The Pharma Innovation

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Characterization of walnut genotypes selected from district Chamba for qualitative characters using DUS test guidelines

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Abstract
The present study was carried out in the Chamba district of Himachal Pradesh during 2019–20 and 2020–21 to characterize the seedling tree population of walnut (*Juglans regia* L.). Based on non-metric characters, 98 seedling walnut trees were categorized and the results showed significant variation that allowed for the addition of better types to the existing walnut diversity. As per the DUS test guidelines, all types of classes were found for the tree growth habit character, *viz.* erect (42 genotypes), spreading (31 genotypes) and semi-erect (25 genotypes). The shape of the leaflet varied from elliptic (41) to broad elliptic (35) to narrow elliptic (22). Bearing was observed to be terminal in all the genotypes. Variation in nut shape in ventral view, i.e., circular (25), broad ovate (15), oblong (31), broad elliptic (16), ovate (7) and elliptic (4); nut shape in lateral view, i.e. oblate (21), circular (65), ovate (10) and broad elliptic (2); nut shape in cross section i.e. oblate (64), circular (33) and reniform (1); shape of base in lateral view i.e. rounded (74), truncate (9), cuneate (6) and emarginate (9); shape of apex in lateral view i.e. rounded (38), truncate (18), obtuse (35) and emarginate (7) were also observed. Nut weight was found to be light in 32 genotypes, heavy in 4 genotypes and medium in the remaining genotypes. Commercially important characters- kernel percentage was low (51) and medium (47); kernel colour was dark amber (13), amber (46), light (33) and extra light (6). Kernel removal was easy (34), moderate (32) and difficult (32), whereas shell thickness was thick (14), medium (81) and thin (3). These characters were highly significant in selecting desirable types.

Keywords: Walnut genotypes, qualitative characters, DUS test guidelines

Introduction

Walnut (*Juglans regia* L.), also known as Persian walnut, is one of the oldest (Sze-Tao and Sathe, 2000; Vahdati *et al.*, 2019) [57, 58] and most common nut crops grown in temperate regions (Bernard *et al.*, 2019; Gaisberger *et al.*, 2020) [13, 23], which has a great scope as a fruit for diversification of fruit crops (Rana *et al.*, 2007) [47]. It belongs to the family Juglandaceae and has a 2n = 32 chromosome number (Woodworth, 1930; Elias, 1972; Kefayati *et al.*, 2019; Bernard *et al.*, 2021) [62, 21, 31, 14]. Genus Juglans consists of approximately 21 species (McGranahan and Leslie, 1991; Ciarmiello *et al.*, 2013; Arab *et al.*, 2019; Gao *et al.*, 2019) [42, 18, 9, 24] and Juglans regia is the most important and commonly grown species in many countries. It is believed to have originated in Iran and its surrounding areas. It requires between 700–1500 hours of chilling. Walnut can be cultivated in all parts of the Himalayan region between elevations ranging from 1100 to 3000 metres above mean sea level (Rana *et al.*, 2007) [47].

Persian walnut is an important fruit crop, commercially grown for excellent quality nut production and is widely grown in the USA, China, France, Italy, Turkey, Poland, Japan and Austria. In India, earlier production of walnut was limited to Jammu and Kashmir and it appears that it only spread to the hills of Himachal Pradesh and Uttarakhand during the last century. Surprisingly, the majority of walnut output originates from seedling trees, which have a wide range of nut size, shape, flavour, shell thickness and quality of kernel (Gazmend *et al.*, 2005) [25]. In India, it is cultivated in an area of 1,07,000 hectares with a production of 2,96,000 metric tons (Anonymous, 2020) [8]. In Himachal Pradesh, it covers an area of 4,406 hectares with a production of 2,872 metric tons (Anonymous, 2019) [7].

Walnuts are divided into three groups based on the hardness of their shells: thin, medium, and hard shelled walnuts (Nainwal and Nainwal, 2013) [45]. Despite the fact that all three varieties are utilized for table purposes, some types of walnuts that are widely grown in Himachal Pradesh and neighboring states are not used for table purposes due to the presence of a very hard shell.

As a result, such fruits may be used to extract oil for a variety of purposes.

Characterization helps in recording and compilation of data for important characteristics that distinguish genotypes among a species or between species and permits simple and quick discrimination among them (Sajwan *et al*., 2020) [51]. It aids in simple accession grouping, gap identification and extraction of valuable germplasm for breeding programs, leading to a larger understanding of the composition of germplasm collections and genetic diversity. It conjointly allows a check on the true-to-type nature of undiversified samples, permitting the detection of misidentifications or duplicates, as well as highlighting possible errors created throughout alternative operations.

Although, the characterization and analysis work on selected walnut genotypes has been disbursed earlier by many workers. The study was advised to examine the performance (qualitatively as well as quantitatively) of those walnut genotypes and conjointly help with the identification, description, assessment of uniformity of traits and stabilization of their expression at completely different locations over a period of time. Keeping in sight its significance, the qualitative characterization of completely different walnut genotypes was performed at different areas of district Chamba (HP).

Material and Methods

This study was conducted from 2019 to 2021 on 98 walnut genotypes. The survey included Mehla, Saluni, Tissa, Bharmaur and Bhattiyat areas (Blocks) of the district of Chamba. The study area was located between - N$32^0$19'9.16968" to N$32^0$49'57.70236" in latitude and E$75^0$56'58.70436" to E$76^0$33'44.73" in longitude with altitude ranges from 1470 to 2154 meters above mean sea level, covering the existing seedling tree population of walnut in the Chamba district of Himachal Pradesh. Initially, the entire area was surveyed and the number of trees was marked on the basis of visual observations and discussions with local people. After a thorough screening, 98 walnut genotypes of seedling origin were identified and geo-referenced for detailed study. The selected trees were healthy, mature, and had a full crop and could grow naturally with no proper management practice. These selected genotypes were named based on their location and these names were supplemented with numerical characters. Seedling trees were more than 7 years (estimated average) of age, and some trees were well over 100 years of age.

We recorded observations for two consecutive years on various morphological, phenological, and carpological characteristics. The genotypes were characterized for 29 morphological characters at specific growth stages when the characters had full expression, as per IPGRI (1994), PPV&FRA (2012) and UPOV (2017) descriptors. Among 29 characters, 2 (Kernel flavor, Kernel shrivel) characters were characterized as per IPGRI descriptor, 9 (Nut: shape in ventral view, Nut: shape in lateral view, Nut: shape in cross section, Nut: shape of base in lateral view, Nut: shape of apex in lateral view, Nut: length of tip, Nut: extent of pad around suture, Nut: width of pad on suture in lateral view, and Nut: structure of surface of shell) characters were characterized as per UPOV descriptor, and 18 (Tree: growth habit, Tree: density of branches, Bearing habit, Shoot colour, Leaf: Leaflet length, Leaf: Leaflet Shape, Leaf: Leaflet Margin, Leaf: Leaflet Colour, Leaf: Leaflet pubescence, Nut diameter,

Nut length, Nut weight, Shell colour, Shell Thickness, Kernel Weight, Kernel Percentage, Kernel Colour and Ease of removal of kernel halves) characters were characterize on the basis of PPV&FRA descriptor. Observations for the assessment of distinctiveness, uniformity and stability were made on plants. All observations on the tree and the branches were made to activate during dormancy. For foliage characters, we randomly picked twenty-five fully matured leaflets from the middle portion of the current season's flush at the end of summer growth. Similarly, for nut and kernel characters, 10 representative fruit samples were taken at optimum maturity and observations were recorded. Assorted characters were classified into different groups and notes were assigned as shown below (Table 1):

Results and Discussion

The ninety-eight seedling genotypes included in this study were evaluated for growing in different parts of District Chamba. Geographical variety has resulted in widespread genetic diversity in walnuts. Therefore, the germplasm has been evaluated in an effort to assess its diversity and various descriptive and qualitative characteristics. According to studies on tree vigour and growth traits, the seedling walnut genotypes cultivated at Chamba show remarkable variability. In the present study, huge variation was reported in tree and foliage characteristics (Figure 1) such as tree: growth habit; leaf: leaflet length; leaf: leaflet shape; leaf: leaflet colour; leaf: leaflet pubescence and shoot colour except for bearing habit and leaf: leaflet margin, where no apparent variation was found.

All the 98 genotypes were found to have terminal bearing and had the "entire" type of leaf: leaflet margin. The results for the character terminal bearing are in accordance with the findings of Joolka and Sharma (2005) [29], Poggetti *et al*. (2017) [46] and Mir *et al*. (2018) [43] who found that most of the seedling walnut trees were terminal bearers in their study. However, Botu *et al*. (2010) [15], Solar *et al*. (2002) [56], Akhiani *et al*. (2017) [2] and Rouskas and Zakynthinos (2001) [50] also reported some lateral-bearing genotypes in their studies. Besides, the results for leaf: leaflet margin are in agreement with those observed by Angmo *et al*. (2013) [3], Angmo *et al*. (2015) [4] and Kabiri *et al*. (2018) [30] who also found the "entire" type of leaf margin in their study. Considering the current study, out of 98 genotypes, 42 (42.86%) had erect, 25 (25.51%) had semi-erect and the rest of the 31 (31.63%) had a spreading type of growth habit. This variability observed in tree growth habits might be caused by factors such as age, soil fertility, and environmental factors (Lone, 2017; Waheed *et al*., 2021) [38]. A similar type of variation was reported by Shah *et al*. (2020) [52], *viz.,* 49 genotypes (58.33%) exhibited a spreading growth habit, 6 genotypes (7.14%) were upright in growth habit, and the remaining showed a semi-upright growth habit. Ahandani *et al*. (2014) [1] also found walnut accession's growth habit as semi-upright to spreading type. Angmo *et al*. (2015) [4] found in their study that spreading growth patterns were more prevalent than semi-erect ones. Rezaei *et al*. (2017) [49] also reported variation in tree habits in their study. Tree vigour and growth habits, for example, are key parameters for yield or yield efficiency. The bearing potential of a walnut tree with strong vigour and spreading behaviour is increased by growing more lateral, sub terminal, or terminal leafy shoots. Because spreading type trees have limited apical dominance, their IAA cumulative effect is diminished (Chanda *et al*., 2017) [17]. The highest percentage

of tree: density of branches falls in the group of intermediate (77.55%), followed by sparse (13.27%) and dense (9.18%). A similar kind of variation for the density of branches was also reported by Simsek et al. (2017) [55], viz., sparse, very sparse, medium and dense. Shah et al. (2020) [52] discovered that 52.38 percent of the genotypes studied had dense branches, followed by 36.90 percent of genotypes with medium density

of branches, 8.33 percent of genotypes with sparse density of branches, and 2.038 percent of genotypes with very dense density of branches. In their study on walnuts, Lone (2017) [38] found that 30 genotypes had sparse branches, 57 genotypes had intermediate branches, and 65 genotypes had dense branches.

Table 1: Descriptor of the qualitative characters utilized for the studied walnuts (Scoring the studied nut characteristics based on IPGRI, PPV&FRA and UPOV descriptor)

Variable	Code and state						
	1	3	5	7	9	11	13
Tree: Growth Habit	Erect	Semi erect	Spreading				
Tree: Density of branches	Sparse	Intermediate	Dense				
Bearing Habit	Terminal	Lateral					
Leaf: Leaflet length (cm)	Short (< 10)	Medium (10-15)	Long (>15)				
Leaf: Leaflet shape	Narrow elliptic	Elliptic	Broad elliptic				
Leaf: Leaflet margin	Entire	Serrate	Dentate				
Leaf: Leaflet colour	Light green	Green	Dark Green	Purplish			
Leaf: Leaflet pubescence	Glabrous	Slightly pubescent	Pubescent				
Shoot Colour	Green	Brown	Dark brown				
Nut length (mm)	Small (< 30)	Medium (30-40)	Large (>40)				
Nut diameter (mm)	Small (<30)	Medium (30-40)	Large (> 40)				
Nut weight (g)	Light (<15)	Medium (15-20)	Heavy (>20)				
Shell colour	Very light	Light	Medium	Dark			
Nut: shape in ventral view	Triangular	Broad ovate	Ovate	Oblong	Elliptic	Circular	Broad elliptic
Nut: shape in lateral view	Triangular	Broad ovate	Ovate	Circular	Broad elliptic	Oblate	
Nut: shape in cross section	Reniform	Oblate	Circular	Elliptic			
Nut: shape of base in lateral view	Cuneate	Rounded	Truncate	Emarginate			
Nut: shape of apex in lateral view	Obtuse	Rounded	Truncate	Emarginate			
Nut: length of tip	Absent or short	Medium	Long				
Nut: extent of pad around suture	On upper half	On upper 2/3 portion	On whole length				
Nut: width of pad on suture in lateral view	Narrow	Medium	Broad				
Nut: shell surface	Moderately smooth	Smooth	Rough				
Ease of removal of kernel halves	Easy	Moderate	Difficult				
Shell thickness (mm)	Thin (<1)	Medium (1-2)	Thick (>2)				
Kernel weight (g)	Light (<6)	Medium (6-10)	Heavy (>10)				
Kernel percentage	Low (<40)	Medium (40-50)	High (50-60)	Very high (>60)			
Kernel colour	Extra light	Light	Amber	Dark amber			
Kernel flavour	Satisfactory	Unsatisfactory					
Kernel shrivel (%)	Kernel exhibiting tip shrivel	Kernel exhibiting < 50% shrivel	Kernel exhibiting 50% or more shrivel	Kernel blank			

In 55.10 percent of genotypes, leaf: leaflet length was medium, followed by long in 44.90 percent, whereas, no genotype had a short leaflet length. This is consistent with the findings of Mir et al. (2018) [43], who also noted a range of leaflet lengths, including short, medium, and long. Among the studied genotypes in terms of leaf: leaflet shape as presented in Figure 1, 41.84 percent of genotypes had an elliptic leaflet shape, followed by 22.45 percent of genotypes that had a narrow elliptic leaflet shape, and 35.71 percent of genotypes that had a broad elliptic leaflet shape, which is in line with the findings of Shah et al. (2020) [52] viz., elliptic (53.57%), followed by narrow elliptic (27.38%) and broad elliptic (19.04%). Sajwan et al. (2020) [51] also reported narrow elliptic, elliptic and broad elliptic types of leaflet shape variation during their survey in the three districts of Himachal Pradesh. In the case of leaf: leaflet colour, 48 genotypes had green colour, whereas, 50 genotypes had dark green colour. Leaf: leaflet pubescence was predominantly slightly pubescent in 90.82 percent of genotypes and pubescent in

9.18 percent of genotypes, but no genotypes were reported to have glabrous pubescence. The majority of the genotypes (91.84%) had brown and a few (8.16%) had green shoot colour. However, no genotype had a dark brown shoot colour as shown in Figure 1. The leaflet colour, leaf: leaflet pubescence, and shoot colour conform to the findings of Shah et al., 2020; Sharma et al., 2014; Angmo et al., 2015; Mir et al., 2018; Sajwan et al., 2020) [52, 4, 54, 43, 51].

In the present survey, the nut length in 1.02 percent of genotypes was small, medium in 54.08 percent of genotypes and large in 44.90 percent of genotypes. The majority of the genotypes (95.92%) had medium nut diameters, while a few (4.08%) had small. However, no genotype had a large nut diameter. Considering nut weight, 32.65 percent of genotypes fell into the class of "light", 63.27 percent in the class "medium" and only 4.08 percent in "heavy". While comparing 27 walnut genotypes, Mir et al. (2018) [43] observed a significant degree of variability in the cases of nut length, net diameter, and nut weight. Only 3 genotypes were found to

have a small nut diameter, 10 had a medium nut diameter and 14 genotypes had a large nut diameter. In terms of nut length, 14 genotypes had large nuts, 11 had medium nuts, and 2 had small nuts. In contrast, their study found that in terms of nut weight, 17 genotypes had heavy nuts, 10 had medium nuts, and only 2 genotypes had light nuts. Nut: shape (in lateral view) varied greatly and included ovate (10.20%), circular (66.33%), broad elliptic (2.04%) and oblate (21.43%) genotypes, but no triangular and broad ovate shaped genotypes were reported (Figure 2). Shah et al. (2020) [52] also found variation in nut shape as broad elliptic/elliptic (39.28%), circular (28.57%), broad ovate/ovate (21.42%), broad trapezium/trapezium (7.14%) and triangular (3.57%), while evaluating 84 walnut seedlings. However, Kouhi et al. (2020) [37] found mostly ovate, round and elliptic types of nut shape, whereas Mirmahdi and Khadivi (2021) [44] reported nut shape variation in genotypes as round (68), board oval (49), oval (70), board ovate (93), ovate (58), trapezoidal (10) and triangular (14) in their trials. The results of the present study

are in accordance with the various previous workers (Caglarırmak, 2003; Fatahi et al., 2010; Asma, 2012; Ghanbari et al., 2018; Angmo et al., 2013; Simsek et al., 2017; Sajwan et al., 2020) [16, 22, 3, 12, 26, 55, 51]. The nut shape (in cross section) was oblate (in 65.31% of genotypes), circular (in 33.67% of genotypes) and reniform (1.02% of genotypes) in the present study. Mir et al. (2018) [43] also found most of the genotypes with oblate (14), followed by 8 genotypes with a round shape and 5 genotypes with an elliptic type of nut shape in cross section. The nut shape (in ventral view) varied from triangular, broad ovate, ovate, oblong, elliptic, circular to broad elliptic. In the present 16 genotypes (16.33%) exhibited a broad elliptic shape, 7 genotypes (7.14%) had ovate, 31 genotypes (31.63%) exhibited oblong, 4 genotypes (4.08%) had elliptic, 25 genotypes (25.51%) had circular, and the rest of the other 15 genotypes (15.31%) had a broad ovate nut shape and no genotypes were categorized under the triangular group (Figure 2).
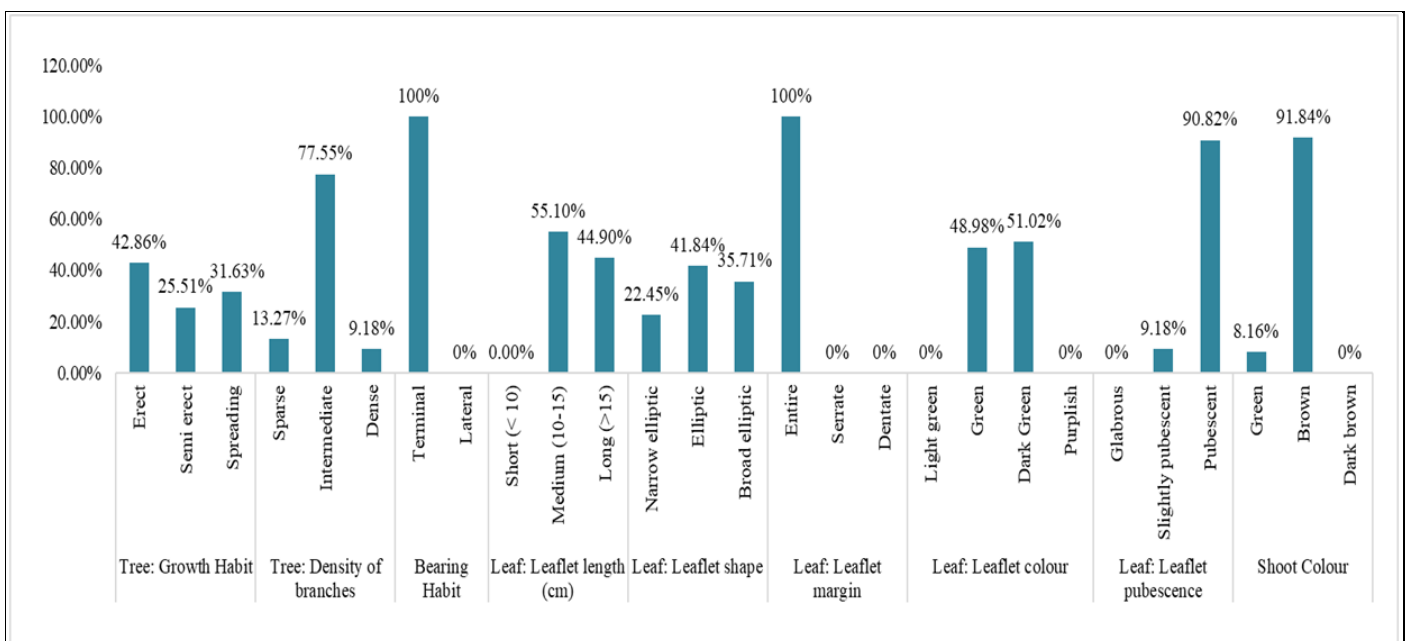

Fig 1: Frequency distribution of 98 walnut accessions based on tree and foliage characters

However, Sajwan et al. (2020) [51] reported to have 37.80 percent broad elliptical genotypes, followed by 24.40 percent ovate, 22.20 percent broad ovate and 15.60 percent circular in their survey. Which is in conformity with the findings of Kilicoglu and Akca, (2018) [36] and Jacimovic et al. (2020) [28] as they also find the same type of variation for nut shape (in ventral view). Nut: shape of the base (in lateral view) was determined as cuneate (in 6.12% of genotypes), rounded (75.51%), truncate (9.18%) and emarginate (9.18%) in the present study, which is in agreement with the findings of Mir et al. (2018) [43] viz., cuneate, rounded, truncate and emarginate.

Among the studied genotypes in terms of nut: shape of apex (in lateral view), 35.71 percent of genotypes were obtuse, 38.78 percent were rounded, 7.14 percent were emarginate and 18.37 percent were of truncate shape. According to Sajwan et al. (2020) [51], 40 percent of the genotypes had an obtuse shape of apex, 28.9 percent had a rounded, 17.8 percent had a truncate, and 13.3 percent had an emarginated. Kilicoglu and Akca (2018) [36] also reported similar kind of variation as pointed and rounded for most of the genotypes

and emarginated and truncate for very few genotypes in their study. However, Jacimovic et al. (2020) [28] reported only pointed and rounded types of variation in their trial, but no genotype was reported to have a truncated or emarginated type of shape of apex. Nut: length of tip was absent or short in 18.37 percent of the genotypes, medium in 54.08 percent of the genotypes and long in 27.55 percent of the total genotypes evaluated. Kilicoglu and Akca (2018) [36] found that the majority of the genotypes under evaluation had medium prominence of the apical tip (14 genotypes), whereas 4 had strong prominence and only 2 had weak prominence. Whereas, Jacimovic et al. (2020) [28] assessed 22 genotypes and discovered that the majority of the genotypes had weak to medium prominence of the apical tip, with a few having strong prominence. Amidst the studied genotypes in terms of nut: extent of pad around suture (Figure 2), 38.78 percent of genotypes had suture on the upper half, 51.02 percent had it on the upper 2/3 portion and 10.20 percent had it on the whole length of the nuts. However, Sajwan et al. (2020) [51] discovered that 55.6 percent of genotypes had suture position on the upper half, 35.6 percent had suture on the upper 2/3

portion, and 8.9 percent had suture on the entire length. Similar results were also reported by Kilicoglu and Akca (2018) [36] and Jacmovie *et al.* (2020). Nut: width of pad around suture (lateral view) was determined as narrow (2.04%), medium (92.86%) and broad (5.10%) in the present study. Similar kind of variation (narrow, medium and broad) for width of pad on suture was also reported by the previous workers (Kilicoglu and Akca, 2018 and Jacimovic *et al.*, 2020) [36, 28].
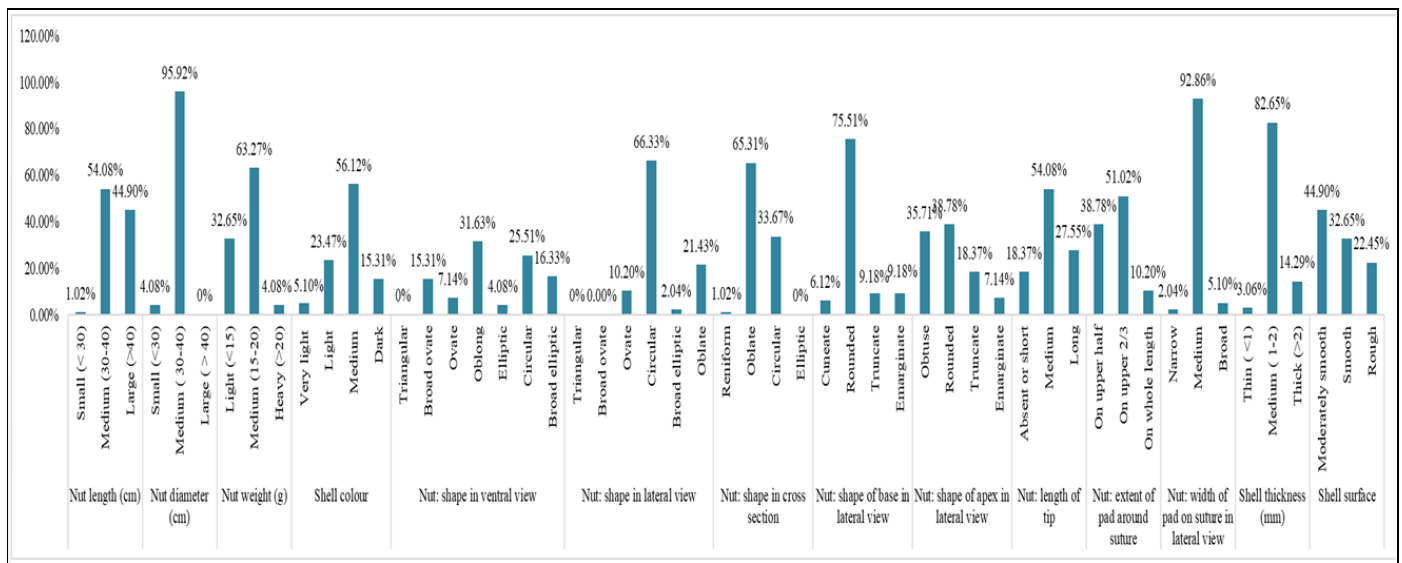

Fig 2: Frequency distribution of 98 walnut accessions based on nut characters

The data on shell thickness revealed that about 81 of the 98 genotypes had medium shell thickness, 14 genotypes had thick shells, and 3 genotypes had thin shells. Ghanbari *et al.* (2018) [26] reported variations of a similar nature for shell thickness. However, Simsek *et al.* (2017) [55] found very thin, thin and medium-sized variations in shell thickness. Among all of the genotypes being studied, 44.90 percent of them were moderately smooth, 32.65 percent showed a smooth texture, and 22.45 percent had a rough nut: shell surface. Khadavi-Khub (2014) discovered that shell texture was smooth in 27 percent of accessions, medium in 41 percent, and rough in 32 percent. However, Kouhi *et al.* (2020) [37] found smooth and medium shell surfaces during their study and Verma *et al.* (2020) found smooth, medium and rough types of shell surfaces. Shell colour among the genotypes revealed that 5.10 percent of the genotypes had very light shell colour, 23.47 percent of the genotypes had light colour, 56.12 percent of the genotypes had medium colour and 15.31 percent of the genotypes showed dark shell colour. The shell colour of several walnut accessions was also noted by Joolka and Sharma (2005) [29] to be very light and light to medium, while Asma (2012) [12] found light and dark shell colour. However, while assessing 11 genotypes, Verma *et al.* (2020) [59] identified light, medium, and dark shell colour.

Among the samples considered in our study (Figure 3), 6 (6.12%) samples had an extra light kernel colour, 33 (33.67%) were light, 46 (46.94%) had amber, and 13 (13.27%) were dark amber coloured. Khan *et al.* (2017) [35] also reported the variation in kernel colour as extra light, light, light amber and amber in their evaluated genotypes, while Keles *et al.* (2014) [32] reported that five selected genotypes had extra light, seven

had light-coloured kernels and only two genotypes had dark-coloured kernels. However, while evaluating 84 genotypes for different characteristics, Shah *et al.* (2020) [52] reported 35 genotypes had light, 33 had light amber, and 16 had amber-coloured kernel colour. The findings of the present work are in agreement with various previous researchers (Arzani *et al.*, 2008; Fatahi *et al.*, 2010; Khadivi-Khub *et al.*, 2015; Sharma *et al.,* 2014 and Joolka and Sharma, 2005) [10, 54, 33, 22, 29]. Commercial growers experience a financial loss as a result of the unmarketability of ambered walnut kernels (Warmund, 2014) [61]. Ambered kernels often contain less stearic and oleic acid than nonambered kernels, according to Mangoff (1980) [40]. The genotypes with light-colored kernels are the most significant from an economic standpoint (McGranahan and Leslie, 1991; Cosmulescu *et al.*, 2010; Botu *et al.*, 2010; McGranahan and Leslie, 2012; Ma *et al.*, 2013) [42, 19, 41, 15]. Besides, ease of kernel removal was easy in 34 (34.69%) genotypes, 32 (32.65%) had moderate and 32 (32.65%) of genotypes showed difficult results in ease of kernel removal (Figure 3). Similar results were found by Arzani *et al.* (2008) [10] and Khadivi-Khub (2014) [34] as very easy, easy, moderate, and difficult. Regarding ease of kernel removal, 136 of the samples were classified as "very easy removal", 22 as "easy", 12 as "intermediate", 8 as "difficult" and just 6 as "very difficult" (Poggettia *et al.*, 2017) [46]. Jacimovic *et al.* (2020) [28] reported very easy to easy (ease of kernel removal) for most of the accessions and difficult for one accession only. Typically, the genotypes with the most attractive kernels are ones whose shells are the easiest to remove (McGranahan and Leslie, 1991; Cosmulescu *et al.*, 2010; Botu *et al.*, 2010; McGranahan and Leslie, 2012) [42, 19, 41, 15].
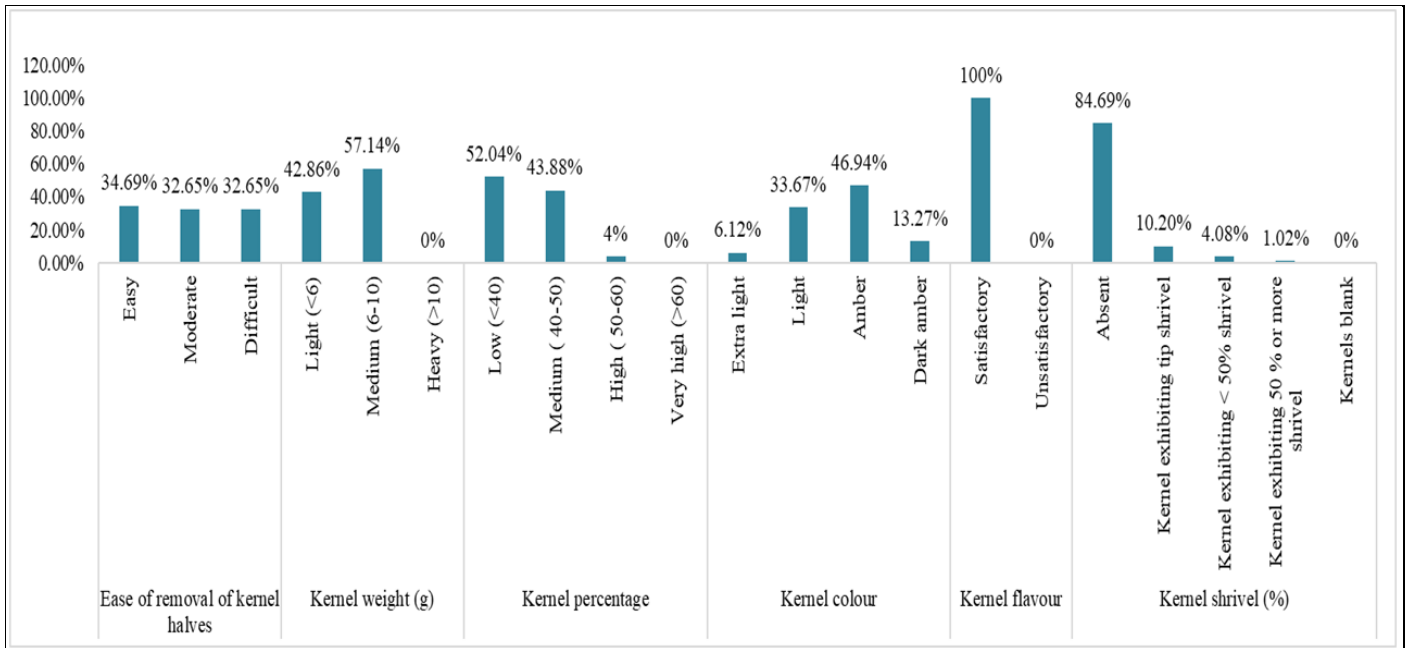
Fig 3: Frequency distribution of 98 walnut accessions based on kernel character

Nut diameter, shell hardness, shell texture, kernel weight, kernel percentage, kernel fill and kernel plumpness all have an impact on how easily the kernel may be removed. As a result, nuts with smooth shell textures will make it simpler to extract the kernels. Nuts with thicker shells contained fewer and smaller kernels (Khadivi-Khub *et al*., 2015) [33]. Superior walnut genotypes have been chosen based on the colour of the kernel and the ease of kernel removal (Sharma and Sharma, 2000; Yarilgac *et al*., 2001; Zeneli *et al*., 2004; Aslantas, 2006; Arzani *et al*., 2008; Cosmulescu and Botu, 2012) [53, 10, 63, 20]. Kernel shrivel was absent in majority of the genotypes (84.69%), with 10.20 percent of the genotypes' kernels exhibiting tip shrivel, 4.08 percent of the genotypes' kernels exhibiting less than 50 percent shrivel, 1.02 percent of the kernels exhibiting 50 percent or more shrivel and none of the genotypes were reported blank. This is in line with the conclusions obtained by Sajwan *et al*. (2020) [51], where they found 82.2 percent of genotypes with no shrivel and 17.8 percent of genotypes with kernels exhibiting tip shrivel in their conducted survey. Considering the present study, 42 genotypes (42.86%) were reported to have light (<6) and 56 genotypes (57.14%) had medium (6-10) kernel weight. However, Mir *et al*. (2018) [43] reported 14 genotypes with heavy kernel weight, 8 with medium and only 3 genotypes with light kernel weight. Kernel percentage was low (52.04% genotypes), medium (43.88% genotypes) and high (4.08% genotypes) among 98 evaluated genotypes. For kernel flavour, all walnut accessions were found to be satisfactory, which is in accordance with the findings of Angmo *et al*. (2013) [3], Angmo *et al*. (2015) [4] and Kabiri *et al*. (2018) [30], who also reported satisfactory kernel flavour for all genotypes in their study. Non-significant differences in kernel flavour have also been reported by Ingels *et al*. (1990) [27] in previous studies.

Conclusion
Considering the significance of walnut genotype diversity, it is critical to survey these populations and identify superior genotypes for future breeding programs through in situ conservation and subsequent hybridization. As a result, the study was taken up to characterize and catalogue the local

gene pool, as well as to identify some promising genotypes for future improvement of this fruit crop species. So, after characterization of 98 walnut genotypes based on DUS test guidelines, nine genotypes with good kernel percentage, nut size, nut weight, kernel weight, thin to medium shelled nuts, easy to remove kernel halves, light to extra light coloured kernels, as well as moderately smooth to smooth shell surface, are recognized as superior genotypes and can be used to establish new orchards or improve cultivars. These traits could serve as selection indices for the development of ideotype cultivars in walnut. The best-known genotypes from this study must be used in future walnut breeding programs to introduce new cultivars and revitalize traditional walnut orchards in order to commercialize them.

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