. 3 a Ferric-reducing and b DPPH radical scavenging activity of spices

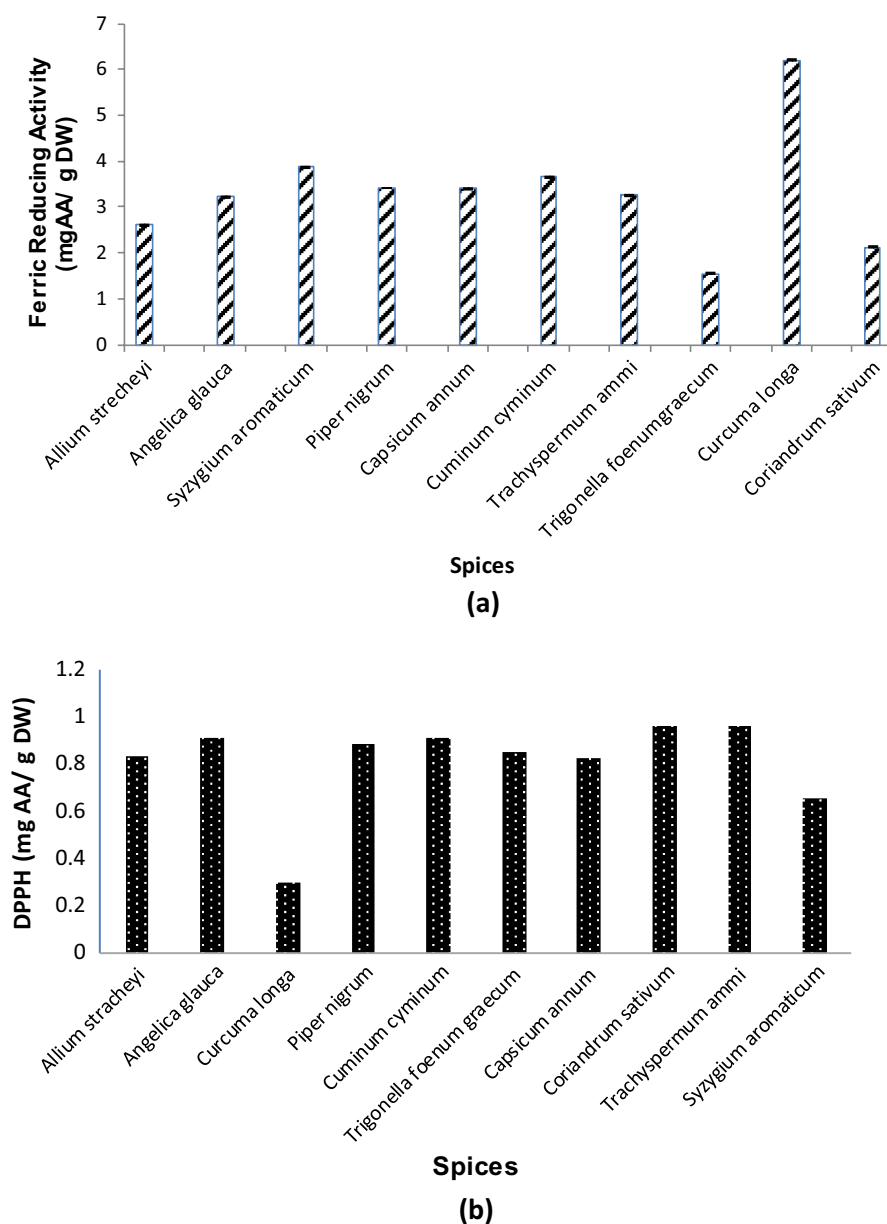


Table 3 Phytic acid and iron ratio for the spices

| Spices | PA/Iron \pm SE |
|----------------------------------|------------------|
| <i>Allium stracheyi</i> | 2.81 \pm 0.04 |
| <i>Angelica glauca</i> | 3.24 \pm 0.11 |
| <i>Curcuma longa</i> | 13.61 \pm 4.21 |
| <i>Piper nigrum</i> | 4.22 \pm 0.07 |
| <i>Cuminum cyminum</i> | 4.33 \pm 0.10 |
| <i>Trigonella foenum graecum</i> | 7.82 \pm 0.22 |
| <i>Capsicum annuum</i> | 11.44 \pm 0.19 |
| <i>Coriandrum sativum</i> | 9.40 \pm 0.10 |
| <i>Trachyspermum ammi</i> | 1.66 \pm 0.01 |
| <i>Syzygium aromaticum</i> | 13.69 \pm 0.48 |

while rest of the spices were clustered in another cluster (C2). Similarity between *Curcuma longa* and *Syzygium aromaticum* is well justified by observing the chemical characteristics of both the spices (Tables 1, 2, 3, and 4 and Figs. 1, 2, and 3). C2 is further grouped into two sub-clusters, where the Himalayan spices *Allium stracheyi* had made separate sub-cluster, and the rest of the spices came in another sub-cluster. *Angelica glauca* has shown proximity with *Piper nigrum* and comparatively far proximity with other remaining spices on the basis of chemical parameters under study.

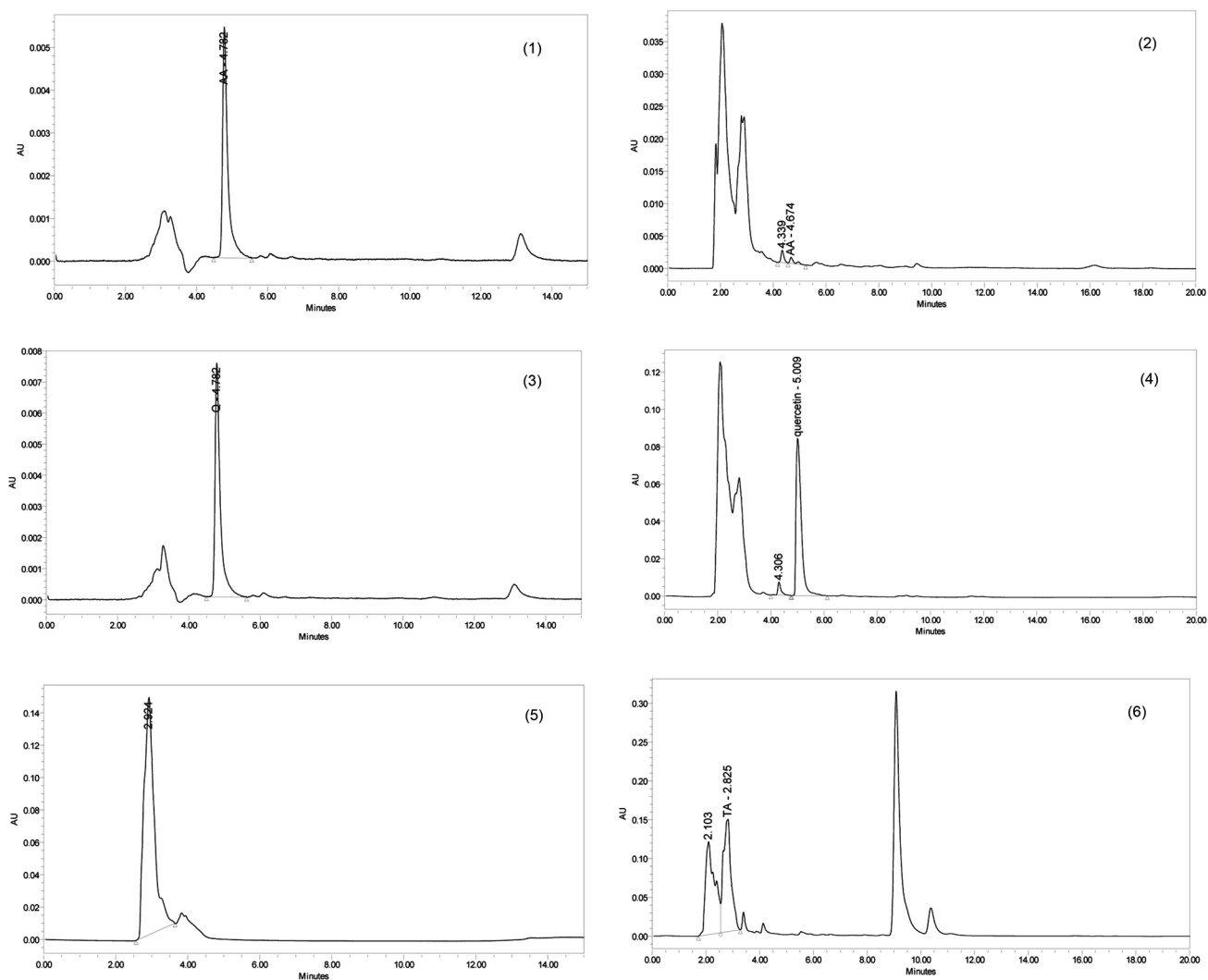


Fig. 4 RP-HPLC Chromatogram of standards (1) Ascorbic acid, (3) Quercetin, (5) Tannic acid and their respective presence in spice samples (2) *Coriandrum sativum* (4) *Trigonella foenum graecum* (6) *Cuminum cyminum*

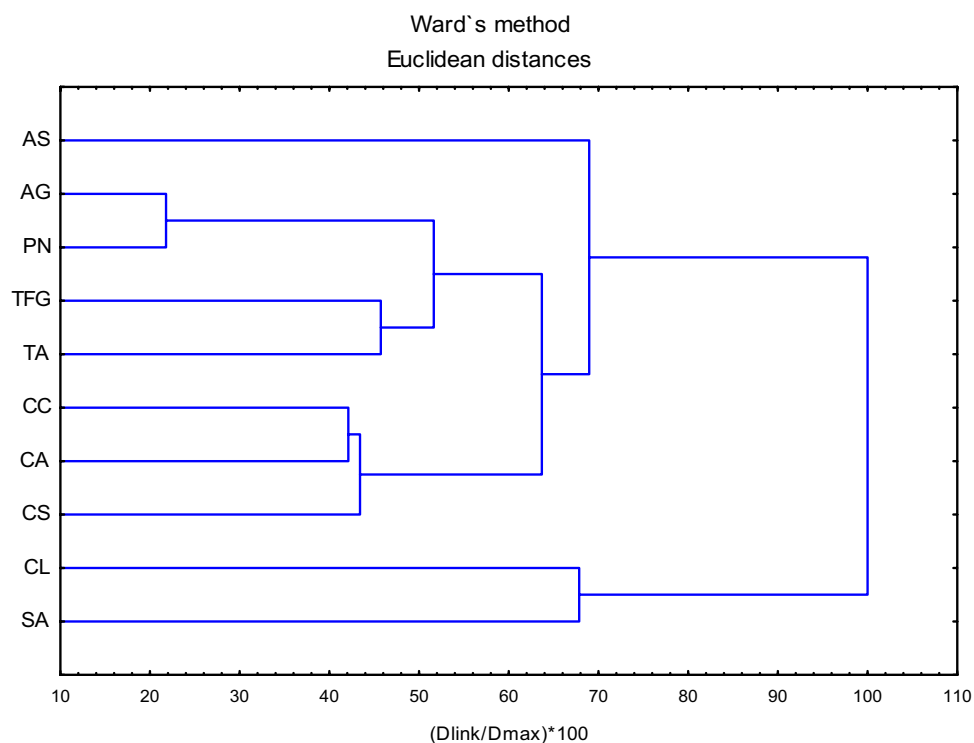
Table 4 Quantification of ascorbic acid, quercetin and tannic acid through RP-HPLC

| Spices | Ascorbic acid (mg/100 g) | Quercetin (mg/100 g) | Tannic acid (mg/100 g) |
|---------------------------------|--------------------------|----------------------|------------------------|
| <i>Allium stracheyi</i> | 82.24 ± 3.21 | 0.156 ± 0.005 | 1.97 ± 0.01 |
| <i>Angelica glauca</i> | 2822.84 ± 129.15 | 0.36 ± 0.05 | 7.76 ± 0.21 |
| <i>Curcuma longa</i> | BDL | 0.18 ± 0.02 | 2.35 ± 0.06 |
| <i>Piper nigrum</i> | BDL | 0.056 ± 0.001 | 0.47 ± 0.04 |
| <i>Cuminum cyminum</i> | BDL | BDL | 9.77 ± 0.04 |
| <i>Trigonella foenumgraecum</i> | BDL | 1.91 ± 0.046 | 1.41 ± 0.02 |
| <i>Capsicum annum</i> | 364.48 ± 16.94 | 0.116 ± 0.02 | 4.34 ± 0.02 |
| <i>Coriandrum sativum</i> | 79.77 ± 2.38 | 0.056 ± 0.005 | 2.63 ± 0.24 |
| <i>Trachyspermum ammi</i> | 306.01 ± 1.49 | 0.096 ± 0.0015 | 1.22 ± 0.06 |
| <i>Syzygium aromaticum</i> | BDL | 0.04 ± 0 | 5.67 ± 0.06 |

Table 5 Loading of chemical variables (17 No.) on Factors for all the spices

| Parameters | Factor 1 | Factor 2 | Factor 3 | Factor 4 | Factor 5 |
|---------------------|--------------|--------------|-------------|---------------|-------------|
| FRAP, mg AAE/g DW | 0.71 | 0.21 | 0.41 | - 0.24 | 0.31 |
| TPC, mg/TAE/g DW | 0.87 | - 0.05 | 0.34 | 0.35 | 0.03 |
| TFC, mg/QE/g DW | 0.83 | 0.41 | 0.26 | 0.17 | 0.11 |
| DPPH, | - 0.67 | - 0.48 | - 0.20 | 0.02 | - 0.19 |
| Total Moisture% | 0.82 | 0.12 | 0.12 | 0.50 | 0.22 |
| Total Ash% | 0.09 | - 0.18 | 0.94 | 0.20 | - 0.02 |
| Total Fat% | 0.65 | - 0.42 | 0.06 | - 0.28 | 0.51 |
| Crude Fibre% | - 0.11 | -0.84 | 0.10 | - 0.05 | 0.36 |
| Protein % | - 0.19 | -0.88 | 0.13 | 0.28 | - 0.23 |
| Carbohydrate% | - 0.03 | 0.76 | 0.20 | 0.12 | 0.02 |
| P (mg/100 g) | 0.17 | 0.06 | - 0.14 | - 0.02 | 0.92 |
| Na (mg/100 g) | - 0.06 | - 0.48 | - 0.37 | 0.43 | 0.52 |
| K (mg/100 g) | 0.14 | 0.13 | 0.88 | 0.19 | - 0.13 |
| Fe (mg/100 g) | -0.93 | 0.16 | 0.15 | - 0.06 | 0.13 |
| Phytic acid (%) | - 0.12 | - 0.27 | - 0.21 | - 0.83 | 0.18 |
| Tannic acid (%) | 0.57 | 0.20 | 0.71 | - 0.03 | - 0.15 |
| Oxalate content (%) | 0.20 | - 0.35 | 0.18 | 0.76 | 0.13 |
| Expl.Var | 4.85 | 3.25 | 2.90 | 2.17 | 1.87 |
| Prp.Totl | 0.29 | 0.19 | 0.17 | 0.13 | 0.11 |
| Eigenvalue | 6.28 | 3.33 | 2.64 | 1.67 | 1.13 |
| % Total | 36.91 | 19.57 | 15.53 | 9.82 | 6.65 |
| Cumulative | 6.28 | 9.60 | 12.24 | 13.91 | 15.04 |
| Cumulative | 36.91 | 56.48 | 72.02 | 81.83 | 88.49 |

Fig. 5 Clustering of spices based on their analysed chemical characteristics



Through this study, it is observed that the Himalayan spices *Allium stracheyi* and *Angelica glauca* are comparable with other commonly used spices in terms of their nutraceutical potential.

Conclusion

Himalayan spices *Allium stracheyi* and *Angelica glauca* are being used both as spices as well as medicinal plants. The present study has shown the nutraceutical potential of these spices with their comparison with other well-known spices. The Himalayan spices were found comparable with other well-known spices on the basis of selected chemical properties except for *Curcuma longa* and *Syzygium aromaticum*. Therefore, use of these spices as a nutrient supplement is highly promising, and these can be used in food fortification.

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Compliance with ethical standards

Conflict of interest There is no conflict of interest.

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