Indian J. Plant Genet. Resour. 32(2): 207-216 (2019)
DOI 10.5958/0976-1926.2019.00026.3

RESEARCH ARTICLE

Citrus Diversity Fair: A Means of Locating Citrus Species Diversity and Selection of Superior Clones of Local Importance

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(Received: 27 April 2018; Revised: 20 December 2018; Accepted: 22 December 2018)

A citrus diversity fair was organized on 5th November 2011 by National Research Centre (NRC) on Litchi, Muzaffarpur in collaboration with Lt. Amit Singh Memorial Foundation, New Delhi at Mahmada, Bihar of Pusa Site of the UNEP/GEF Project to know the extent of citrus variability and selection of superior genotypes of citrus. Considerable diversity of citrus is observed in the study area due to conducive climate. A total of 109 farmers displayed 135 samples of nine *Citrus* species. Richest variability was displayed for pummelo (out of the 135 samples displayed, 127 belonged to pummelo). This fruit species showed rich natural variability due to its out-breeding habit and also the pummelo plants are raised traditionally through seeds in this area. Sixty-seven plants could be validated in the farmer's fields and only those samples were included in these studies. Based on morpho-physico-chemical traits, *viz*; fruit size, fruit shape, flesh colour, TSS (^{0}Brix), acidity percentage, number of seeds/fruit, rind thickness, juice percentage, taste, etc., 11 accessions, *viz;* 9, 62, 8, 17, 4, 6, 31, 39, 44, 51 and 52, were identified as superior genotypes, which could be multiplied and distributed to the farmers for quality fruit production of pummelo.

Key Words: Bio-diversity fair, *Citrus maxima* L., Clone, Genetic variability, Local seedling population, Pummelo

Introduction

Citrus is one of the most popular fruit crops and cultivated in around 140 countries. The genus has its centre of diversity in Northeast India, Malayan Archipelago, China, Japan and Australia (Swingle and Reece, 1967). In India, many *Citrus* species including mandarin (*Citrus reticulata*), sweet orange (*C. sinensis*), acid lime (*C. aurantifolia*) and lemon (*C. limon*) are grown commercially, whereas grapefruit (*C. paradisi*), pummelo (*C. maxima*), galgal (*C. pseudolimon*), citron (*C. medica*), etc. are grown in home gardens or mixed orchards for domestic consumption (Sharma *et al*., 2004). Most of these non-commercial types are grown as seedlings, and citrus being a cross-pollinated crop, considerable variability in the seedling population is observed. This genetic diversity is important from global as well as local point of view and is traditionally maintained in home gardens. The wealth of local citrus genetic diversity by and large remains non-collected.

Breeding of fruit crops is a lifetime work for any plant breeder because of the perennial nature. The variability in fruit trees, growing in home gardens and mixed orchards has been observed for a very long time for undertaking selection, exchange and breeding by fruit growers and villagers (Zeven, 1998). On farm conservation in collaboration with custodians of diversity is a cost-effective method. Genetic improvement in horticultural crops can be done through identification of elite materials (plus trees) available in the community followed by their characterization, evaluation and multiplicationfor community benefits (Sthapit *et al*., 2006; Sthapit *et al*., 2013; Sthapit *et al*., 2016). Diversity fair was initiated as a common tool for raising awareness among public on the importance of conserving diversity (Tapia and Rosa, 1993). The method is presently being used for identifying superior cultivars/accessions, promoting exchange of traditional knowledge and planting materials amongst farmers, identification of diversity rich areas and selection of elite materials (Sthapit *et al*., 2003; Gajanana

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et al., 2015). The present study with bio-diversity fair as a tool was undertaken with the objectives to raise citrus biodiversity awareness among rural communities, locate genetic diversity, assess extent of diversity, and identify elite materials for mother plant selection in order to introduce superior clones in the local production systems.

Citrus fruits and particularly pummelo are grown in almost every household of the study area in the eastern Bihar state of India and the fruits are mainly used for the holy festival "*Chhath Pooja*". Due to the poor quality of the seedling borne fruits, pummelo is not very much popular as a table fruit in the area under study. The identified clones from this diversity fair shall contribute in consolidating the genetic base of *Citrus* species which in turn might be helpful in improving the livelihood and nutritional security of the local people.

Materials and Methods

Selection of Site

The communities which were studied are based at Pusa (Samastipur), Bihar the site of study of the UNEP/ GEF project. The project aimed at conservation and sustainable use of cultivated diversity in the Samastipur district of Bihar in India. The climate of Pusa is humid and subtropical, with maximum and minimum average temperatures ranging from 31ºC to 19ºC respectively, and with an average annual rainfall of 1200 mm, distributed over 35–40 rainy days during the monsoon season. It is located at latitude of 25º46 N and longitude of 86º10 E with an altitude of 53m. Four project communities/ villages, form part of Pusa site namely *viz*; Mahmada, Jagdishpur, Dhobgama and Murliyachak. The Table 1 shows site characteristics data of these communities. Initially, the focus group discussion, four cell analysis and baseline survey were carried out to assess the citrus diversity in these project communities of Pusa site and it was observed that considerable amount (richness) and distribution (evenness) of citrus tree diversity is being maintained on-farm. Farmers are maintaining and using diverse traditional fruit tree diversity for their own welfare and benefit (Singh *et al*., 2016a). It is indicative of the cultural and religious importance of the citrus diversity and its subsequent conservation by the local peasant farmers.

Citrus Diversity Fair

A citrus diversity fair was organized at Mahmada (Pusa, Samastipur, Bihar), under the Project, on 5th November 2011. The methods of diversity fair were reviewed from the literature (Tapia and Rosa, 1993; Sthapit *et al*., 2003; Adhikari *et al*., 2012; May *et al*., 2014) and relevant checklists were developed for community consultations. Several discussions were held with different community groups regarding the objectives, venue and the time of the fair and based on the responses of the villagers, date and venue of the diversity fair was fixed and communicated to all the participating farming communities. The fair was organized in the premises of a local school to stimulate awareness among school children as an additional and fruitful outcome. An enthusiastic response was received from all the participating communities. A total of 109 farmers exhibited 135 samples of citrus diversity and 260 farmers visited the fair and perused the displayed diversity. The different species exhibited were sweet orange (*Citrus sinensis*), pummelo (*C. maxima*), acid lime (*C. aurantifolia*), lemon (*C. limon*), rough lemon (*C. jambhiri*), Rangpur lime (*C. limonia*), Cleopatra mandarin (*C. reshni*), galgal (*C. pseudolimon*) and sweet lime (*C. limettioides*).

Sample Collection

The fruit samples were collected in the fruit diversity fair. Based on visual observations and organoleptic

Table 1. Site characteristics and community richness of citrus diversity measured by four cell analysis (Source: Gajanana *et al*., 2014)

Characteristics	Mahmada	Jagdishpur	Dhobgama	Murliyachak
Total number of households	1124	500	744	200
Total fruits HHs	169	60	250	55
Community citrus richness	6	3	3	3
Common Citrus species	Acid lime$^{\delta}$	Acid lime$^{\delta}$	Acid lime$^{\delta}$	Acid lime$^{\delta}$
Rare Citrus species		Rough Lemon$^{\beta}$/Lemon$^{\Omega}$, Sweet lime*		Lemon$^{\Omega}$
Unique *Citrus* species	Pummelo**	Pummelo**	Pummelo**	Pummelo**

↑*Rare means few households and few trees; Unique means many households and few trees; Common means many households and many trees;* δ= *C. aurantifolia* L. (Acid lime; Kagzi nimbu); β =*C. jambhiri* (Rough lemon); Ω = *C. limon* (Lemon);*= *C. limettioides* (Sweet lime; Mitha nimbu) and ** *C. grandis/maxima* (Gagar/Bhogate)

taste by a group of experts, comprising of scientists, State Horticulture Officer and farmer's representatives, best fruit samples were identified. The genetic diversity collected through the diversity fairs was monitored at farmer's fields to validate the results of the fair and to characterize these landraces for physico-chemical characteristics. In the follow up programme, out of 127 exhibited samples of pummelo, 67 types of pummelo could be precisely located growing in the farmer's fields. After the confirmation of clones in the field, two fruits from each of these 67 samples were analysed for different physico-chemical characteristics *viz;* fruit colour, weight, length, breadth, presence or absence of oil glands, rind thickness, flesh colour, number of segments and seeds, TSS (ºBrix) and acidity.

Fruit Characteristics Study

The external characters of fruit *viz*; height and diameter were measured by Vernier callipers and fruit weight was measured by electronic digital balance. The qualities of fruit like peel, pulp and juice percentage were measured based on fruit weight. Titrable acidity (TA) was measured using the titration method (AOAC, 1989). The taste was judged by an organoleptic test (tasting by eight persons in the laboratory and giving the scores for edibility and acceptability). The TSS was measured with the help of hand-held digital refractometer (Singh, 2016). The juice percentage, TSS: acid ratio and fruit length: breadth ratio was calculated following standard procedures.

Selection of Elite Clones

Selection of superior clones was carried out based on the evaluation of fruit characters and the number of desirable characters such as fruit size, rind thickness, T.S.S., pulp colour, juice percentage and flavour. These characters are used as indicators for edible fruits and/ or for religious/ cultural utilization of the fruits. The preference of farmers is for sweet taste, red pulp and medium-sized fruits. The quality genotypes were adjudged by the number of desirable characteristics possessed by them.

Statistical Analysis

The experiment was conducted in Completely Randomized Design (CRD) with two replications. Data were subjected to one-way analysis of variance. *P* values ≤ 0.05 were considered as significant. All the seventeen physico-chemical characteristics were converted into bi- and multi-state code. Cluster analysis was performed using simple matching coefficient method using NTSYS ver. 2.10e software (Rohlf, 2000) based on unweighted pair group method with arithmetic average (UPGMA). Principal Components Analysis (PCA) of all clones was done by NCSS 2007 v 07.1.18 (Hintze, 2007).

Results

Maximum variability was represented in the case of pummelo at the citrus diversity fair, which was further validated by variability in the farmer's fields. Out of 127 pummelo samples collected during the diversity fair, only 67 were confirmed in the farmer's fields and one of the main reasons behind low on-field confirmation was that the farmers already harvested the fruits and marketed/ distributed for the festival of *Chhath Pooja*, the major reason for which the pummelo plants are maintained. A wide range of variability was observed amongst the pummelo plant samples for various characteristics observed. The fruit colour varied between 1-6 with 1 being the green and the 6 being the dark yellow. The fruit weight varied between 0.60 (Sample/clone no. 42) and 2.50 kg (Sample/clone no. 2). The range for fruit length was from 11.25cm to 23.30 cm in clone no. 8 and 12, respectively and the fruit width varied between 12.05 (in clone 39) and 23.90 cm (in clone 2). The flesh colour varied from white to dark red on a scale of 1 (white) to 6 (dark red). Rind thickness also exhibited variability and ranged from 0.80cm (in clone 62) and 3.75 cm (in clone 2). The number of segments/fruit varied between 11.50 (in clone 31) and 19.50 (in clone 16). The T.S.S. varied between 7.95 (in clone 14) and 12.85 ºBrix (in clone 39).

Seedlessness is an important character in context of edibility and consumer preference, however, no clone with seedless fruits could be found among the collected genotypes. The range for seeds/fruit varied between 17.50 (in clone 33) to 168.00 (in clone 16). The juice sacs could be classified in to either soft or hard categories. Acidity also exhibited wide variation amongst the selected clones with a minimum of 0.32% in clone no.46 and a maximum of 1.74% in clone no.14. From the ratio of fruit length and breadth, it is evident that the fruits were either elongated, round or flattened. Seed size and number were very small with a 100-seed weight of 23.14g (in clone 33) to bold with a 100-seed weight of 68.47 (in clone 21). Juice percentage also exhibited a wide range, 17.24 (in clone 4) to 45.86 (in clone 20). TSS: acid ratio varied between 5.44 (in clone 14) to 32.58 (in clone 46) (Table 2).

Table 2. Variability of the fruit characteristics in the collected (during diversity fair) seedling clones of pummelo

S. No.	Characteristics	Range (Clone No.)	Mean	SE (m)	SE (d)	CV (%)
1.	Fruit colour***	1.000 (7)-4.000 (67)	2.582	0.335	0.473	18.325
2.	Fruit weight (g)	0.632 (42)-2.497 (2)	1.350	0.127	0.179	13.260
3.	Fruit length (cm)	11.250 (8)-23.300 (12)	15.818	0.778	1.100	6.957
4.	Fruit width (cm)	12.050 (39)-23.900 (2)	16.468	0.851	1.203	7.306
5.	Flesh colour*	1.000 (14)-6.000 (66)	4.642	0.299	0.423	9.117
6.	Rind thickness (cm)	0.800 (62)-3.750 (2)	1.720	0.189	0.267	15.520
7.	Segments/ fruit	11.500 (31)-19.500 (16)	14.769	0.756	1.069	7.235
8.	TSS (ºBrix)	7.950 (14)-12.850 (39)	10.672	0.387	0.547	5.127
9.	No. of seeds/fruit	17.500 (33)-168.00 (16)	91.537	19.821	28.031	30.622
10.	Nature of juice sacs**	1.000 (1)-2.000 (66)	1.619	0.162	0.229	14.114
11.	Acidity (%)	0.320 (46)-1.736 (14)	0.596	0.056	0.080	13.399
12.	Fruit length: breadth	0.700 (65)-1.195 (12)	0.963	0.043	0.060	6.262
13.	100 seed wt. (g)	23.140 (33)-68.470 (21)	45.180	5.228	7.393	16.364
14.	Juice weight/fruit (g)	227.530 (3)-798.960 (16)	478.556	43.223	61.127	12.773
15.	Juice %	17.240 (4)-45.855 (20)	36.721	1.027	1.453	3.956
16.	TSS:Acid ratio	5.435 (14)-32.575 (46)	19.392	2.110	2.983	15.385
17.	Taste****	1.000 (14)- 4.000 (64)	2.433	0.323	0.457	18.789

* *Flesh colour*: 1 = white, 2 = light pink, 3 = pink, 4= light red, 5= red, 6 = dark red
***Nature of juice sacs*: 1= soft, 2= hard
*** *Fruit colour*: 1= Green, 2= Green Yellow, 3= yellow green, 4= yellow, 5= Dark yellow
*****Taste*: 1= Poor, 2= Good, 3= Very good, 4= Excellent

The organoleptic taste of the fruits also varied on a scale of 1-4 with 1 being the poor and the 4 being the excellent. Maximum coefficient of variation (30.62%) was recorded for number of seeds/fruit followed by taste (18.79%), fruit colour (18.33%) and 100-seed weight (16.36%), indicating a large amount of variability for these traits (Table 2). Other significantly variable traits were fruit weight, skin thickness, type of juice sacs, acidity and TSS: acid ratio on which selection can be exercised for the selection of superior clones. As far as the range for the quantitative and qualitative traits and the coefficient of variation are concerned, there exists a wide variability amongst the selected clones of pummelo and few of them can be selected for further testing in the replicated trials.

Cluster Analysis for Morpho-physico-chemical Traits

Based on the dendrogram (Fig. 1) generated based on morpho-physico-chemical data, all the 67 clones of pummelo were grouped into two major groups A and B at similarity value of 0.15. Major cluster A comprised 23.88% of studied clones and was further sub-divided into two sub-gruoups as A1 and A2 at 10.25% similarity value. Clone number 4 and 59 were found most distant the sub-groups A1 and A2, respectively. Sub-group A1 and A2 shared 37.50% and A2 62.50 % variability in clones, respectively. Major cluster B comprised most of the studied clones (76.11%). It is further grouped into two sub-groups as B1 and B2 at similarity value of 0.13. Sub-group B1 comprised 94.11% clones and divided into 8 minor groups *viz*., B1.1 to B1.8. Minor cluster B1.1 and B1.4 each comprised 12 clones. In minor cluster B1.1, clone number 5 and 61 exhibited 100% genetic similarity. Minor cluster B1.3 only comprised clone number 8 which was presented as an out-lier in the group (Table 3).

Principal Component Analysis (PCA) for Morpho-physico-chemical Traits

The result of the PCA showed that first eight components accounted for 99.99% of the total variability (Table 4). The fruit colour accounted for 93.38% while the fruit weight accounted for 5.65 % of the total variation. Thus, these traits should be given greater emphasis while making selections. PCA analysis successfully separated out different clusters of pummelo clones. The clone nos. 10, 16, 20, 23, 28 and 35 were scattered into first half of the coordinate whereas, clone nos. 3, 4, 33, 41, 44 and 64 were presented separately in second half axis of the coordinate (Fig. 2). The clone nos. 13 and 61 showed more relatedness (Fig. 1) and PCA results also revealed the same (Fig. 2). Moreover, clone no. 4 presented as an out group in cluster analysis was also confirmed to be a separate group by PCA analysis (Fig. 1).

Selection of Elite Clones

For the selection of the elite clones of pummelo, the clones were grouped in different groups for having desirable scores for the qualitative and quantitative traits (Table 5). The desirability of the quality was decided on the basis of participatory four cell analysis (Sthapit *et al*., 2006) and with farmer group discussion, 12 traits were identified as desirable for this grouping

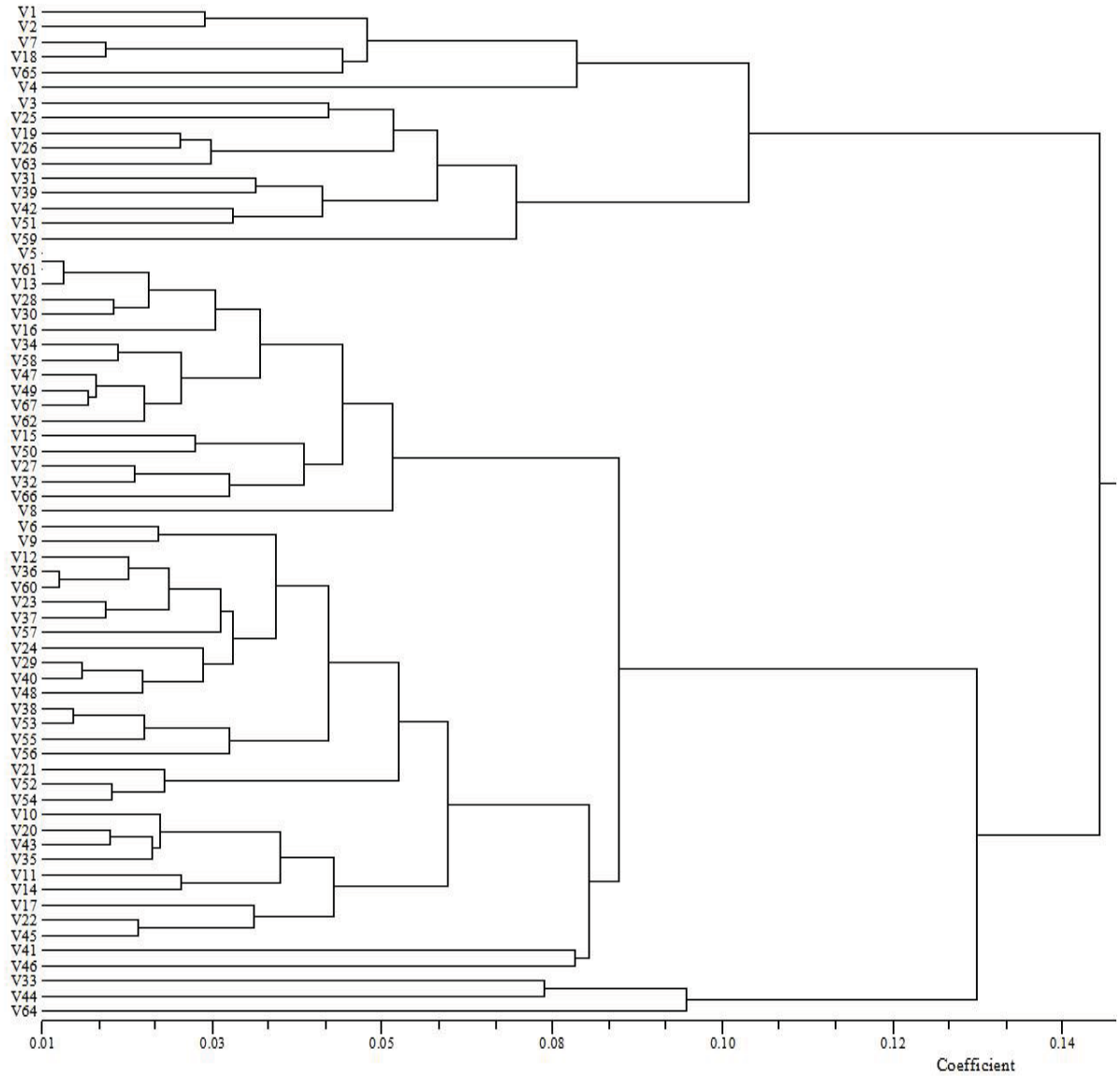

Fig. 1. Dendrogram of sixty-seven selected clones of pummelo based on morpho-physico-chemical traits using the UPGMA method

Table 3. Cluster details of 67 selected clones of pummelo using various morpho-physico-chemical parameters

Cluster Name and Percent clone	Sub-cluster	Total no of clones	Percent clones in sub-cluster	Clone number
A (23.88%)	A1	6	37.50	1, 2, 4, 7, 18, 65
	A2	10	62.50	3, 19, 25, 26, 31, 39, 42, 51, 59, 63
B (76.11%)	B1	48	94.11	
	B1.1	12	25.00	5, 13, 16, 28, 30, 34, 47, 49, 58, 61, 62, 67
	B1.2	5	10.41	15, 27, 32, 50, 66
	B1.3	1	2.08	8
	B1.4	12	25.00	6, 9, 12, 23, 24, 29, 36, 37, 40, 48, 57, 60
	B1.5	4	8.33	38.53, 55, 56
	B1.6	3	6.25	21, 52, 54
	B1.7	9	18.75	10, 11, 14, 17, 20, 22, 35, 43, 45
	B1.8	2	4.16	41, 46
	B2	3	5.88	33, 44, 64

Table 4. Principal Component Analysis among sixty-seven selected clones of pummelo showing the correlations of the first eight principle components (Eigen value >1) with the variables

Principal Component	Fruit Traits	Eigen-Value	%Variance	%Cumulative Variance
PC1	Fruit color	15256.31	93.38	93.38
PC2	Fruit weight (g)	923.33	5.65	99.04
PC3	Fruit length (cm)	83.71	0.51	99.55
PC4	Fruit width (cm)	42.70	0.26	99.81
PC5	Flesh color	23.10	0.14	99.95
PC6	Skin thickness(cm)	2.60	0.13	99.97
PC7	No of segments	1.49	0.02	99.98
PC8	TSS (%)	1.39	0.01	99.99

Table 5. Pummelo clones identified for different characteristics

Fruit character	Selected number of clones	Specific Clone selected
1. Fruit size (500-1000g)	14	3, 8, 17, 25, 26, 31, 39, 41, 42, 44, 46, 51, 59, 64
2. Flesh colour (Dark Red->5.00)	18	1, 4, 5, 6, 9, 16, 17, 22, 27, 28, 38, 43, 45, 50, 61, 62, 63, 66
3. T.S.S. (>11.50%)	11	2, 8, 9, 17, 28, 31, 34, 39, 51, 55, 64
4. Acidity (≤0.50 %)	20	1, 3, 4, 6, 8, 9, 10, 21, 22, 24, 33, 37, 46, 52, 54, 57, 59, 63, 65, 66
5. Number of seeds/fruit (<100)	42	11, 14, 22, 33, 44, 64, 3, 6, 8, 9, 10, 12, 17, 20, 24, 25, 29, 35, 36, 38, 39, 40, 41, 42, 43, 45, 46, 48, 49, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 62, 66, 67
6. Fruit length: breadth (0.96-1.05)	20	1, 4, 7, 9, 13, 19, 20, 21, 22, 25, 29, 38, 43, 45, 49, 54, 55, 56, 60, 62
7. Rind thickness (<1.00 cm)	8	9, 17, 29, 31, 39, 44, 51, 62
8. Fruit colour (>3.00)	16	6, 9, 26, 27, 28, 31, 33, 36, 48, 49, 52, 55, 59, 62, 65, 67
9. 100 Seed weight (< 40.00g)	18	1, 2, 3, 4, 8, 9, 11, 14, 16, 33, 39, 41, 42, 44, 48, 50, 62, 65
10. Juice % (>40.00%)	24	5, 6, 8, 9, 16, 17, 20, 21, 23, 27, 28, 29, 31, 40, 41, 42, 44, 46, 51, 52, 54, 61, 62, 66
11. TSS:Acid ratio (>20.00)	21	1, 4, 6, 8, 9, 10, 21, 22, 33, 37, 44, 46, 52, 53, 54, 57, 59, 62, 63, 65, 66
12. Taste (>3.00)	14	2, 4, 8, 9, 17, 18, 28, 31, 34, 39, 51, 52, 55, 64

were fruit size (500-1000g), flesh colour (Dark red > 5.00 score), T.S.S. (>11.50 ºBrix), acidity (<0.50%), no. of seeds/fruit (<100), fruit length : width (0.96-1.05), rind thickness (<1.00cm), Fruit colour (a score of >3.00), 100 seed weight (<40.00g), juice (>40.00%), TSS : acid (>20.00) and taste (a score of >3.00). Better genotypes of fruit trees can be selected by evaluating the fruit quality. Fruit weight, peel thickness, juice %, TA% and TSS are the major parameters to determine the quality of pummelo fruits. Therefore, elite genotypes of pummelo were selected on the basis of scoring of these fruit parameters.

Plant traits of these genotypes are almost similar, but the fruit physico-chemical characters exhibited variation. After this grouping, the clonal selection was made, and those clones were selected which were possessing more desirable fruit traits.

In this study, significant variation for qualitative and quantitative traits was found in pummelo accessions identified in the citrus diversity fair. On the basis of scoring of fruit characters, total 11 elite genotypes, *viz*; clone nos. 9, 62, 8, 17, 4, 6, 31, 39, 44, 51, and 52 were selected for conservation, breeding and variety development purpose (Table 6). Out of 12 characters considered very important for the evaluation of fruit quality, clone no. 9 (Fig. 3) was possessing eleven characters followed by clone numbers 62 and 8 (8 characters), 17 (7 characters) and clone numbers 4, 6, 31, 39, 44, 51, 52 (6 characters). Thus, it was concluded that diversity fair plays an important role for the identification and selection of elite genotypes of fruit crops for further conservation and utilization directly as superior clonal varieties or as breeding materials for the development of fruit crops.

Discussion

The diversity fairs are effective for documenting and collecting germplasm along with the associated knowledge pertaining to the use of fruits for edible and cultural purposes. They serve as useful tools of practical significance for assessment and collection of diversity of a geographic area or ethnic group. It is a visual method of assessment, and to locate diverse genetic materials, and custodians of diversity across locations (Sthapit *et al.,* 2012). A close interaction with farmers and communities

Table 6. Pummelo clones with six or more desirable fruit characteristics

S. No.	Clone No.	Farmer's name	Desirable fruit characteristics*
1	9	Mohammed Ishaque, Govindpur Chhapra	2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12 (11)
2	62	Reet Lal Sahni, Mushahari	2, 5, 6, 7, 8, 9, 10, 11 (8)
3	8	Joginder Bhagat Shah, Govindpur Chhapra	1, 3, 4, 5, 9, 10, 11, 12 (8)
4	17	Ram Kumar Rai, Dhobgama	1, 2, 3, 5, 7, 10, 12 (7)
5	4	Sudhir Kumar, Mahmada	2, 4, 6, 9, 11, 12 (6)
6	6	Sabdi Devi, Mahmada	2, 4, 5, 8, 10, 11 (6)
7	31	Upender Pathak, Malinagar	1, 3, 7, 8, 10, 12 (6)
8	39	Roshan Kumar Thakur, Malinagar	1, 3, 5, 7, 9, 12 (6)
9	44	Kamal Rai, Mahmada	1, 5, 7, 9, 10, 11 (6)
10	51	Baleshwar Ram, Bhuskaul	1, 3, 5, 7, 10, 12 (6)
11	52	Krishna Shah, Gundi Bandra	4, 5, 8, 10, 11, 12 (6)

*1. *Fruit size* (500-1000g), 2. *Flesh colour* (Dark Red), 3. *Rind thickness* (<1.00 cm), 4. *T.S.S.* % (>11.50%), 5. *Acidity* (0.31-0.50%), 6. *Number of seeds/fruit* (<100 seeds/fruit), 7. 100 seed weight (<40g), 8. *Fruit length: breadth* (0.96-1.05), 9. *Fruit colour* (>3.00), 10. *Juice %* (>40.00), 11. *TSS: acid ratio* (>20.00), 12. *Taste* (>3.00)

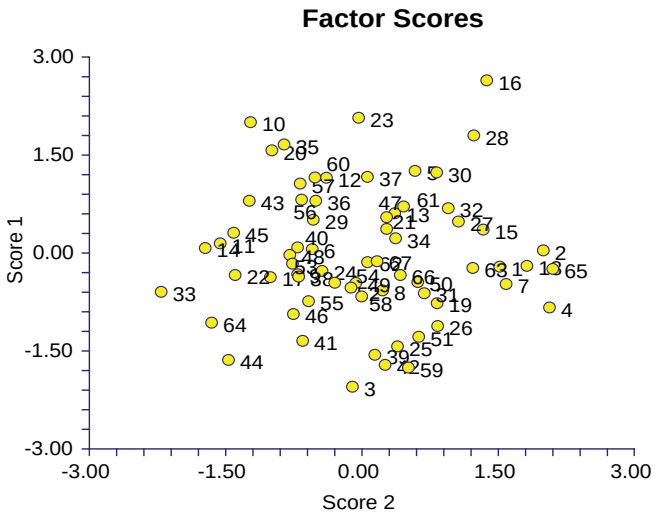

Fig. 2. A principal components analysis (PCA) scatter plot of 67 indigenous seedling clones of pummelo using seventeen morpho-physico-chemical parameters

also motivated them to participate in the germplasm conservation efforts (Mal *et al*., 2011).

The breeding of fruit trees is a long-term activity mainly because of the seedling juvenility. Most of the fruit crops are out-breeders and maintain heterozygosity. As pummelo is grown from seedlings in the study area, a lot of genetic variability can be expected that can be exploited for superior genotype selection. This variability can be explored by field surveys, focused group discussions with the farmers or through organization of diversity fairs as done in the present study. Nine *Citrus* species and varieties were displayed during the citrus diversity fair compared to five *Citrus* species in baseline survey indicate that diversity fairs encourage farmers and farming communities who display rare and unique species and diversities (Upadhyay *et al*., 2012). Selection of superior varieties is rewarding in pummelo owing to its genetic traits; the seeds of most citrus tend to reproduce the traits of the mother plant due to nucellar embryony, but pummelo seeds are monoembryonic and give rise to plants with entirely new horticultural traits. This produces a pool of variability.The results of this study revealed that fruit morphological and physico-chemical characterization may be key in in distinguishing cultivar groups within pummelo population also for selection of potentially better genotypes by the breeders as well as the farmers. Similar observations were made in the clonal selection of mango from the farmer's fields by Singh *et al*. (2016b). Cameron and Soost (1961) stated that pummelo species exhibits considerable variability due to sexual recombination and self-incompatibility. The present study confirmed these findings, re-emphasizing the importance of morphological characterization for improvement through selection and as a base for further studies involving biotechnological and biochemical tools as reported by Bozokalfa *et al*. (2009) and Martasari *et al*. (2012). It was emphasized that high degree of variability in pummelo is due to fruit traits. Further, use of fruit morphology is considerably effective for the recognition of cultivar (Susandarini *et al*., 2013).

Shrestha *et al*. (2012) reported that fruit traits, especially juice, TSS and TA were important parameters for the selection of elite genotypes of citrus trees, also confirmed by the present study. Elite genotypes of citrus fruits can be selected through the assessment of tree morphological traits and consumer's preference. It was reported earlier that farmers highly preferred

Fig. 3. Mature fruits of some of the identified elite seedling clones of pummelo collected during citrus diversity fair

the quality rather than size and yield of the fruits in pummelo (Paudyal and Haq, 2008) whereas consumers prefer the quality of acid lime as round, thin-skinned, yellow colour, juicy and medium size (Dhakal *et al*., 2003), indicating the selection and consumption are governed by farmers and consumers, respecitvely.

The monitoring of genetic diversity at community level helps to develop options for adding value to local crops. Knowing the extent and distribution of the genetic diversity of selected crops over space and time is one of the important outputs of this study. From this citrus diversity fair, it is expected that farmers and communities felt encouraged and appreciative of the efforts and interest to conserve citrus trees. Diversity fair followed by four cell analysis and *in situ* evaluation resulted in the identification of 11 superior clones of pummelo. These studies are in agreement of the previous studies in citrus where the Midsweet, Sunstar and Gardner varieties of sweet orange were selected as the naturally occurring seedling cultivars (Hearn, 1988). The citrus varieties presently grown are mainly the selections from bud-sport mutations and chance seedlings (Pena and Navarro, 1999).

Owing to the presence of natural variability, selection of superior varieties can be developed by this participatory method, which will encourage the cultivation and consumption of pummelo for improving the health and economic status of the farmers, maintaining these

plants. The most important is on-farm conservation for sustainable conservation and utilization of pummelo.

Acknowledgements

The financial assistance received under UNEP/GEF-supported regional project "Conservation and sustainable use of cultivated and wild tropical fruit diversity: promoting sustainable livelihoods, food security and ecosystem services" is duly acknowledged.

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