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Characterization of phenotypic and floral diversity in seedling populations of *Punica granatum* L. in the North-Western Indian Himalayas

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Abstract
This study investigated the phenotypic and floral morphological variability among seedling-derived *Punica granatum* L. genotypes in the northwestern Himalayan region of India. Conducted over two growing seasons (2022-2023), a survey and characterization of 46 indigenous pomegranate genotypes were performed in the Doda and Kishtwar districts of the Jammu subtropics. Our results revealed significant variation in key floral traits, with specific genotypes exhibiting superior characteristics; for instance, JMU (Pom-2) displayed the longest calyx length and width (5.12 cm, 1.90 cm) and similarly JMU (Pom-2) the longest corolla (3.44 cm) and widest corolla (3.11 cm). Importantly, these seedling-origin genotypes demonstrated distinct morphological profiles compared to existing commercial cultivars. This identified phenotypic diversity represents a valuable genetic resource, offering promising avenues for future pomegranate breeding programmes aimed at crop improvement and adaptation.

Keywords: Pomegranate, *Punica granatum* L., floral morphology, phenotypic variability, genetic diversity, genotypes, breeding, Jammu region, India

Introduction
Pomegranate (*Punica granatum* L.) is one of the oldest known edible fruit crops, valued for its nutritional, medicinal, and economic significance worldwide (Hasnaoui *et al.* 2012) [2], (Parshuram *et al.* 2022) [4]. In recent decades, the species has garnered increasing scientific interest due to its remarkable health-promoting properties and adaptability to diverse agro-climatic conditions (Hasnaoui *et al.* 2012) [2], (Parshuram *et al.* 2022) [4]. The north-western Indian Himalayas, with their unique topography and climatic heterogeneity, represent an important center for pomegranate cultivation and genetic resource conservation.

Understanding the extent of phenotypic and floral diversity within seedling populations of *P. granatum* is fundamental for effective breeding, conservation, and sustainable utilization strategies. Previous studies have highlighted substantial morphological and pomological variation in both wild and cultivated pomegranate accessions across different regions, particularly in traits such as fruit weight, aril characteristics, peel color, and floral morphology (Ashrafi *et al.* 2023) [1], (Parshuram *et al.* 2022) [4]. For instance, research on wild pomegranate populations in Iran and India has revealed high diversity in fruit and flower traits, which are crucial for selecting superior genotypes and improving crop performance (Parshuram *et al.* 2022) [4], (Ashrafi *et al.* 2023) [1]. Despite these advances, comprehensive characterization of phenotypic and floral diversity in seedling populations-especially within the context of the north-western Indian Himalayas remains limited.

Moreover, the genetic base of cultivated pomegranate is often considered narrow, underscoring the need for detailed evaluation of existing variability to inform breeding programs and germplasm management (Hasnaoui *et al.* 2012) [2]. Integrative approaches combining morphological, floral, and molecular data have proven effective in elucidating the complex diversity patterns within *P. granatum* collections (Ashrafi *et al.* 2023) [1]. However, floral diversity, in particular, has received comparatively less attention, despite its relevance to reproductive biology and fruit set (Parshuram *et al.* 2022) [4].

This research aims to systematically characterize the phenotypic and floral diversity present in seedling populations of *Punica granatum* L. in the north-western Indian Himalayas. By documenting and analyzing this diversity, the study seeks to provide a scientific basis for germplasm conservation, breeding program enhancement, and the sustainable development of pomegranate cultivation in this ecologically significant region.

Materials and Methods

The present investigation of different pomegranate genotypes was carried out during 2022-2023 and 2023-2024 in the subtropical areas of Jammu subtropics. The various morphological analysis was performed at the Division of Fruit Science, Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu (SKUAST-Jammu).

Geographic area of the Study

The study site is located in the Northern Western Himalayan region of Jammu and Kashmir, encompassing parts of the Doda and Kishtwar districts. Doda district, in the eastern Jammu region, is characterized by hilly terrain at 33.080°N latitude and 75.320°E longitude, with an elevation of 1,107 meters. It experiences a relatively dry climate with limited rainfall (average 926 mm annually) primarily during the monsoon season (July-September), and an average snowfall of 135 mm. Kishtwar district, located in the northeastern Jammu region at 33°19'12''N and 75°46'12''E, lies along the Chenab River at 1,640 meters elevation. Its landscape is predominantly hilly and mountainous with valleys like Dachhan, Marwah, Paddar, and Chatroo. Kishtwar's climate ranges from subtropical to temperate, with summer daytime temperatures between 6°C and 27°C and average monthly temperatures in colder months ranging from-1°C to 13°C. It receives an average annual rainfall of 865 mm, with significant snowfall, especially on mountain peaks, contributing the majority of its precipitation.

Technical programme

The survey for seedling genotypes was conducted in major pomegranate growing areas of Doda and Kishtwar districts of Jammu province to select the most promising accession among the seedling pomegranate genotypes and to assess the variability in their morphological characteristics. The location was selected with respect to the availability of diversity in seedling pomegranate genotypes. During survey, 46 seedling pomegranate genotypes with divergent characters were selected at fruit maturity stage on the basis of size and fruit quality. Codes were allotted to each selection on the basis of their location and geo tagging was done on selected plants. The observations on flower morphological characteristics were recorded as per guidelines for the conduct of test for Distinctiveness, Uniformity and Stability of pomegranate (*Punica granatum* L.)) Protection of Plant Varieties and Farmers Right's Authority (PPV&FRA) Government of India along with the pomegranate Descriptor of International Union for the Protection of New Varieties of Plants (UPOV, 2013).

Results and Discussion

The table 1 shows the data on the calyx and corolla traits of different types of pomegranates. The calyx length of pomegranate genotypes ranged from 2.93 cm to 5.12 cm. The longest calyx length was 5.12 cm in genotype JMU (Pom-2), followed by 4.84 cm in JMU (Pom-34), and 4.60 cm in JMU (Pom-17) and 4.51 cm in JMU (Pom-45). Genotype JMU (Pom-4) had the shortest calyx, which was only 2.93 cm long. There was a lot of difference in the width of the calyx, which ranged from 1.12 cm to 1.90 cm. Genotype JMU (Pom-2) had the widest calyx at 1.90 cm, followed by JMU (Pom-25) at 1.87 cm and JMU (Pom-12) at 1.81 cm. JMU (Pom-19) and JMU (Pom-43) had the smallest calyx width, which was 1.12 cm. The length of the corolla in some pomegranate genotypes ranged from 2.13 cm to 3.44 cm. The longest corolla, 3.44 cm, was found in JMU (Pom-2). JMU (Pom-45) had the next longest corolla, 3.42 cm. JMU (Pom-5) had the shortest corolla length, which was 2.18 cm. The corolla width was between 1.42 cm and 3.11 cm. The widest corolla was 3.11 cm in JMU (Pom-2), followed by 2.94 cm in JMU (Pom-30) and 2.81 cm in JMU (Pom-21). JMU (Pom-6) and JMU (Pom-19) had the smallest corolla width, which were 1.42 cm and 1.43, respectively. These differences in floral morphology can be attributed to both genetic factors and environmental influences. Studies have shown that pomegranate accessions exhibit statistically significant variability in calyx and corolla dimensions, which are largely governed by genotype but can also be affected by local growing conditions and agronomic practices. Parashuram *et al.* (2022) [4] reported calyx lengths ranging from 2.74 cm to 4.37 cm and widths from 0.90 cm to 1.43 cm among 40 Indian pomegranate accessions, with significant coefficients of variation for these traits. Radunic *et al.* (2024) [5] found similar ranges in European pomegranate cultivars, with calyx lengths from 3.30 cm to 4.28 cm and widths from 1.36 cm to 1.98 cm. These findings confirm that the variability observed in the present study is consistent with global patterns of morphological diversity in pomegranate.

The figure 1 shows the number of genotypes and frequency percent of calyx colour of pomegranate (*Punica granatum* L.) genotypes from the Jammu region. The data revealed that the calyx colour of the genotypes was categorized into four types: orange, orange red, Medium red, and Dark red. Out of the observed genotypes, 11 genotypes (23.91%) had an orange calyx colour. The highest percentage was found in the Orange red category, with 19 genotypes (41.3%) exhibiting this colour. These results align with the findings of Khadivi *et al.* (2020) [3]. For Medium red calyx colour, there were 13 genotypes, accounting for 28.26% of the total. Lastly, only 3 genotypes (6.52%) showed a Dark red calyx colour. The diversity in calyx and corolla colouration is primarily attributed to genetic variation among the genotypes, as supported by earlier studies (Parashuram *et al.*, 2022; Radunic *et al.*, 2024) [4, 5]. Environmental factors, such as light intensity, temperature, and soil nutrients, may also play a role in the expression of floral pigments (Ashrafi *et al.*, 2023) [1]. The predominance of orange red and medium red hues suggests that these colour phenotypes may be adaptive or preferred in the local agro-climatic conditions of Jammu, possibly influencing pollinator attraction and fruit set.

The figure 2 displays the number of genotypes and frequency percent of corolla colour of pomegranate (*Punica granatum* L.) genotypes from the Jammu region. The data indicated that the corolla colour of the genotypes was categorized into six types: White, Pink, Light orange, medium, red, and medium red. No genotypes (0%) exhibited a White corolla colour. For Pink corolla, 8 genotypes (17.39%) were observed. Light orange corolla was found in 4 genotypes (8.69%), while Medium orange corolla was present in 3 genotypes (6.52%). The most prevalent corolla colour was orange red, with 19 genotypes (41.3%) showing this trait. Lastly, 12 genotypes (26.08%) displayed a Medium red corolla colour. Floral colour traits, including calyx and corolla pigmentation, are important descriptors in pomegranate taxonomy and breeding. The observed variability aligns with the findings of Radunić *et al.* (2024) [5], who noted significant diversity in floral and fruit colour among European pomegranate cultivars. Colour traits are often used to distinguish between genotypes and can be linked to consumer preferences and marketability.

The figure 3 illustrates the number of genotypes and frequency percent of petal surface characteristics of pomegranate (*Punica granatum* L.) genotypes from the Jammu region. The data revealed that the petal surface of the genotypes was categorized into three types: smooth, moderately wrinkled, and strongly wrinkled. Ten genotypes (21.73%) had a Smooth petal surface. The highest percentage was found in the Moderately wrinkled category, with 22 genotypes (47.82%) exhibiting this trait. Fourteen genotypes (30.43%) displayed a Strongly wrinkled petal surface. Variation in petal surface texture is primarily governed by genetic factors, as supported by previous studies on pomegranate floral morphology (Parashuram *et al.*, 2022; Radunić *et al.*, 2024) [4, 5]. Environmental conditions and developmental factors may also influence petal surface characteristics, though to a lesser extent than genetic makeup.

The figure 4 presents the number of genotypes and frequency percent of the number of flowers per node for pomegranate (*Punica granatum* L.) genotypes from the Jammu region. The data revealed that the number of flowers per node was categorized into four groups: One, Two, Three, and More than three. Four genotypes (8.69%) produced one flower per node. The majority of genotypes, 24 (52.17%), had two flowers per node. Fourteen genotypes (30.43%) displayed three flowers per node. Lastly, four genotypes (8.69%) had more than three flowers per node. The number of flowers per node is a key reproductive trait influenced by both genetic factors and environmental conditions. Previous studies have demonstrated that this trait varies widely among pomegranate genotypes and is strongly affected by genotype-specific flowering habits (Parashuram *et al.*, 2022; Radunić *et al.*, 2024) [4, 5]. Environmental factors such as temperature, light, and nutrient availability can also modulate flower production, but the predominance of two and three flowers per node in this collection suggests a genetic predisposition toward higher floral density in the Jammu region germplasm.

The figure 5 illustrates the number of genotypes and frequency percent of the time of beginning of flowering for pomegranate (*Punica granatum* L.) genotypes from the Jammu region. The data revealed that the onset of flowering occurred across several weeks from April to May. Four genotypes (8.69%) began flowering in the 3rd week of April. Five genotypes (10.86%) started in the 4th week of April. Seven genotypes (15.21%) commenced flowering in the 1st week of May, and 10 genotypes (21.73%) began in the 2nd week of May. The highest percentage of genotypes, 17 (36.95%), started flowering in the 3rd week of May. Finally, 3 genotypes (6.52%) began flowering in the 4th week of May. Variation in flowering time is a critical trait for both agronomic management and breeding. Early-flowering genotypes can be advantageous for extending the harvest season or escaping late-season pests and diseases, while later-flowering genotypes may be better suited to areas prone to early spring frosts. The presence of genotypes with staggered flowering times provides breeders with the opportunity to develop cultivars tailored to different climatic zones and market needs. Similar patterns of phenological diversity have been reported in other pomegranate collections. For example, Ashrafi *et al.* (2023) [1] observed a wide range of flowering times among wild pomegranate populations in Iran, attributing this variation to both genetic background and local environmental adaptation. Radunić *et al.* (2024) [5] also noted that European pomegranate cultivars exhibited significant differences in flowering onset, which could be exploited for breeding.

Table 1: Variation in calyx length, calyx width, corolla length and corolla width of pomegranate (*Punica granatum* L.) genotypes of Jammu region

Genotypes	Calyx Length	Calyx Width	Corolla Length	Corolla Width
JMU (Pom-1)	4.40	1.53	2.46	1.60
JMU (Pom-2)	5.12	1.90	3.44	3.11
JMU (Pom-3)	3.95	1.19	2.18	1.95
JMU (Pom-4)	2.93	1.34	2.55	2.23
JMU (Pom-5)	4.63	1.58	2.13	1.48
JMU (Pom-6)	4.39	1.33	2.53	1.42
JMU (Pom-7)	4.17	1.32	2.85	2.23
JMU (Pom-8)	3.50	1.25	2.54	2.17
JMU (Pom-9)	4.40	1.73	2.33	2.09
JMU (Pom-10)	3.71	1.37	2.44	2.21
JMU (Pom-11)	3.24	1.23	2.57	1.92
JMU (Pom-12)	4.02	1.81	2.70	2.04
JMU (Pom-13)	4.27	1.87	2.16	1.86
JMU (Pom-14)	3.01	1.30	2.61	1.83
JMU (Pom-15)	3.53	1.38	2.91	2.01
JMU (Pom-16)	3.66	1.21	3.45	2.37
JMU (Pom-17)	4.60	1.24	3.29	2.80
JMU (Pom-18)	3.12	1.47	2.80	1.96
JMU (Pom-19)	3.50	1.12	2.36	1.43
JMU (Pom-20)	4.21	1.32	2.94	1.65
JMU (Pom-21)	3.61	1.24	3.28	2.81
JMU (Pom-22)	4.30	1.58	2.13	1.86
JMU (Pom-23)	4.31	1.24	2.74	1.82
JMU (Pom-24)	3.86	1.41	3.04	1.96
JMU (Pom-25)	3.95	1.55	3.00	2.04
JMU (Pom-26)	3.90	1.57	2.53	1.84
JMU (Pom-27)	3.72	1.43	2.77	2.17
JMU (Pom-28)	4.06	1.49	2.46	1.74
JMU (Pom-29)	4.23	1.73	2.64	1.90
JMU (Pom-30)	3.51	1.32	3.15	2.94
JMU (Pom-31)	3.75	1.51	2.53	1.92
JMU (Pom-32)	3.53	1.21	2.90	1.54
JMU (Pom-33)	4.21	1.65	2.87	2.06
JMU (Pom-34)	4.84	1.60	2.50	1.71
JMU (Pom-35)	4.20	1.36	2.39	1.76
JMU (Pom-36)	3.80	1.40	3.64	1.93
JMU (Pom-37)	3.72	1.24	3.19	2.79
JMU (Pom-38)	3.57	1.26	3.12	2.53
JMU (Pom-39)	3.25	1.43	2.94	2.20
JMU (Pom-40)	3.45	1.47	3.31	1.93
JMU (Pom-41)	3.64	1.64	2.49	1.52
JMU (Pom-42)	3.12	1.44	2.86	1.74
JMU (Pom-43)	3.11	1.12	2.71	1.96
JMU (Pom-44)	4.53	1.41	3.18	2.43
JMU (Pom-45)	4.51	1.24	3.42	2.07
JMU (Pom-46)	4.56	1.35	2.77	2.20
Mean	3.90	1.42	2.78	2.03
Standard Deviation	0.51	0.19	0.37	0.39
CV	13.3	13.58	13.39	19.32

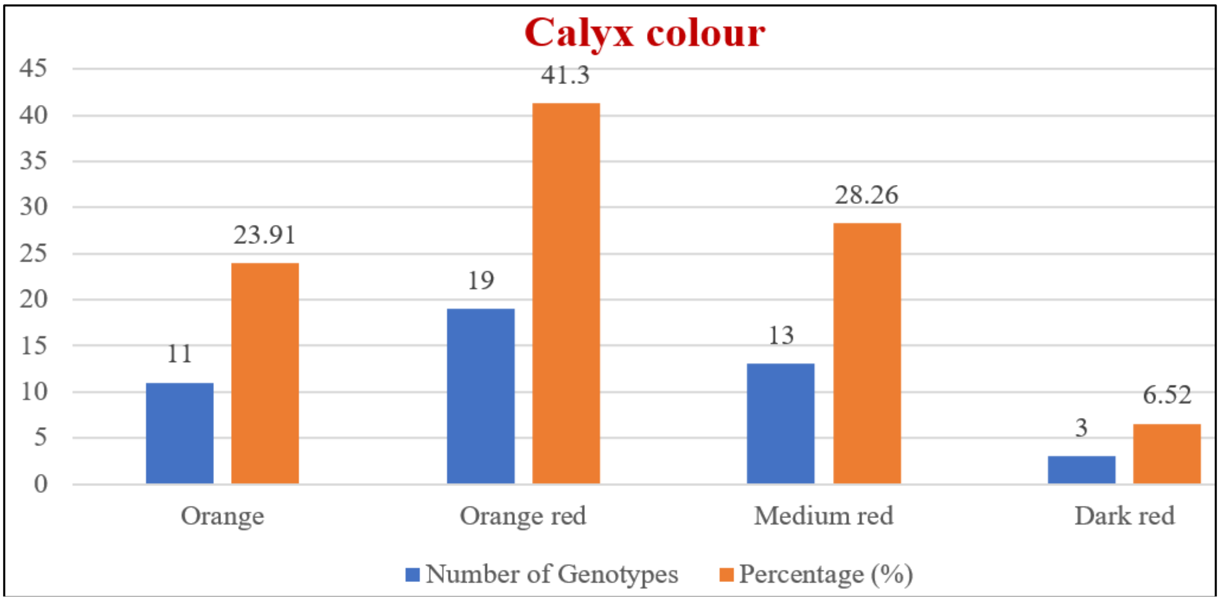

Fig 1. Number of genotypes and frequency percent of calyx colour of pomegranate (*Punica granatum* L.) genotypes of Jammu region

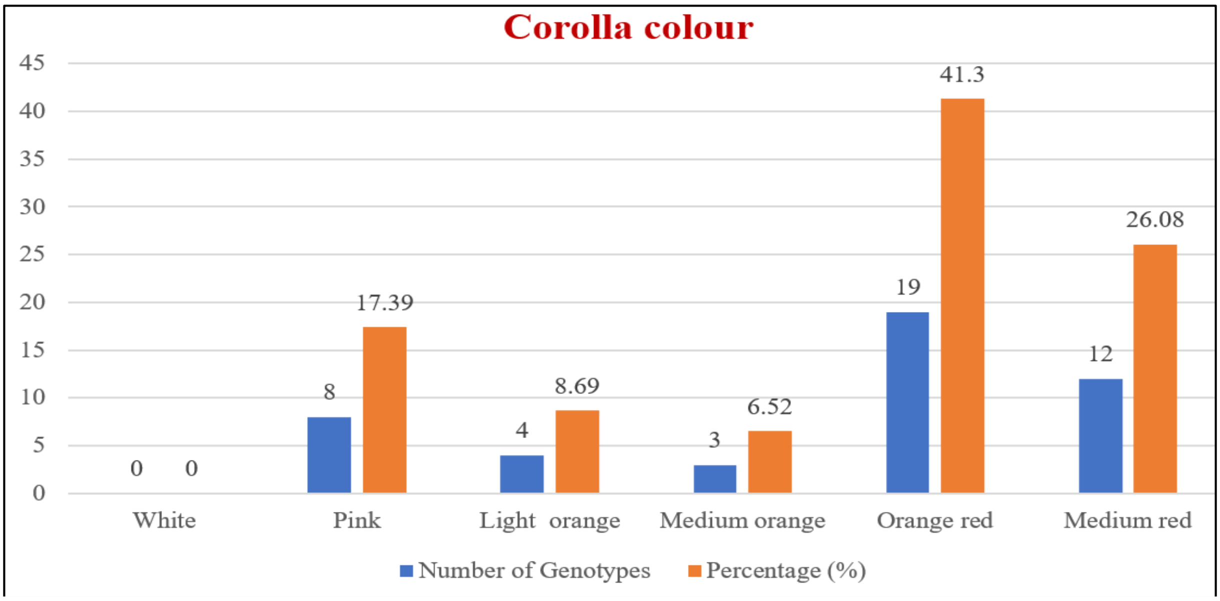

Fig 2. Number of genotypes and frequency percent of corolla colour of pomegranate (*Punica granatum* L.) genotypes of Jammu region

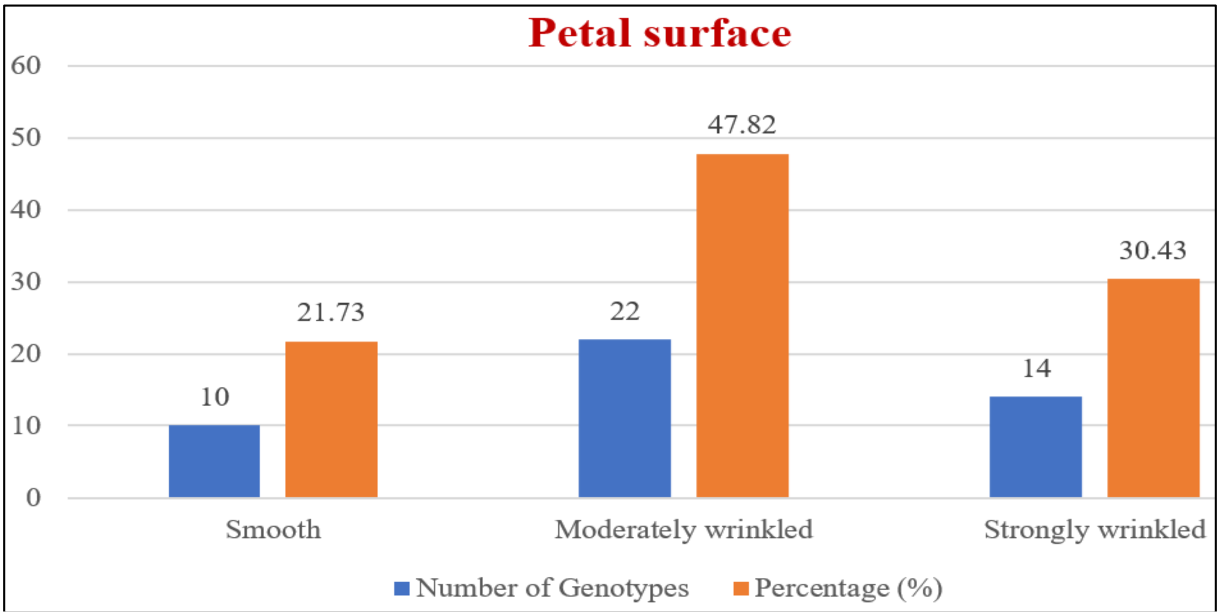

Fig 3. Number of genotypes and frequency percent of petal surface of pomegranate (*Punica granatum* L.) genotypes of Jammu region

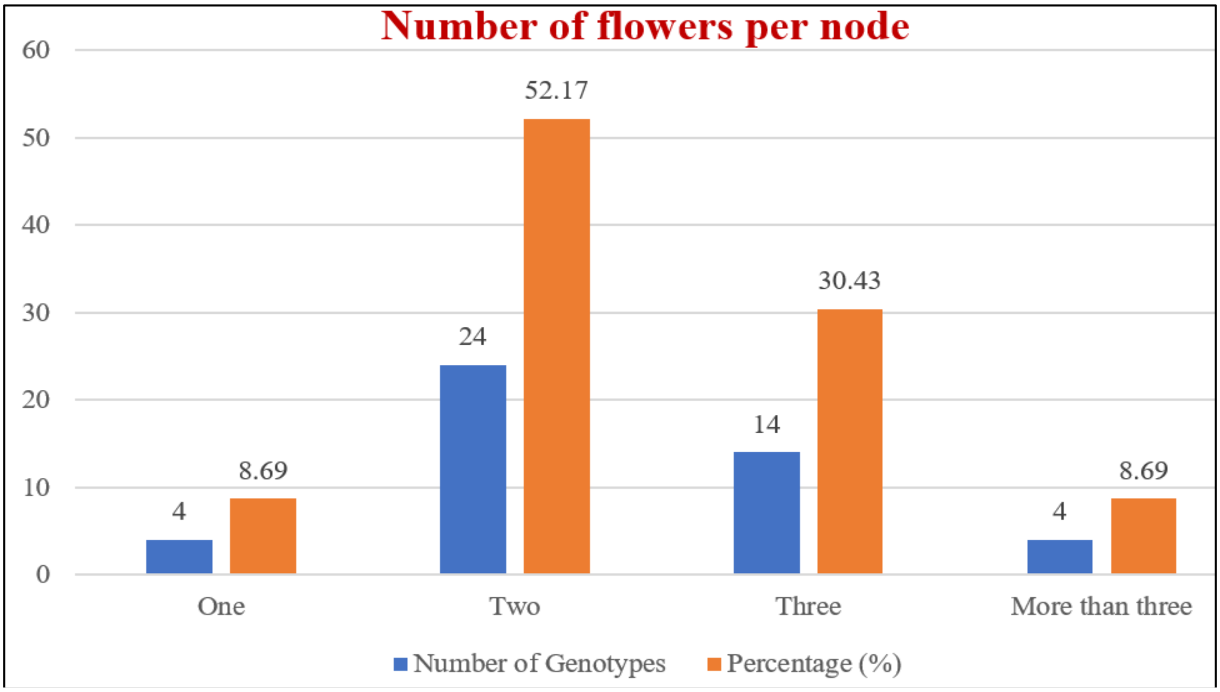

Fig 4. Number of genotypes and frequency percent of number of flowers per node of pomegranate (*Punica granatum* L.) genotypes of Jammu region

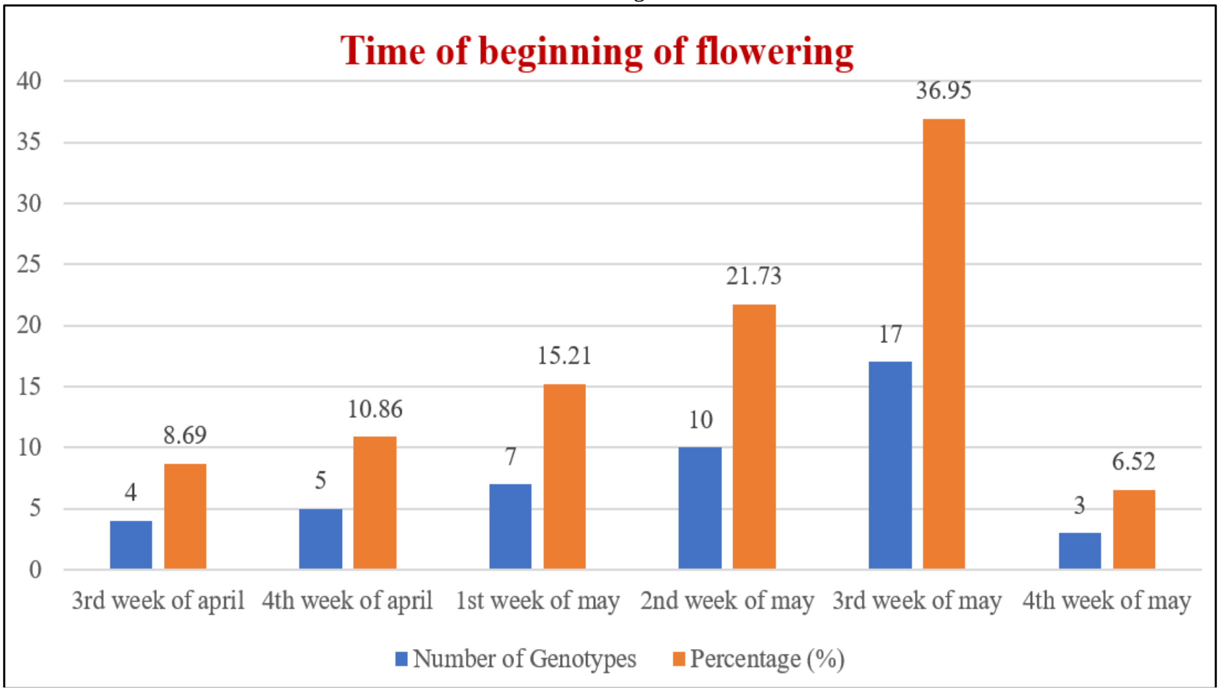

Fig 5. Number of genotypes and frequency percent of time of beginning of flowering of pomegranate (*Punica granatum* L.) genotypes of Jammu region

Conclusion

Wide genetic variability was found in morphological traits among forty-six seedling origin pomegranate genotypes explored under this study. Based on morphological characterization of pomegranate (*Punica granatum* L.) revealing that JMU-Pom (Sel-2) have been found better than other accessions. This exploration and spotting of variability in pomegranate will be helpful in screening and identification of better pomegranate strains for successful crop improvement programme.

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