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MORPHO-PHYSIOLOGICAL ANALYSIS OF LOW CHILLING APPLE VARIETIES IN ASSAM INDIA

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ABSTRACT

The present experiment was conducted with 3 low chilling apple varieties viz., HRMN-99(V1), Dorsett Golden(V2) and Anna(V3) at the Experimental Farm, Department of Horticulture, Assam Agricultural University, Jorhat during 2023-24. The experiment was laid out in Randomized Block Design (RBD) with seven replications. The data on tree and leaf morphological parameters revealed a statistically significant variation among the varieties except for no. for primary branches/tree and petiole length and length of leaf tip respectively. In flower morphological parameters, the data on petal width and size of king bloom was found to be non-significant while the highest no. of flowers/inflorescence (5.74) and flowering to fruiting duration (23.74 days) were recorded in V3 while V2 had the highest flowering duration of 37.42 days. For fruit morphological parameters, the data varied significantly with the highest fruit weight (95.15g) and no. of fruits/tree (42.43) were recorded in V3 while V2 had the highest no. of seeds/fruit (2.73). The physiological parameters also varied significantly with the highest values recorded in V3. Thus, from the study, V3(Anna) was found to be superior due to maximum no. of fruits. / tree (42.43), fruit weight (95.15g), highest RLWC (53.06%) and MSI (89.91%) values as compared to the other two varieties.

***Keywords*:** low chilling apple varieties, morphological, physiological, HRMN-99, Dorsett Golden, Anna

Introduction

Apple (*Malus domestica*) is one of the most important temperate fruit crops in the world and popularly known as the king of temperate fruits. It is grown in high latitude regions of the world where temperatures may reach up to -40°C at high elevations (Janick, 1974). With a world production estimated to be 68.7 million tons in 2018, apples rank third worldwide among the fresh fruits traded (Iriarte *et al.*, 2019). Genetic diversity in crop species can be determined using morphological and agronomic characteristics, as well as biochemical and DNA marker analysis (Liu, 1997). In India, the cultivation of apple is largely done in Jammu & Kashmir, Himachal Pradesh and Uttarakhand. Presently, there are more than 7,500 known cultivars of apples, with a wide range of fruit characteristics and climatic suitability. Apple cultivars are generally propagated by grafting onto favorable rootstocks, which determine the size of the tree. Generally, for most apple varieties the chilling hours required are 1000-1500 hr below 7° C. After the bud break, during the growth, long day hours with high light intensity, warm days (not hot days) viz., 12-15°C and cool nights (not freezing nights) viz., 7– 8°C are favourable for production of quality fruits. However, with the ongoing research, various low chilling varieties of apple have been developed that are suitable for growing in the tropical and sub-tropical climatic condition of the world. Low chilling varieties are those apple varieties which can be successfully cultivated in warm areas with around 200-300 hrs of chilling temperature (below 7°C). Some of the Low Chilling Apple varieties are Anna, Tropical Beauty, Early Fuji, Tamma, Neomi, Parlin's Beauty, Golden Dorsett, HRMN-99 etc (Carter, 2007; Rai *et al.,* 2015).

Morphological characterization of trees and fruits is the first and the most important step for the description, classification and characterization of germplasm collections (Verma *et al*., 2006). It is therefore important to characterize cultivars so that well known cultivars can be clearly distinguished. Being a new temperate crop to be cultivated in the sub-tropical conditions of Assam, it is important to study the morpho-physiological characters of low chilling apple varieties in our soil and climatic condition to seek the answers of possible questions that would arise regarding its successful cultivation in the state.

Materials and Methods

The present study was conducted at the Experimental Farm, Department of Horticulture, Assam Agricultural University, Jorhat (located at 26˚45' N latitude, 94˚12' E longitude and at an altitude of 87 m above mean sea level) during the period 2023-24. The experiment was laid out in a randomized block design (RBD) with three low-chilling apple varieties, viz. V1 (HRMN-99), V2 (Dorsett Golden) and V3 (Anna) and seven replications. The plants were planted on 10th May, 2022 in pits of size 1m x 1m x 1m with a spacing of 3m x 3m covering an experimental area of 540 m^2. The fertilizer dose includes 5kg FYM + 150 g urea+ 210g SSP+ 120 g MOP/ plant for Assam condition.

Quantitative parameters such as tree height (cm), tree girth (cm) and crown spread (cm) were recorded at an interval of 30 days for 3 consecutive months i.e, April, May and June. The crown diameter was recorded by measuring the length of North-south spreading crown branch and again the crown spread at East -West directions and the average value of both crown spread was taken as crown diameter and expressed in centimeter(cm) using the formula: Crown diameter=(W1+W2)/2, where; W1= crown spread at NS, W2= crown spread at EW. Leaf length (cm), leaf width (cm), petiole length (cm), internode length (cm), leaf blade ratio, leaf area and RLWC (%) (Relative Leaf Water Content) were recorded at an interval of 60 days for 2 months i.e., April and June. Petal length (cm), petal width (cm), size of king bloom (cm), pedicel length (cm), fruit length (cm), fruit width (cm), stalk length (cm), seed length (cm) and seed width (cm) and fruit weight (gm) were recorded following the standard procedure.

Qualitative characters were recorded visually during the respective growth stages and some parameters such as tree vigour, tree habit, type of bearing, shoot pubescence, suckering tendency, leaf Blade: attitude in relation to shoot, leaf blade colour (intensity of green), leaf margin, pubescence on lower leaf surface, petal arrangement, pubescence on pedicel, fruit shape, fruit colour at maturity, fruit: flesh colour at maturity and russet type were recorded following IBPGR Apple Descriptors, 1982 and UPOV guidelines for Apple, 2005. Leaf area (cm^2) was measured with the help of leaf area meter and calculated the average. The total chlorophyll content (mg/g) was estimated according to the method given by Anderson and Boardman (1964) in which the absorbance of the extract from the fresh leaf samples was measured at 645 and 663 nm wavelengths using a spectrophotometer. Leaf membrane stability index (MSI)(%) was determined according to the method described by Sairam (1994). The significance of variance was analysed by computing the respective 'F' values as per the procedures given by Panse and Sukhatma (1978).

Results and Discussion

Tree characters

The data on quantitative tree parameters differed significantly under the influence of the varieties except for no. of primary branches/tree (Table 1 & 2). The maximum tree height of 241.95 cm in April, 245.77 cm in May and 251.80cm in June were recorded in V3(Anna), where in June, V3 was statistically at par with V2 (Dorsett Golden) (250.71cm). The maximum tree girth was recorded in V3(Anna)(25.62cm in April, 25.65cm in May and 25.68cm in June) which was statistically at par with V1(HRMN-99)(25.10cm in April, 25.12cm in May and 25.15cm in June). The highest no. of secondary branches/tree was recorded in V3(Anna) (31.00) which was statistically at par with V2(Dorsett Golden) (27.00). The highest crown spread was recorded in V1(HRMN-99) (116.52 cm, 117.92 cm and 119.45 cm in April, May and June respectively) while lowest in V2 (Dorsett Golden) (105.65 cm, 107.18 cm and 108.45 cm in April, May and June respectively) and both the varieties were statistically at par with V3(Anna)(108.10 cm in April,110.92 cm in May and 111.62 cm in June). Similar findings for tree height, tree girth and crown spread were reported by Kumari (2024) in Anna (271-332cm, 14.86-21.38cm and 117-148cm respectively), HRMN-99 (231-291cm, 15.62-22.99cm and 107-137cm respectively) and Dorsett Golden (185-228cm, 10.91-14.83cm and 191-197cm). As per Shyamali (2006) and Chavan *et al*. (2007), these variations in tree characters is might be due to the different genetic makeup of the cultivars under study and also might be the influence of prevailing climatic condition of the region, effect of soil and different management practices. The data on qualitative tree parameters are presented in Table 3.

Table 1 : Tree parameters

Variety	Tree height(cm)			Tree girth(cm)		
	April	May	June	April	May	June
V1 (HRMN-99)	235.28	239.75	246.68	25.10	25.12	25.15
V2 (Dorsett Golden)	238.88	243.50	250.71	22.47	22.51	22.52
V3 (Anna)	241.05	245.77	251.80	25.62	25.65	25.68
SED (±)	0.84	0.84	0.53	1.18	1.18	1.18
CD (P=0.05)	1.86	1.85	1.18	2.60	2.60	2.59

Table 2 : Tree parameters

Variety	No. of primary branches/tree	No. of secondary branches/tree	Crown spread(cm)		
			April	May	June
V1(HRMN-99)	4.42	19.28	116.52	117.92	119.45
V2 (Dorsett Golden)	4.57	27.00	105.65	107.18	108.45
V3 (Anna)	3.85	31.00	108.10	110.25	111.62
SED (±)	0.69	2.75	4.02	3.96	3.86
CD (P=0.05)	NS	6.06	8.86	8.72	8.51

Table 3 : Qualitative tree parameters

Variety	Tree vigour	Tree habit	Type of bearing	Shoot pubescence	Suckering Tendency
V1 (HRMN-99)	Medium	Upright	On spurs only	High	Low
V2 (Dorsett Golden)	Medium	Upright	On spurs only	High	Low
V3 (Anna)	Medium	Upright	On spurs only	High	Low

Leaf parameters

The data on quantitative leaf parameters differed significantly among the varieties except for petiole length and length of leaf tip (Table 4 & 5). The maximum leaf length in April was recorded in V3(Anna) (12.02cm) while minimum in V2(Dorsett Golden) (11.28cm) and both the varieties were statistically at par with V1(HRMN-99) (11.61cm) and in June, the maximum was recorded in V1(HRMN-99) (13.91cm) which was statistically at par with V3(Anna)(13.90cm). In April, the highest leaf width was recorded in V1(HRMN-99) (5.65cm) which was statistically at par with V2(Dorsett Golden) (5.63cm) and in June, the highest was obtained in V1(HRMN-99) (6.29cm) while lowest in V3(Anna)(6.01cm) and both the varieties were statistically at par with V2(Dorsett Golden) (6.15cm). Similar results for leaf blade length (6.26-12.06 cm) and leaf blade width (2.73-7.23 cm) were reported by Hassan *et al.* (2017) in certain apple accessions. The highest internode length was recorded in V1(HRMN-99) (1.79cm in April and 1.92cm in June) while lowest in V3(Anna) (1.33cm in April and 1.47cm in June) and both the varieties were statistically at par with V2(Dorsett Golden)(1.51cm in April and 1.63cm in June). For leaf blade ratio during both April and June, V1(HRMN-99) (2.05 in April and 2.21 in June) was statistically at par with V2(Dorsett Golden) (1.99 in April and 2.22 in June). As per Rana *et al.* (2015) and Handique *et al.* (2022), these variations in leaf characters with respect to the varieties is might be the influence of their genetic makeup or due to the interaction with the environmental factors. The data on qualitative leaf parameters are presented in Table 6.

Table 4 : Leaf parameters

Variety	Leaf length(cm)		Lead width(cm)		Petiole length(cm)	
	April	June	April	June	April	June
V1 (HRMN-99)	11.61	13.91	5.65	6.29	2.90	3.20
V2 (Dorsett Golden)	11.28	13.71	5.63	6.15	3.01	3.16
V3 (Anna)	12.02	13.90	5.49	6.01	2.93	3.19
SED (±)	0.25	0.05	0.05	0.08	0.11	0.12
CD (P=0.05)	0.56	0.12	0.12	0.18	NS	NS

Table 5 : Leaf parameters

Variety	Internode length(cm)		Length of leaf tip(cm)	Leaf blade ratio: length/ breadth	
	April	June		April	June
V1 (HRMN-99)	1.79	1.92	0.62	2.05	2.21
V2 (Dorsett Golden)	1.51	1.63	0.61	1.99	2.22
V3 (Anna)	1.33	1.47	0.65	2.18	2.30
SED (±)	0.13	0.15	0.11	0.04	0.03
CD (P=0.05)	0.29	0.34	NS	0.10	0.06

Table 6 : Qualitative Leaf parameters

Variety	Leaf blade: attitude in relation to shoot	Leaf attachment	Leaf blade colour: intensity of green	Leaf margin	Leaf shape	Arrangement of leaves	Pubescence on lower leaf surface
V1 (HRMN-99)	Outward	Petiolate	Medium and dark	Serrate	Elliptical	Alternate	Medium
V2 (Dorsett Golden)	Outward	Petiolate	Medium and dark	Crenate (40%) and Serrate	Elliptical	Alternate	Medium
V3 (Anna)	Outward	Petiolate	Medium and dark	Serrate	Elliptical	Alternate	Medium

Flower parameters

The data on quantitative flower parameters differed significantly except for petal width and size of king bloom (Table 7). The longest petal and pedicel length were recorded in V1(HRMN-99) (2.04 cm and 1.96 cm respectively) followed by V2(Dorsett Golden) (1.72 cm and 1.77 cm respectively) which was statistically at par with V3(Anna) (1.69 cm and 1.73 cm respectively). The highest no. of flowers/ inflorescence was recorded in V3(Anna) (5.74), which was statistically at par with V2(Dorsett Golden) (5.62). The longest flowering duration was recorded in V2(Dorsett Golden) (37.42 days), which was statistically at par with V3(Anna) (34.10 days). The longest flowering to fruiting duration was recorded in V3(Anna) (23.74 days) while the shortest in V1(HRMN-99) (18.90 days) and both the varieties were statistically at par with V2(Dorsett Golden) (21.98 days). Similar results for no. of flowers/ inflorescence (4-6) and pedicel length (1.72-1.98cm) were reported by Devi (2021) in different Ambri genotypes. As per Singh *et al*. (2005), these variations in flower morphological parameters and also in the time and duration of flowering may be attributed to genetic make-up of the cultivars and also might be the influence of the prevailing climatic conditions of the experimental site. The data on qualitative flower parameters are presented in Table 8(a) and 8(b).

Table 7 : Flower parameters

Variety	Petal length (cm)	Petal width (cm)	Pedicel length (cm)	No. of flowers/ inflorescence	Size of king bloom (cm)	Flowering duration (days)	Flowering to fruiting duration (days)
V1 (HRMN-99)	2.04	1.13	1.96	5.30	3.50	29.84	18.90
V2 (Dorsett Golden)	1.72	1.22	1.77	5.62	3.45	37.42	21.98
V3 (Anna)	1.69	1.21	1.73	5.74	3.80	34.10	23.74
SED (±)	0.11	0.06	0.08	0.13	0.22	2.07	1.74
CD (P=0.05)	0.24	NS	0.18	0.30	NS	4.56	3.83

Table 8(a) : Qualitative flower parameters

Variety	Type of inflorescence	Pubescence on pedicel	Position of inflorescence	Nature of inflorescence
V1 (HRMN-99)	Cymose	Medium	Both terminal and axillary	Cluster
V2(Dorsett Golden)	Cymose	Medium	Both terminal and axillary	Cluster
V3 (Anna)	Cymose	Medium	Both terminal and axillary	Cluster

Table 8(b) : Qualitative flower parameters

Variety	Time of flowering	Type of flower	No. of petals per flower	No. of sepals per flower	Flower colour	Petal arrangement
V1 (HRMN-99)	Mid (2nd March, 2024)	Hermaphrodite	Five	Five	Pale pink	Free (30%) and intermediate
V2 (Dorsett Golden)	Mid (2nd March, 2024)	Hermaphrodite	Five	Five	Pale pink	Free (40%) and intermediate
V3 (Anna)	Early (25th February, 2024)	Hermaphrodite	Five	Five	Pale pink	Free (25%) and intermediate

Fruit parameters

The data on quantitative fruit parameters differed significantly among the varieties (Table 9). The highest fruit weight was recorded in V3(Anna) (95.15 g), which was statistically at par with V2(Dorsett Golden) (91.02 g). Similar result for fruit weight was reported by Bodh *et al.* (2020) in Nishiki (85.93 g) to Early Amber (100.33 g) peach accessions. The variation in fruit weight can be attributed to environmental conditions, management practices and genotypic variations among the varieties as producing bigger fruits might be the inherent ability of a genotype by utilizing resources efficiently to achieve a certain fruit size (Basak *et al*., 2022). The highest fruit length was recorded in V2(Dorsett Golden) (5.96 cm) while the lowest in V1(HRMN-99)(5.44 cm) and both the varieties were statistically at par with V3(Anna)(5.78 cm). The highest breadth was noted in V3(Anna)(5.36 cm) while the lowest in V1(HRMN-99)(4.77cm) and both the varieties were statistically at par with V2(Dorsett Golden)(5.05cm). The longest stalk length was observed in V1(HRMN-99)(2.06 cm) which was statistically at par with V2(Dorsett Golden)(1.80 cm). Kumar *et al*. (2023) reported similar results for fruit length and breadth in HRMN-99 (5.58 cm and 5.04 cm respectively), Dorsett Golden (5.52 cm and 5.02 cm respectively) and Anna (5.71 cm and 5.26 cm respectively) and similar stalk length (1.9-2.25cm) by Baba (2023) in Ambri apple genotypes. The variations in fruit length, width and stalk length among the varieties are probably due to differences in genetic makeup and micro-climate conditions (Kayesh *et al*., 2018). The maximum volume was recorded in V3(Anna)(84.72 cc) followed by V2(Dorsett Golden)(74.12 cc), which was statistically at par with V1(HRMN-99) (71.37 cc). The variation in fruit volume with respect to different varieties is mainly attributed to the inter-varietal differences associated with genetic make-up of the varieties (Kishor *et al*.,2017) and governed mainly by fruit weight as bigger size fruits will have more volume as compared to a smaller one. The data on qualitative fruit parameters are presented in Table 10.

Table 9 : Fruit parameters

Variety	Fruit wt (g)	Length (cm)	Breadth (cm)	Stalk length (cm)	Volume (cc)	No. of fruits / tree
V1 (HRMN-99)	84.26	5.44	4.77	2.06	71.37	32.83
V2 (Dorsett Golden)	91.02	5.96	5.05	1.80	74.12	41.32
V3 (Anna)	95.15	5.78	5.36	1.81	84.72	42.43
SED (±)	2.15	0.17	0.16	0.10	2.27	1.25
CD (P=0.05)	4.74	0.38	0.35	0.23	5.00	2.75

Table 10 : Qualitative fruit parameters

Variety	Fruit shape	Fruit colour at maturity		Fruit flesh colour	Russet Type
		Ground colour	Over colour		
V1 (HRMN-99)	Oblong, globose and ellipsoid	Yellow	Red	Cream	Very fine
V2 (Dorsett Golden)	Oblong, globose and ellipsoid	Yellow	Red	Cream	Very fine
V3 (Anna)	Oblong, globose and ellipsoid	Yellow	Red	Cream	Very fine

Seed parameters

The data on quantitative seed parameters varied significantly (Table 11). The maximum no. of seeds/fruit were recorded in V2(Dorsett Golden) (2.73), which was statistically at par with V3(Anna)(2.67). Similar result was recorded by Hassan *et al*. (2017), who observed 2.32, 2.51 and 2.52 no. of seeds/fruit in some apple accessions of Kashmir. The varied data for no. of seeds is probably due to different pulp content of the fruits belonging to different varieties (Meghana *et al*., 2020). The longest seed length was recorded in V1(HRMN-99) (0.89cm) followed by V2(Dorsett Golden) (0.79cm), which was statistically at par with V3(Anna) (0.78) while the maximum seed breadth was recorded in V3(Anna)(0.30cm). The results are in accordance with the findings of Handique *et al*. (2022), who reported similar seed length (0.80cm and 0.97cm) and seed width (0.23cm and 0.33cm) in two different types of custard apple. The variation in seed length and width is probably due to different genetic makeup of the varieties or it might be due to variation in fruit size (Handique *et al*., 2022).

Table 11 : Seed parameters

Variety	No. of seeds/fruit	Seed length(cm)	Seed breadth(cm)	Presence of seed	Colour of seed	Seed shape
V1 (HRMN-99)	2.40	0.89	0.24	Present	Brown	Ovoid
V2 (Dorsett Golden)	2.73	0.79	0.27	Present	Brown	Ovoid
V3 (Anna)	2.67	0.78	0.30	Present	Brown	Ovoid
SED (±)	0.10	0.02	0.01			
CD (P=0.05)	0.23	0.05	0.03			

Physiological parameters

The data on physiological parameters varied significantly among the varieties (Table 12). The highest leaf area was recorded in V3(Anna) (40.59cm^2 in April and 42.57cm^2 in June), which was statistically at par with V2(Dorsett Golden) (39.95cm^2 in April and 41.83cm^2 in June). The results are in accordance with the findings of Kumari (2024), who reported similar leaf area in HRMN-99(39.32-40.37cm^2), Dorsett Golden (40.35-41.34cm^2) and Anna (40.75-42.84cm^2). The variation is probably due to varied leaf morphology of the varieties, which is influenced by its individual genetic makeup. In April, the maximum RLWC was recorded in V3(Anna)(47.71%) while minimum in V1(HRMN-99)(45.05%) and both the varieties were statistically at par with V2(Dorsett Golden)(45.49%) and in June, the maximum was recorded in V3(Anna)(58.42%) while minimum in V2(Dorsett Golden)(54.97%), which was statistically at par with V1(HRMN-99)(54.99%). Similar results were reported by Sharma (2024) in HRMN-99, Dorsett Golden and Anna. The probable reason for this variation is due to the genotypic differences among the varieties, favouring better maintenance of internal water relations of the leaf tissues (Chavan *et al*., 2007). The highest total chlorophyll content was recorded in V3(Anna)(1.11 mg/g) while lowest in V1(HRMN-99)(0.78 mg/g) and both the varieties were statistically at par with V2(Dorsett Golden)(0.95 mg/g). This variation in the chlorophyll content may be attributed to the genotypic difference in the photosynthetic activity with respect to the cultivars. Similar findings by Mihaljevic *et al*. (2021) in apple and Sharma (2024) in HRMN-99, Dorsett Golden and Anna presented results on varietal differences in leaf chlorophyll content. The highest MSI (Membrane Stability Index) was recorded in V3(Anna)(89.91%), which was statistically at par with V2 (Dorsett Golden) (89.76%). Similar variation was recorded by Singh *et al*. (2020) in different India wheat cultivars, which is probably due to the varied stress tolerant ability of the varieties to cope up with stress condition as per the prevailing climatic condition of the site.

Table 12 : Physiological Parameters

Variety	Leaf area(cm^2)		RLWC (%)		Total chlorophyll content (mg/g)	Membrane Stability Index (%)
	April	**June**	**April**	**June**		
V1 (HRMN-99)	37.13	40.09	45.05	54.99	0.78	84.70
V2 (Dorsett Golden)	39.95	41.83	45.49	54.97	0.95	89.76
V3 (Anna)	40.59	42.57	47.71	58.42	1.11	89.91
SED (±)	0.89	0.59	0.99	1.06	0.09	0.21
CD (P=0.05)	1.97	1.30	2.19	2.34	0.21	0.46

Conclusion

The study on the morpho-physiological parameters of these low chilling apple varieties were essential to evaluate its morphology and performance as the per the prevailing climatic condition of the experimental site, since it's a new and temperate crop planted in our sub-tropical climatic condition. From the present study, it could be concluded that the sub-tropical climatic condition of Assam would offer opportunities for cultivation of low chilling apple varieties. Successful growth and development of the varieties under study were achieved which was marked by adequate flowering and fruit set as per the age of the plants so far. In terms of no. of fruits/tree, fruit weight and membrane stability index(MSI), V3(Anna) was found to be better than the other two varieties.

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