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Discrimination in North-eastern pomelo using ionomic profiling and multivariate statistics

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ABSTRACT

The study was conducted at Dept. of Horticulture, Sikkim University, Gangtok during 2017-2018 with an objective to discriminate northeastern pomelo using ionomic profiling and multivariate statistics. Five different states of northeastern region were selected which are considered to have maximum pomelo diversity in India and fruits along with its soil from rhizosphere were examined. In this paper, discrimination of different pomelo germplasm was carried out based on their geographical origin using multi-elemental profiling. Twenty-one elements (Ba, Pb, Ag, Al, Ca, Cd, Co, Cs, Cu, S, Li, Mg, Na, U, B, Zn, Hg, Si, I, Ce, Mo)were determined by Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The ANOVA, PCA and LDA were applied to discriminate the origin of pomelo. ANOVA had revealed significant difference between twenty one elements from different geographical origin, both in soil and fruit. Magnesium (Mg) was found to be most abundant element however its concentration was relatively higher in fruit peel than pulp. Multivariate tools such as PCA and LDA have shown the geographical origin of pomelo, distinctly producing five individual groupings.

Key words : *Geographical origin, ICP-MS, LDA, PCA, pomelo*

Introduction

Citrus is one of the most economically important fruit crops, belonging to the family *Rutaceae*. It is free from sodium and cholesterol and also rich source of vitamin C, folic acid, potassium, flavonoids, coumarins, pectins and dietary fibres (Roy *et al.* 2014). Pomelo (*Citrus grandis*), a member of genus *Citrus* is native to Southeast Asia and usually identified with the wild species *Citrus maxima* (Burm.) Merr. based on morphology and molecular markers (Singh *et al*., 2015), where it is known under a wide variety of names. It is the largest fruit grown in almost all the citrus growing countries of the world and is considered to be principal ancestor of grapefruit (*Citrus paradisi*) (Morton, 1987). It is known for good source of nutritional and mineral composition like ascorbic acid, dietary fibres, essential mineral nutrients which acts as strong antioxidants having variety of medicinal and nutritional property serves as a protective food. Pomelos are grown in more than 50 countries, majorly in China, Vietnam, United States, Mexico, South Africa, Thailand *etc.* However, the pomelo production is mainly contributed by China (53.12%) followed by Vietnam (8.94%) and United States (5.56%) respectively (FAO, 2020, http://www.fao.org/3/cb6492en/cb6492en.pdf). It is a monoembryonic species which produces seedlings with genes from its two parents which resulted in large genetic variability in their population in terms of plant morphology as well as fruit characters. Pomelo is usually employed in many artificial breeding programs as one of the donor parents (Sameh *et al.* 2014). There is wide variation in physical and chemical characteristics in pomelo across South Asia (Morton, 1987). The fruit is mainly popular for its tolerance to insects and disease and large variability exists in the country (Karnataka, Bihar, W.B., North east, Tamil Nadu *etc.*) (Singh *et al.*, 2015). In India, pomelo trees are found growing wild or semi-wild in marginal land or in home gardens without any commercial cultivation however maximum diversity is reported from North east region (Singh and Singh, 2003; Roy *et al.*, 2014; Singh *et al.*, 2015). Recently, *Devenhalli* pomelo from Karnataka got GI protection likewise many untapped pomelo germplasms have potential as export crop which needs to be validated and linked to its geographical origin for easy identification and recognition in the international market. Some studies have been done to morphological and chemical characterisation of the fruit as a screening tool for better quality, marketing and conservation (Shen *et al*., 1999, Maiti *et al*., 2001, Samarasinghe, 2005) however no ionomic profiling studies was done earlier to link or connect the pomelo germplasm to its geographical locations in India. In this paper, we try to incorporate ionomic approach to multi-elemental profiling of pomelo for any discrimination possibility using ionomic signatures.

Many researchers have worked on developing new analytical and identification tool for verification of geographical origins yet multielement testing is promising due to its high reproducibility (Yan *et al.*, 2015). In addition, multielement testing is particularly sensitive to the soil and climate conditions under which agricultural crops are grown. Hence, it is one of the most common techniques employed to characterize and verify different food samples. Multi-elemental profiling followed by multi-variate analysis has been effectively used to differentiate food samples from the different geographical locations *viz.* potato (Anderson *et al.*, 1999), onions (Ariyama *et al.*, 2007), mutton (Sun *et al.*, 2011), fruit juices (Tormen *et al.* 2011), wheat (Zhao *et al*., 2011), rice (Li *et al.* 2012), wines (Martin, *et al*., 2012), pomelo (Yan *et al*., 2015), heliconia (Avendaño-arrazate *et al.*, 2017), cabbage (Bong *et al.* 2012), rose (Singh *et al.*, 2020) and jackfruit (Debbarma *et al.*, 2021).

Therefore, in this paper, multi-element fingerprinting coupled with principal component analysis (PCA) and linear discriminant analysis (LDA) were carried out to verify the geographical origins of pomelo grown in the region with highest diversity as reported.

Materials and Methods

Sample collection and its preparation

Matured, ripened and fresh edible quality pomelo fruits from the bearing trees aged ranging about 15 to 25 years were collected from five different states of North eastern region (NER), India. Five pomelo fruits were collected from five representative trees for the experiment. The pomelo growing states of NER, India *viz.* Arunachal Pradesh (Boleng, Lileng, Rengo, Pangkang), Sikkim (Tokdey, Makha, Tadong, Pakyong), Meghalaya (Kling, Byrnihat, Nongpoh, Pahammawlein), Assam (Lokhra, Khanapara, Lalmati, Jalukbari), Mizoram (Khawlailung, Sairang, Luangmaul, Tlangnuam). The pomelo fruits were harvested when the fruits turn its dull color to shiny bright color approx. 5-6 months after fruit set. For storage, the fruits were kept in air-sealed plastic bags and stored in -20^{o}C until further analysis were carried out. Soil samples collected from different geographical origins were taken from the depth of approximately 15 to 20 cm and kept in plastic bags. These collections were carried out during the month of July, 2017. Soils were completely air-dried in a hot air oven, and dried soils were milled into powder with an agate mortar and pestle, homogenized and shifted through a plastic sieve with a diameter of 2 mm. Both soil and fruit (peel and pulp) samples were digested through open air digestion. The samples were digested in a di-acid *i.e.* Nitric acid (HNO_3) and Perchloric acid ($HClO_4$) in a ratio of 9:4. The samples were weighed for 0.5g in a weighing scale, the weighed samples were taken in a 50 ml conical flask, then 10 ml of the Di-acid was added into the conical flask containing the samples, the conical flask was then heated in a hot plate in open condition, the flask was heated till a white fumes appeared and the liquid became colourless, the leftover amount of the digested sample was filtered into a 50 ml volumetric flask using parchment 4.0 filter paper, finally the volume was made up to 50 ml with distilled water. Then, the digested samples were subject to ICPMS analysis for ionome profile.

Multi-element analysis

Twenty-one elements *viz.* Ba, Pb, Ag, Al, Ca, Cd, Co, Cs, Cu, S, Li, Mg, Na, U, B, Zn, Hg, Si, I, Ce, Mo were determined by Inductively Coupled Plasma Mass Spectrometry (Perkin Elmer Nex ion 300X, USA) with cross flow nebulizer. The digested samples were analysed for the ionic constitution using multi-elemental standard supplied by the manufacturer.

Statistical analysis

The elemental profile of soil and fruit derived from ICPMS were subjected to analysis of variance (ANOVA) with Completely Randomized design using statistical package (WASP 2.0) developed by ICAR- Central Coastal Agriculture Research Institute (https://ccari.icar.gov.in/wasp2.0/index.php) to check means of multi-elemental content of soil and fruit are significantly different from each other or not. The two-dimensional multi-variate *viz*. principal component analysis (PCA) and linear discriminant analysis (LDA) were carried out to identify the geographical origins of pomelo collected from different states of NER, India using ionome profile of their soil and fruits. All statistical analyses were performed by using SAS JMP Statistical Discovery Pro 2022.

Results

Multi-elemental profiling of pomelo fruit (peel and pulp)

ANOVA conducted for the fruit samples collected from five different states of North East Region (NER), India had showed significant differences between them. ANOVA had revealed in Table 1 that elements like magnesium, calcium, barium and silicon both in peel and pulp of pomelo showed higher concentrations than the other elements. The pomelos from Arunachal Pradesh had highest concentration for elements like Al, Li, B, Si in both peel and pulp of the fruit. However, pomelos from Sikkim had highest concentration of Ca, S, Na in peel and Mo, I, Na, S in pulp. The Mg content of pomelo was found to be higher in peel than pulp of the fruit. Magnesium was found to be highest in pomelos from Mizoram for both peel and pulp of the fruit. The pomelos from Assam contained 80.97 mg Kg^{-1} in peel and 60.87 mg $Kg^{-}1$ pulp, which were highest concentration of Barium as compared to pomelos from other states. For trace elements like Co and B, fruit pulp of Arunachal recorded highest concentration and least concentration was observed in Assam pomelo. The pulp of pomelo contained higher concentration of these elements than compared to peel. For elements like S, Mo and I, Sikkim pomelo showed highest concentration in pulp than peel. In case of Cu and Zn, the concentration of these elements was recorded highest in pomelo from Mizoram. The concentration of these two elements was observed higher in pulp that peel of the pomelo fruit.

For harmful heavy metal analysis, lead (Pb) content was highest in pomelo from Mizoram in both peel and pulp and lowest in Meghalaya pomelo. The lowest cadmium (Cd) content (0.005 mg Kg^{-1}) was found in peel of pomelo from Meghalaya and highest in pulp of Sikkim pomelo (9.87 mg Kg^{-1}). The pomelo fruit from Assam contain lowest concentration of mercury (Hg) and the highest concentration was found in pulp of Mizoram pomelo.

Multi-elemental profiling of soil

Majority of the elemental content collected from soil samples *viz.* Assam and Sikkim recorded highest element concentration than soils from other states. Elements like Ag, Al, Cd, Hg and I were recorded as 1.0 mg Kg^{-1}, 736.24 mg Kg^{-1}, 0.16 mg Kg^{-1}, 0.18 mg kg^{-1}, 3.95 mg Kg^{-1} respectively in soil from Sikkim. In contrast, the soil samples from Sikkim contained lowest concentration of elements like Li, S and B. Soils from Assam also recorded highest content in element *viz.* Barium (1375.70 mg Kg^{-1}), Lead (33.34 mg Kg^{-1}), Copper (51.43 mg Kg^{-1}), Sodium (197.30 mg Kg^{-1}), Uranium (53.09 mg Kg^{-1}). While, soil samples from Assam also had the lowest concentration of elements like Al, Ca, Co, Mg, Zn and Hg.

Elements like Ba, Cs, Li and Ce were observed highest in soil collected from Meghalaya while this soil had also lowest concentration of elements like Cd, Hg, Si and I. Elements like Co, S, B and Zn were present in highest concentration in soil samples from Mizoram. The value for these elements were recorded as 9.58 mg Kg^{-1}, 24.36 mg Kg^{-1}, 0.97 mg Kg^{-1}, 4.57 mg Kg^{-1} respectively. In Table 2, it also showed highest content of two macro elements like Ca (36.69 mg Kg^{-1}) and Mg (986.93 mg Kg^{-1}) in soil from Arunachal Pradesh. While, the elements like Ba, Pb, Ag, Cu, Na, Ce had the lowest concentration in the soil.

Table 1. Multi-elemental concentrations of pomelo fruit (peel & pulp) and soil collected from different states in NER.

Elements	Arunachal Pradesh			Sikkim			Meghalaya			Assam			Mizoram			C.D. @ 5%		
(mg kg^{-1})	Peel	Pulp	Soil	Peel	Pulp	Soil	Peel	Pulp	Soil	Peel	Pulp	Soil	Peel	Pulp	Soil	Peel	Pulp	Soil
Ba	29.44	26.65	232.94	45.92	53.55	647.74	42.80	20.05	1375.70	80.97	60.87	1375.70	30.52	28.50	891.35	2.64	1.91	28.68
Pb	1.15	1.11	1.13	0.73	0.51	3.61	0.19	0.13	16.35	0.27	0.35	33.34	1.32	1.22	29.40	0.17	0.14	2.50
Ag	0.82	0.82	0.67	1.05	1.26	1.00	0.78	0.44	0.89	0.68	0.57	0.93	0.80	0.71	0.75	0.09	0.12	0.19
Al	18.80	15.00	262.28	10.95	10.00	736.24	3.28	2.05	0.00	7.88	4.97	85.27	8.35	7.07	185.28	0.61	1.51	15.13
Ca	72.38	64.48	36.69	12.25	19.43	13.38	44.08	50.87	6.82	66.45	20.93	5.72	69.00	76.97	23.63	1.82	1.16	2.84
Cd	0.03	7.31	0.06	0.03	9.87	0.16	0.005	2.14	0.03	0.02	0.64	0.07	0.03	8.17	0.10	NS	0.63	0.02
Co	0.13	12.01	9.37	0.17	1.53	8.04	0.05	5.66	8.54	0.14	0.58	7.75	0.13	7.76	9.58	0.04	0.74	0.69
Cs	0.14	5.03	2.82	0.31	6.51	3.46	0.18	6.75	4.77	0.22	2.25	4.31	0.12	1.92	2.76	0.04	0.55	0.47
Cu	11.70	39.10	28.66	14.31	47.99	41.65	3.77	36.61	49.89	9.13	47.89	51.43	12.94	63.68	39.10	1.06	1.00	1.92
S	18.50	22.44	17.21	25.12	31.30	14.79	11.14	21.56	17.62	22.75	24.21	19.12	22.51	22.28	24.36	1.31	0.88	1.23
Li	16.53	21.10	65.86	0.30	0.27	58.06	0.24	0.28	127.19	0.38	6.31	105.54	0.48	0.56	120.84	0.55	0.30	9.91
Mg	324.15	248.00	986.93	335.85	269.52	941.37	224.67	183.33	567.80	356.46	117.05	337.01	382.58	292.56	751.01	1.52	3.15	127.90
Na	3.77	3.35	15.85	5.36	4.97	16.67	2.75	1.65	16.99	4.21	2.33	197.30	4.33	2.25	33.26	1.40	0.78	2.45
U	0.007	2.11	1.08	0.009	1.36	1.77	0.001	0.83	2.16	0.007	1.02	53.09	0.008	1.04	0.64	0.00	0.31	1.46
B	19.13	42.56	0.69	2.07	29.91	0.34	1.25	2.60	0.40	2.41	4.06	0.43	1.60	2.15	0.97	0.87	1.58	0.09
Zn	0.21	11.80	2.01	0.23	2.57	3.39	0.11	3.79	1.15	0.21	20.38	0.97	0.17	26.17	4.57	0.04	2.41	0.64
Hg	0.10	0.82	0.12	0.095	1.14	0.18	0.093	3.46	0.09	0.07	0.30	0.09	0.09	2.63	0.17	0.006	1.06	0.02
Si	1.39	6.72	85.63	1.33	1.64	74.22	0.64	2.15	68.53	0.84	1.26	69.06	1.22	1.29	72.87	0.12	0.84	5.71
I	0.62	1.60	0.09	0.94	4.07	3.95	0.63	0.76	0.06	0.32	1.97	0.13	0.40	1.76	0.25	0.06	0.92	0.72
Ce	0.40	4.40	33.13	0.51	2.72	61.48	0.52	2.32	263.25	0.87	1.47	96.54	0.50	3.84	55.27	0.03	0.99	24.78
Mo	0.10	1.00	0.16	0.04	4.00	0.12	0.01	0.52	0.15	0.01	0.00	0.19	0.10	0.00	0.20	0.01	0.34	NS

Principal component analysis of pomelo fruit (peel and pulp) and soil ionome

PCA was separately performed for fruit (peel and pulp) and soil ionome collected from different states of NER (Fig. 1A & 1B). The analysis was motivated to bring better insights into vital information which helped to determine distinction of samples. In case of pomelo peel, the first four principal component comprised of 90.25 % of the total variation in the data sample, in which PC1 covered 42.20% of variation followed by 25.94%, 14.35%, 7.75% for PC2, PC3 and PC4 respectively. According to eigen vector shown in Table 2, PC1 had variables like Pb, Al, Ca, Cd, Co, Cu, S, Mg, U, Zn, Si and Mo which represented 42.20% of the total variation. For PC2, the dominant variable were Ba, Co, Cs, S, Li, Na, B, Hg, Ce and Mo representing 25.94% of variance. In PC3, the major contributing variation of 14.35% was associated with Ag, Ca, Cs, Mg, I, Ce and Mo. While, the variation of 7.75 % in PC4 were represented by Ba, Pb, Al, Li, B, Zn. In summary, PCA for pomelo peel indicated that the samples collected from Sikkim and Arunachal Pradesh were relatively abundant in elements like Cs, Na, S, Zn, Ca, U, Cu, Cd, Ag, Si, Al, Pb, Mo, B, Li and Hg. Similarly, the samples from Assam had higher content of elements like Barium and Celium.

However, the PCA results for pomelo pulp in first four principal components had total variation of 95.23%. The PC1 had the highest variation of 35.37%, mostly represented by Ag, Al, Cd, Mg, Na, U, B, Si, Ce and Mo. In PC2, the total variation of 30.06% was contributed by Ba, Pb, Ca, Co, S, Li, Na, Si, Ce and Mo. While, the total variation of 16.86% and 12.92% for PC3 and PC4 respectively were associated with Pb, Cd, Cs, Cu, Li, Mg, Si for PC3 and Ba, Cd, Cs, Li, Mg, Zn, Hg for PC4. PCA results for pomelo pulp implicated that the samples from Arunachal and Sikkim

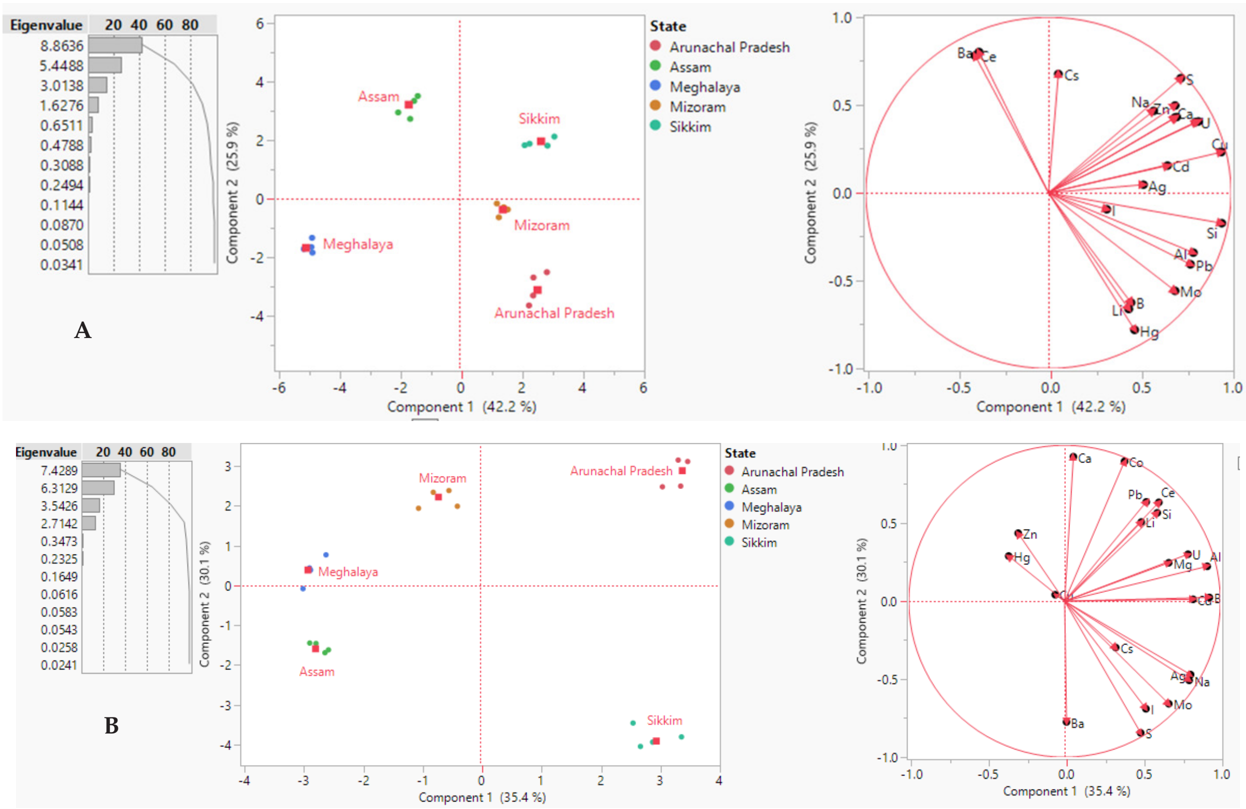

Fig. 1. Principal component analysis (PCA) of pomelo peel (A) and pulp (B) ionome from different states of NER

had relatively higher occurrence of elements as compared to the other states.

As depicted in the Table 2, the total amount of variation of first four PCs accounted for 90.74% in raw data. PC1 accounted for 45.04% of variation wherein highest contributing variable includes Ba, Pb, Al, Ca, Cs, Cu, Li, Mg, Na, U, Zn, Hg, Si and Ce. Variables like Ag, Al, Co, S, B and I had majorly contributed in percentage variation for PC2, which accounted for 22.40%. In PC3, Zn had more weightage in variation followed by Pb, Cd, S and Hg which carried 13.74% of total variation. PC4 was mostly represented by Na, U and Ce, which was 9.55% of variation. According to soil PCA result, the soil samples were grouped into three in which samples from Assam and Meghalaya were observed to be grouped together followed by Mizoram and Arunachal. However, sample of Sikkim was separately grouped from other samples (Fig. 2).

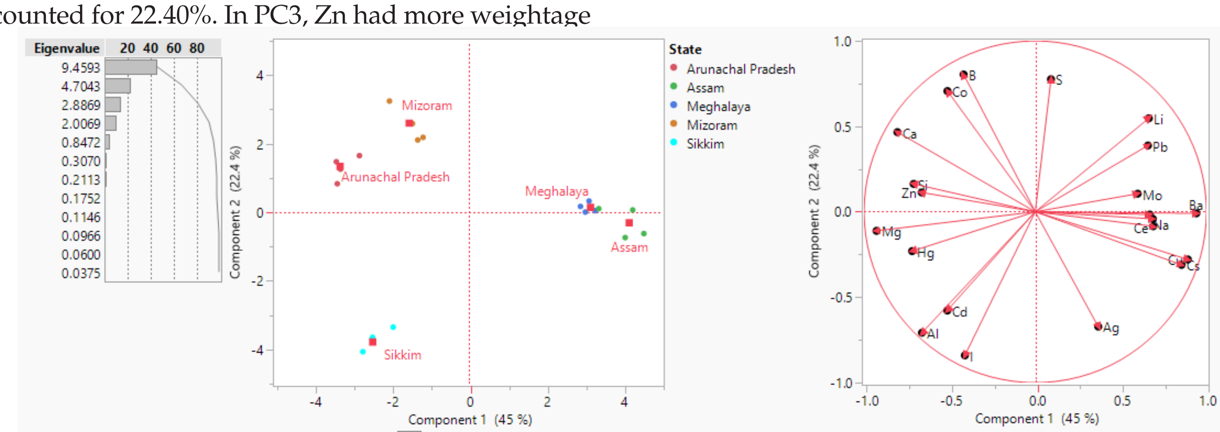

Fig. 2. Principal component analysis (PCA) of soil ionome from different states of NER

Table 2. The eigenvectors, % variance and accumulative variance for the first 4 principal components (PCs) of pomelo peel, pulp and soil samples

	Peel				Pulp				Soil			
Elements	PC1	PC2	PC3	PC4	PC1	PC2	PC3	PC4	PC1	PC2	PC3	PC4
Ba	-0.13	0.34	-0.12	0.30	0.00	-0.31	0.11	-0.35	0.31	0.00	0.13	0.14
Pb	0.26	-0.17	-0.14	-0.28	0.19	0.25	0.28	-0.09	0.22	0.19	0.35	-0.11
Ag	0.17	0.02	0.45	-0.10	0.30	-0.19	0.13	0.05	0.12	-0.31	0.14	0.06
Al	0.27	-0.15	-0.07	0.38	0.34	0.09	0.00	-0.16	-0.21	-0.33	0.13	-0.03
Ca	0.28	0.17	0.21	0.05	0.02	0.37	0.12	0.17	-0.27	0.21	-0.13	-0.14
Cd	0.22	0.07	-0.14	-0.11	0.30	0.00	0.23	0.20	-0.16	-0.27	0.37	-0.01
Co	0.23	0.21	-0.07	0.06	0.14	0.36	-0.07	0.08	-0.17	0.32	-0.03	0.17
Cs	0.02	0.29	0.37	0.19	0.12	-0.12	-0.35	0.36	0.28	-0.14	-0.16	0.15
Cu	0.32	0.10	0.00	-0.11	-0.02	0.02	0.53	-0.02	0.30	-0.12	0.13	0.11
S	0.24	0.28	-0.07	-0.08	0.18	-0.34	0.09	0.03	0.03	0.36	0.35	0.03
Li	0.15	-0.28	-0.12	0.44	0.18	0.20	-0.22	-0.34	0.22	0.26	0.14	0.28
Mg	0.24	0.19	-0.29	-0.17	0.25	0.10	0.26	0.30	-0.31	-0.06	-0.10	0.12
Na	0.19	0.20	0.12	-0.07	0.29	-0.20	0.04	0.00	0.22	0.00	0.14	-0.49
U	0.27	0.17	-0.09	-0.05	0.29	0.12	-0.14	-0.19	0.23	-0.04	0.09	-0.49
B	0.15	-0.27	-0.12	0.45	0.34	0.01	-0.17	-0.09	-0.14	0.37	0.21	0.00
Zn	0.23	0.18	-0.05	0.31	-0.11	0.17	0.36	-0.30	-0.21	0.05	0.40	0.17
Hg	0.16	-0.33	0.16	-0.05	-0.13	0.12	0.02	0.49	-0.23	-0.11	0.35	0.13
Si	0.32	-0.07	0.04	-0.01	0.22	0.23	-0.27	-0.12	-0.24	0.07	-0.22	-0.22
I	0.11	-0.04	0.53	0.11	0.19	-0.27	0.17	-0.01	-0.13	-0.39	0.16	0.09
Ce	-0.14	0.33	-0.24	0.16	0.22	0.25	0.10	0.11	0.23	-0.02	-0.20	0.44
Mo	0.23	-0.24	-0.20	-0.19	0.25	-0.26	-0.03	0.18	0.19	0.05	-0.19	-0.10
Total Variance (%)	42.20	25.94	14.35	7.75	35.37	30.06	16.86	12.92	45.04	22.40	13.74	9.55
Accumulated Variance (%)	42.20	68.15	82.50	90.25	35.37	65.43	82.30	95.23	45.04	67.44	81.19	90.74

Linear discriminant analysis of fruit (peel and pulp) and soil ionome

According to the results of LDA (Fig. 3A, 3B & 3C), it was observed that all the samples were distinctly distinguishable from each other in all cases *viz.* peel, pulp and soil. There was significant separation between the samples from the five states of NER. However, the samples from Mizoram and Sikkim were observed to be grouped together with minor overlapping in LDA peel followed by Arunachal and Sikkim in pulp adjacently joined but with no overlapping. In case of soil LDA, all the samples were easily separable. As in comparison with PCA results, LDA proves to be more efficient in distinguishing the samples and hence a powerful data modelling tool for discriminating geographical origins (Yan *et al.*, 2015).

Discussion

Elemental composition of soil at a particular region is differentially influenced by its topography, soil parent material, soil type, pH and climatic conditions. Variations in the distribution of these elements among different geographical locations can give distinct elemental signatures in organic tissues and the element contents in vegetation were also influenced by plant species, since the element absorption capacity of vegetation is species dependent (Sun *et al.*, 2011). The concentration of the analysed element was different in peel and pulp of the fruit which was in compliance with the findings of (Watanabe *et al.*, 2016). Based on Pearson correlation, there is a negative correlation of soil element with most of the fruit nutrients. Elements like Na and Ba have the most positively correlated elements. Our results have revealed that the richness of soil nutrient may not directly influence the fruit elements rather contributed by the genetic makeup of the plant and its ability to absorption and retention on its edible parts (Sun *et al.*, 2011; Yang and Volker, 1999). Further, the fertility status of soil in different states based on the multi-element profile has found to be distinguishable has been reflected in the Table 1. The soil in the five states studied varied from hill soil, lateritic and alluvial soil mostly being rich in humus and acidic

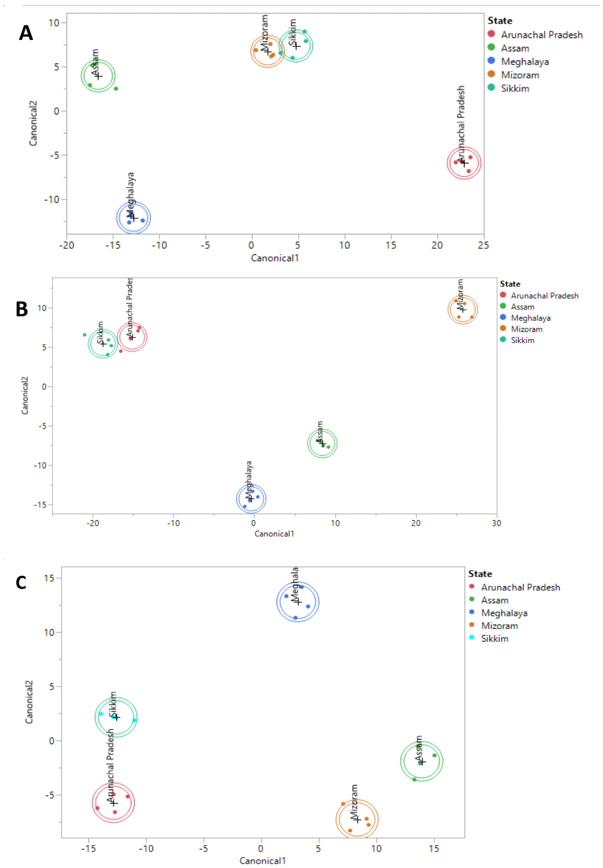

Fig. 3. Linear discriminant analysis of pomelo peel (A), pulp (B) & soil (C) from different states of NER

in nature. However, the soils from Arunachal and Mizoram were enriched with basic elements like Mg, Na and Ca while low in Al, Zn, Cu and S as compared to other states. Comparatively, soils from Sikkim were relatively richer in acidic elements like Al and Zn. However, the multi-variate analysis of soil and fruit only do not shed light on the geographical origin of the pomelos. Therefore, the implementation of visualisation method has become of prime importance to validate the geographical origin of pomelos.

According to PCA and LDA results, the geographical origins of pomelo were easily distinguishable and distinct. PCA for fruit ionome (both peel and pulp) and soil had yielded five different groups. The most representative variable of PCA for fruit ionome was Na and U in variable clustering 1 and 2 respectively with proportion of variation explained by clustering was 82.5%.

Similarly, little proximity between Sikkim/ Mizoram in case of peel and Sikkim/Arunachal in pulp were observed in LDA whereas LDA for soil ionome has given five different groups. The distinguished and distinct grouping for fruit and soil ionome in LDA could be an indicator of geographical locations and genetic constitution of pomelo studied in this paper. Due to the differences in elemental concentration, the five distinguishable grouping were seen in LDA results was in accordance with (Ariyama *et al*., 2007).

Conclusion

The multi-elemental and multi-variate analysis such as PCA and LDA results have shown that ionome profiling combined with dimensionality reduction methods are efficient in discriminating pomelo based on their geographical origins. In this paper, the pomelo originated from five different states of north east region have found to be discriminated based on their elemental composition. Further, this discriminating signature of elemental composition may be incorporated in bringing the superior pomelo strains in breeding programme in future. However, this work needs to be further validated with more samples from other regions of India in order to develop more reliant and robust model.

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Declaration of No Conflict of interest

The authors declare that there is no conflict of interest in this work.

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