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Effect of intercroping geometry inorganic-based cropping models of broccoli (*Brassica oleracea* var. italica)

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ABSTRACT

A field experiment was conducted to evaluate the efficacy of intercropping and crop geometry in organic production of broccoli (*Brassica oleracea* var. italica) at CCS Haryana Agricultural University, Hisar, during winter season of 2012-13. The experiment was laid out in a split plot design, replicated thrice with three different organic manures and two spacing as main plot treatments and five intercrops including sole crop of broccoli as sub-plot treatments, thus making a total of 30 treatment combinations. Of the 30 treatment combinations, application of vermicompost coupled with single row spacing and sole crop of broccoli ($M_2S_1C_1$) recordered maximum values of number of leaves (31.6), weight of main head (206.6), girth of head (16.5 cm), number of sprouts/plant (10.8), yield of sprouts/plant (413.4 g), yield of sprouts (main head + sprouts)/plant (620.0 g) and total yield (306.2 q/ha) significantly,while plant height (68.2 cm) was highest in treatment $M_2S_2C_1$ and yield of intercrops (146.5 q/ha) was recorded maximum in $M_2S_2C_5$ treatment. Amongst intercropping treatments, broccoli + fenugreek along with vermicompost application following the spacing of 45 cm × 45 cm ($M_2S_1C_4$) recorded highest growth and yield-attributing characters except number of sprouts/plant which was found significantly better in the treatment ($M_2S_1C_3$).

Key Words: Broccoli, Crop geometry, Cropping models, Organic manures, Sole crop

Sprouting broccoli (*Brassica oleracea* var. *italica* L.), a member of cole group, is considered as a minor vegetable crop. However, its cultivation is now gaining popularity with Indian growers for the last couple of years due to increasing awareness of its high nutritive values and tourist influx. To stabilize crop production and to provide insurance mechanism against aberrant weather situation characterizing rainfed agriculture, intercropping could be a viable cultural means of risk minimizing farmers profit and subsistence-oriented, energy-efficient and sustainable venture. Our strategy should be to produce more vegetables from less land, less water with less detrimental to soil and environment as well. Growing of two or more different crops on the same piece of land during the same crop season interacts agronomically (Faroda *et al.* 2007). It is well established that plant spacing has significant effect on growth and yield of broccoli. Optimal plant spacing is important for crop production through efficient utilization of nutrients, water and light by plants. In general, higher plant population adversely affects the yield per unit area hampering vegetative and reproductive growth of plant. Paired row planting (30/60 cm × 45 cm) may facilitate the growing of intercrops like fenugreek, coriander, beet leaf and radish in broccoli because the space available between rows of main crop is more than that available in normal row spacing, *i.e.* 45 cm × 45 cm (Singh, 1992). In India, pure organic farming is possible partially only for the crops having high export potential in international markets. Organic vegetable cultivation offers one of the most sustainable farming systems with recurring benefits to only long-term soil health but provides a lasting stability in production by importing better resistance against various biotic and abiotic stresses. Further, organic manures can serve as an alternative practice in place of mineral fertilizers for

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improving soil structure, microbial biomass and producing quality crop yield. Therefore, an experiment was conducted to find out the effect of intercropping and crop geometry on organic production of cauliflower.

MATERIALS AND METHODS

The experiment was conducted at the research farm and laboratory of Department of Vegetable Science, CCS Haryana Agricultural University, Hisar, during winter season of 2012-13. Hot and dry winds during summer and dry severe cold in winter are common features of this region. The experiment was laid out in a split plot design replicated thrice with three different organic manure (FYM, vermicompost and poultry manure) and two spacing, *viz.* single row (45 cm × 45 cm) and paired row (30/60 cm × 45 cm) as main plot treatments and five intercrops, *viz.* broccoli (CBH-1), beet leaf (HS-23), coriander (HisarBhumit), fenugreek (Hisar Suwarna) and radish (HS-1), including sole crop of broccoli as sub-plot treatments, thus making a total of 30 treatment combinations.

The FYM, vermicompost and poultry manure were applied at the rate of 8.0, 5.0 and 5.0 tonnes/ha, respectively. The available N, P and K contents (%) in organic manures were in FYM (0.5, 0.3 and 0.3), vermicompost (1.5, 1.2 and 1.1) and poultry manure (1.1, 0.8 and 0.8), respectively. The available N content was determined by alkaline permanganate method (Subhiah and Asija, 1956) while available phosphorus content by Olsen method (Olsen *et al.*, 1954). The available potassium was determined by extraction of manures and with 1 N neutral ammonium acetate and estimated by flame photometer (Metson, 1956).

The seeds of intercrops were sown between the rows of broccoli five of days after transplanting. The intercrops were harvested 25-35 days after sowing. The harvesting of head and sprouts was done 55 days after transplanting and onwards when heads and buds are compact and unopened. The study on intercropping, crop geometry, organic manure and their interactions on vegetative growth and yield parameters of broccoli were studied. Mean values of the parameters in each replication were statistically analyzed in split plot design as suggested by Panes and Sukhatme (1985) and by using software of CCS HAU, Hisar_website http://hau.ernet.in/opstat.html for analysis of variance and test of significance.

RESULTS AND DISCUSSION

The individual effect of treatmentslike organic manures, crop geometry and intercropping of different intercrops in between rows of broccoli showed significant effects on growth and yield of broccoli (Table 1). The maximum plant height (57.52 cm), number

Table 1. Effect of organic manures, crop geometry and intercrops on growth and yield parameters of broccoli and total yield of intercrops

Treatment	PH	NL	WMH	GMH	NS	YS	TYPP	TYPH	TYI
Manures (M)									
FYM (M_1)	54.19	23.1	163.0	11.7	5.1	325.3	488.2	241.1	47.8
Vermicompost (M_2)	57.52	26.0	188.2	13.4	7.0	376.4	564.6	278.8	58.1
Poultry manure (M_3)	55.49	24.4	178.2	13.2	6.2	357.0	534.6	264.0	51.5
CD (5%)	0.40	0.2	4.4	0.3	0.2	8.3	13.3	6.6	1.3
Spacing (S)									
45 cm × 45 cm (S_1)	53.57	25.0	180.2	13.2	6.5	360.7	540.5	266.9	40.7
30/60 cm × 45 cm (S_2)	57.90	24.0	172.7	12.3	5.7	345.1	517.7	255.7	64.2
CD (5%)	0.33	0.2	3.6	0.2	0.1	6.8	10.8	5.4	1.0
Intercrops (C)									
Broccoli (C_1)	64.23	27.7	187.7	15.0	8.8	375.3	561.9	277.5	0.0
Beet leaf (C_2)	55.69	23.2	176.5	11.6	5.2	353.5	530.2	261.8	61.5
Coriander (C_3)	54.69	25.0	177.6	13.0	6.3	355.2	532.8	263.1	24.5
Fenugreek (C_4)	55.52	25.4	179.4	13.0	6.3	358.2	538.2	265.8	57.9
Radish (C_5)	48.53	21.1	160.8	11.2	3.8	321.7	482.5	238.3	118.5
Mean (M, S, C)	55.73	24.5	176.4	12.8	6.1	352.9	529.1	261.1	52.5
CD (5%)	0.14	0.3	0.1	0.3	0.1	1.7	1.0	0.5	1.4

PH, plant height (cm); NL, number of leaves; WMH, weight of main head (g); GMH, girth of main head (cm); NS, number of sprouts; YS, yield of sprouts (g/plant); TYPP, total yield/plant (g); TYPH, total yield per hectare (q) and TYI, total yield of intercrops (q/ha)

Table 2. Interaction effect of organic manures, crop geometry and intercrops on plant height, number of leaves and weight of main head

Intercrop grown with broccoli	Plant height (cm)											
	M_1			M_2			M_3			Mean for plant spacing		Mean for organic manures
	S_1	S_2	Mean	S_1	S_2	Mean	S_1	S_2	Mean	S_1	S_2	
C_1	60.60	64.90	62.75	63.10	68.23	65.67	61.97	66.57	64.27	61.89	66.57	64.23
C_2	52.03	56.03	54.03	54.83	60.40	57.62	53.63	57.20	55.42	53.50	57.88	55.69
C_3	51.00	55.27	53.14	54.13	58.50	56.32	52.67	56.60	54.64	52.60	56.79	54.70
C_4	51.83	55.80	53.82	53.43	62.20	57.82	51.87	58.00	54.94	52.38	58.67	55.52
C_5	46.03	48.37	47.20	49.20	51.20	50.20	47.20	49.20	48.20	47.48	49.59	48.53
Mean	52.30	56.07	54.19	54.94	60.11	57.52	53.47	57.51	55.49	53.57	57.90	55.73
CD (5%) for interaction : manures × spacing = 0.57, manures × intercrops = 0.24, spacing × intercrops = 0.20, manures × spacing × intercrops = 0.34												
Intercrop	Number of leaves											
C_1	25.6	25.1	25.4	31.6	28.0	29.8	29.2	26.8	28.0	28.8	26.6	27.7
C_2	22.8	22.4	22.6	25.5	24.3	24.9	22.4	21.8	22.1	23.6	22.8	23.2
C_3	22.4	23.9	23.2	26.3	26.6	26.5	25.6	25.2	25.4	24.9	25.2	25.0
C_4	23.9	25.0	24.5	27.4	27.1	27.3	24.6	24.6	24.6	25.7	25.6	25.4
C_5	21.4	18.8	20.1	23.0	19.7	21.4	23.8	20.1	22.0	21.2	19.5	21.1
Mean	23.2	23.0	23.1	26.8	25.1	26.0	25.1	23.7	24.4	24.6	24.0	24.5
CD (5%) for interaction : manures × spacing = 0.34, manures × intercrops = 0.51, spacing × intercrops = 0.42, manures × spacing × intercrops = 0.7												
Intercrop	Weight of main head (g)											
C_1	177.0	172.0	174.5	206.6	197.0	201.8	190.2	183.2	186.7	191.3	184.1	187.7
C_2	163.7	157.6	160.7	191.3	188.1	189.7	186.4	173.3	179.9	180.5	173.0	176.7
C_3	165.0	159.3	162.2	192.3	187.4	189.9	185.2	176.2	180.7	180.8	174.3	177.6
C_4	167.3	161.7	164.5	195.6	188.3	192.0	187.5	175.8	181.7	183.5	175.3	179.4
C_5	157.0	149.0	153.0	172.3	162.6	167.5	165.7	158.4	162.1	165.0	156.7	160.8
Mean	166.0	159.9	163.0	191.6	184.7	188.2	183.0	173.4	178.2	180.2	172.7	176.4
CD (5%) for interaction : manures × spacing = NS, manures × intercrops = 0.1, spacing × intercrops = 0.1, manures × spacing × intercrops = 0.1												

Table 3. Interaction effect of organic manures, crop geometry and intercrops on girth of head, number of sprouts per plant and yield of sprouts per plant.

Intercrop grown with broccoli	Girth of head (cm)											
	M_1			M_2			M_3			Mean for plant spacing		Mean for organic manures
	S_1	S_2	Mean	S_1	S_2	Mean	S_1	S_2	Mean	S_1	S_2	
C_1	14.2	13.4	13.8	16.5	15.6	16.1	15.6	14.9	15.3	15.4	14.6	15.0
C_2	11.4	9.9	10.7	12.8	11.6	12.2	12.1	12.0	12.1	12.1	11.2	11.6
C_3	12.5	10.6	11.6	14.0	13.1	13.6	14.4	13.2	13.8	13.6	12.3	13.0
C_4	12.7	10.7	11.7	14.4	13.5	14.0	13.2	13.6	13.4	13.4	12.6	13.0
C_5	11.3	9.8	10.6	11.4	11.5	11.5	12.1	11.3	11.7	11.6	10.9	11.2
Mean	12.4	10.9	11.7	13.8	13.1	13.4	13.5	13.0	13.2	13.2	12.3	12.8
CD (5%) for interaction : manures × spacing = 0.4, manures × intercrops = 0.5, spacing × intercrops = NS, manures × spacing × intercrops = 0.7												
Intercrop	Number of sprouts per plant											
C_1	7.0	6.5	6.8	10.8	9.6	10.2	10.0	9.1	9.6	9.3	8.4	8.8
C_2	4.9	4.5	4.7	6.3	5.5	5.9	5.3	4.8	5.1	5.5	4.9	5.2
C_3	6.1	5.3	5.7	7.5	6.7	7.1	7.1	5.2	6.2	6.9	5.7	6.3
C_4	6.1	5.5	5.8	7.3	6.9	7.1	6.8	5.5	6.2	6.7	6.0	6.4
C_5	2.8	2.4	2.6	5.1	4.3	4.7	4.6	3.8	4.2	4.2	3.5	3.8
Mean	5.4	4.8	5.1	7.4	6.6	7.0	6.8	5.7	6.2	6.5	5.7	6.1
CD (5%) for interaction : manures × spacing = 0.2, manures × intercrops = 0.3, spacing × intercrops = 0.2, manures × spacing × intercrops = 0.4												
Intercrop	Yield of sprouts per plant (g)											
C_1	351.7	339.7	345.7	413.4	394.1	403.8	386.9	366.4	376.7	384.0	366.7	375.4
C_2	327.4	315.2	321.3	382.7	376.3	379.5	372.8	346.6	359.7	361.0	346.0	353.5
C_3	330.0	318.7	324.4	384.7	374.9	379.8	370.4	352.4	361.4	361.7	348.7	355.2
C_4	334.7	323.3	329.0	391.4	376.7	384.1	375.1	351.6	363.4	367.1	350.5	358.8
C_5	314.0	298.0	306.0	344.7	325.3	335.0	331.4	316.8	324.1	330.0	313.4	321.7
Mean	331.6	319.0	325.3	383.4	369.5	376.4	367.3	346.8	357.0	360.8	345.1	352.9
CD (5%) for interaction : manures × spacing = NS, manures × intercrops = 3.0, spacing × intercrops = 2.4, manures × spacing × intercrops = 4.2												

Table 4. Interaction effect of organic manures, crop geometry and intercrops on total yield per plant, total yield per hactare and total yield of intercrops

Intercrop grown with broccoli	Total yield per plant (g)											
	M_1			M_2			M_3			Mean for plant spacing		Mean for organic manures
	S_1	S_2	Mean	S_1	S_2	Mean	S_1	S_2	Mean	S_1	S_2	
C_1	528.7	511.7	520.2	620.0	591.1	605.6	570.6	549.6	560.1	573.1	550.8	562.0
C_2	491.1	472.8	482.0	574.0	564.4	569.2	559.2	519.9	539.6	541.4	519.0	530.2
C_3	495.0	478.0	486.5	577.0	562.3	569.7	555.6	528.6	542.1	542.5	523.0	532.8
C_4	502.0	485.0	493.5	587.0	565.0	576.0	562.6	527.4	545.0	550.5	525.8	538.2
C_5	471.0	447.0	459.0	517.0	487.9	502.5	497.1	475.2	486.2	495.0	470.0	482.5
Mean	497.6	478.9	488.2	575.0	554.1	564.6	549.0	520.1	534.6	540.5	517.7	529.1
CD (5%) for interaction : manures × spacing = NS, manures × intercrops = 1.5, spacing × intercrops = 1.8, manures × spacing × intercrops = 2.5												
Intercrop	Total yield (q/ha)											
C_1	261.1	252.7	256.9	306.2	291.9	299.1	281.8	271.4	276.6	283.0	272.0	277.5
C_2	242.5	233.5	238.0	283.5	278.7	281.1	276.2	256.7	266.5	267.4	256.3	261.9
C_3	244.4	236.0	240.2	284.9	277.7	281.3	274.4	261.0	267.7	267.9	258.2	263.1
C_4	247.9	239.5	243.7	289.9	279.0	284.5	277.8	260.4	269.1	271.9	259.6	265.8
C_5	232.6	220.7	226.7	255.3	240.9	248.1	245.5	234.7	240.1	244.5	232.1	238.3
Mean	245.7	236.5	241.1	284.0	273.6	278.8	271.1	256.8	264.0	266.9	255.7	261.3
CD (5%) for interaction : manures × spacing = NS, manures × intercrops = 0.9, spacing × intercrops = 0.7, manures × spacing × intercrops = 1.2												
Intercrop	Total yield of intercrops (q/ha)											
C_1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
C_2	38.2	78.2	58.2	48.6	88.9	68.8	37.6	77.4	57.5	41.5	81.5	61.5
C_3	16.2	29.4	22.8	17.0	37.1	27.1	14.5	32.7	23.6	15.9	33.1	24.5
C_4	42.2	61.4	51.8	41.6	84.1	62.9	40.7	77.3	59.0	41.5	74.3	57.9
C_5	94.1	118.5	106.3	116.8	146.5	131.7	103.7	131.3	117.5	104.9	132.1	118.5
Mean	38.1	57.5	47.8	44.8	71.3	58.1	39.3	63.7	51.5	40.7	64.2	52.5
CD (5%) for interaction : manures × spacing = 1.8, manures × intercrops = 2.5, spacing × intercrops = 2.0, manures × spacing × intercrops = 3.5												

of leaves (26.0), weight of main head (188.2 g), girth of main head (13.4 cm), number of sprouts / plant (7.0), yield of sprouts / plant (376.4 g), total yield (main head+sprouts) / plant (564.6 g), total yield / (278.8 q / ha) of broccoli and total yield of intercrops (58.1 q / ha) were found with the application of vermicompost. The application of vermicompost was found more effective due to better aeration, water-holding capacity and might have increased the nutrient-use efficiency, supply of micronutrients and availability of major nutrients due to favourable soil condition, followed by poultry manure and FYM in respect all characters. The results indicated that the treatments receiving high doses of nutrients resulted in hastening of different reproductive growth phases of broccoli, where as commencement of different reproductive phases were drastically delayed in plants receiving low rate of nutrients or no nutrients (Choudhary *et al.*, 2012; Kumar *et al.*, 2013). Similar results were presented in different vegetables.

The crop geometry on broccoli crop had shown a significant effect on growth and yield attributes. The maximum number of leaves, weight of main head, girth of main head, number of sprouts / plant, yield of sprouts / plant, total yield (main head + sprouts) / plant and total yield / ha of broccoli were observed in normal spacing due to less competition for growing space and light. However, maximum plant height and total yield of intercropswas recorded in paired row planting (30 / 60 cm × 45 cm) due to closer spacing that supported erect growth as compared to normal spacing. Singh *et al.* (2004) suggested that closer spacing of broccoli tended to produce taller plant than widely-spaced plant. This might be due to competition of solar energy couple with shallow root system and higher plant height at closer spacing (Table 1).

The result revealed that plant growth and yield significantly influenced. The growing of sole crop of broccoli recorded maximum plant height,number of leaves, weight of main head, girth of main head, number of sprouts / plant, yield of sprouts / plant, total yield / plant and total yield / ha of broccoli. However, amongst the intercropping pattern, maximum plant height of broccoli plants was recorded with growing of beet leaf (C_2) which was statistically at par in the treatment C_4. This was due to fast and vigorous growth of beet leaf that resulted in erect growth of broccoli plants. The maximum girth of main head and number of sprouts was recorded in the treatment C_3 which was statistically at par in the treatment C_4. Whereas, all other growth and yield parameters were recorded with growing of fenugreek that was due to fixing of atmospheric nitrogen and small canopy growth of fenugreek, which was statistically at par in the treatment C_3 and C_2. However, intercropping of radish had adverse effect on growth and yield of broccoli due to intense competition for light, space, nutrients and moisture.

The interaction between organic manure, crop geometry and intercrops remarkably influenced the growth and yield parameters of broccoli and yield of intercrops (Tables 2-4). The treatment $M_2S_1C_1$ was found superior in respect to all growth and yield parameters of broccoli. Application of vermicompost coupled with paired row spacing of 30 / 60 cm × 45 cm having sole broccoli crop recorded higher plant height, followed by $M_3S_2C_1$. However, application of vermicompost with a spacing of 45 cm × 45 cm with sole crop of broccoli recorded maximum number of leaves and weight of main head, maximum girth of head, number of sprouts / plant, yield of sprouts / plant, total yield (main head + sprouts) / plant and total yield / ha.

The intercropping pattern, best results were observed in the treatment $M_2S_1C_4$ except plant height which was superior in the treatment $M_2S_2C_4$. The treatment, broccoli + fenugreek along with vermicompost application following the spacing of 45 cm × 45 cm ($M_2S_1C_4$) recorded highest growth and yield-attributing characters except number of sprouts / plant which was found in treatment ($M_2S_1C_3$). $M_2S_1C_3$ was found statistically at par with the treatment $M_2S_1C_4$. There were non-significant difference in between $M_2S_2C_2$ and $M_3S_2C_3$, $M_2S_2C_2$ and $M_3S_2C_4$ and, $M_3S_2C_3$ and $M_3S_2C_4$ for total yield / ha. Application of organic based nutrients mainly biogas slurry + FYM, vermicompost + FYM and vermicompost alone recorded the maximum fruit size and more number of fruits / plant in tomato (Renuka and Ravishankar, 1998).

These findings clearly indicated that vermicompost played a significant role on enhancing growth of broccoli. Improvement in plant growth attributes with the application of vermicompost might be due to better photosynthesis, energy storage, cell division and cell enlargement, moisture holding capacity, supply of micronutrients and availability of major nutrients due to favourable soil condition (Uddin *et al.*, 2009 and Sinha *et al.*, 2013). Poultry manure also enhanced the vegetative growth of broccoli. It might be due to the fact that poultry manure contains uric acid having 60 percent nitrogen.

The uric acid rapidly changes to ammonia form causing its immediate and efficient utilization for better plant growth and development. These results were supported by Chaterjee *et al.* (2005) and Maurya *et al.* (2008). These results are in partial conformity with the findings of Maurya *et al.* (2008), Choudhary *et al.* (2012), Devi and Singh (2012), Kumar *et al.* (2013) and Mohapatra, *et al.* (2013).thus, it may be concluded that application of vermicompost coupled with paired row

spacing of 30/60 × 45 cm + fenugreek intercropped with broccoli improved the growth and yield of broccoli and yield of intercrops.

REFERENCES

Chaterjee B, Ghanti P, Thapa U and Tripathy P. 2005. Effect of organic nutrition in sprouting broccoli (*brassica oleracea* I. var *Italica* Plenck). *Vegetable Science* **32**(1): 51-54.

Choudhary S, Soni A K and Jat N K. 2012. Effect of organic and inorganic sources of nutrients on growth, yield and quality of sprouting broccoli cv CBH-1. *Indian Journal Horticulture* **69**(4): 550-54.

Devi K B and Singh N I. 2012. Yield response of cabbage (*Brassica oleraceae* var *capitata*) cv Pride of India to varying levels of chemical fertilizers and vermicompost. *Journal of Agriculture and Veterinary Science* **1**(3): 8-11.

Faroda A S, Joshi N L, Singh R, Sexena A. 2007. Resource management for sustainable crop production inarid zone-A review. *Indian Journal Agronomy* **52**: 181-193.

Kumar M, Das B, Prasad K K and Kumar P. 2013. Effect of integrated nutrient management on growth and yield of broccoli (*Brassica oleracea* var *italica*) under Jharkhand conditions. *Vegetable Science* **40**(1): 117-20.

Maurya A K, Singh M P, Srivastava B K, Singh Y V, Singh D K, Singh S and Singh P K. 2008. Effect of organic manures and inorganic fertilizers on growth characters, yield and economic of sprouting broccoli cv Fiesta. *Indian Journal of Horticulture* **65**(1): 116-118.

Metson A J 1956. Methods of chemical analysis for soil survey samples Bull NZ Deptt Sci *Nat Res. Soil Bureau* **12**.

Mohapatra S K, Munsi P S and Mahapatra P N. 2013. Effect of integrated nutrient management on growth, yield and economics of broccoli (*Brassica oleracea* L var *italic* Plenck). *Vegetable Science* **40**(1): 69-72.

Olsen S R, Cale C V, Watonable F S and Dean L A. 1954. Estimation of available phosphorus in soil by extraction with sodium bicarbonate. Washington Circular No 939.

Panse V G and Sukhatme P V. 1985. *Statistical Methods for Agricultural Workers*. Indian Council of Agricultural Research, New Delhi, pp 381.

Renuka B and Ravishankar C. 1998. Effect of organic manures on growth and yield of tomato. *In: Proceedings of National Seminar on Changing Scenario in the Production System of Horticultural crops*, held at Tamil Nadu Agricultural University, Coimbatore, 28-30 August, pp 215-18.

Singh A 1992. 'Effect of crop geometry, nitrogen levels and intercrops on growth, yield and quality of cauliflower and cabbage'. PhD Thesis, CCS HAU, Hisar.

Singh K, Dhaka R S and Fageria M S. 2004. Response of cauliflower (*Brassica oleracea* var *botryris* L) cultivars to row spacing and nitrogen fertilization. *Progressive Horticulture* **36**(1): 171-173.

Sinha R K, Soni B K, Agarwal S, Shankar B and Hahn G. 2013. Vermiculture for organic horticulture: producing chemical free, nutritive and health protective foods by earthworms. *Agricultural Sciences* **1**(1): 17-44.

Subhiah B V and Asija G L. 1956. A rapid procedure for the estimation of available nitrogen in manures. *Current Science* **25**: 259-260.

Uddin J, Solaiman A H M and Hasanuzzaman M. 2009. Plant characters and yield of kohlrabi (brassica oleraceae var gongylodes) as affected by different organic manures. *Journal of Horticultural Science and Ornamental Plants* **1**(1): 01-04.

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Effect of sodium hypochlorite, citric acid, ascorbic acid, calcium chloride and packaging material on quality of minimally-processed cauliflower *(Brassica oleracea)*

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ABSTRACT

An experiment was conducted to find out the effect of minimal processing of cauliflower at Indian Agricultural Research Institute, New Delhi, during. The material of cauliflower, cv. Sweta was purchased from Najafgarh area, New Delhi. Pre-treatment of cauliflower florets with 150 ppm sodium hypochlorite for 2 minutes were found to be effective in reducing the bacterial population. Dipping of florets in 1% citric acid for 5 minutes was found to be highly effective in checking bacterial population (3.65 log cfu / g) in minimally-processed cauliflower. The maximum retention of firmness (18.08 N) was found in 1% $CaCl_2$ treated cauliflower followed by 0.5% citric acid + 0.5% $CaCl_2$. The minimally-processed cauliflower treated with 150 ppm sodium hypochlorite for 2 minutes and 1% citric acid for 5 minutes, packed in HDPE (500 gauge), LDPE (100 gauge) and PP (100 gauge) and stored at 5°C for 20 days. The least weight loss was observed in cauliflower stored in HDPE (2.35%) than florets of LDPE (4.64%). The respiratory rate and ethylene evolution were higher in cauliflower stored in LDPE packaging than HDPE and PP packaging. During storage, glucosinolates content was found to be higher in cauliflower packed in HDPE films (55.78 µmol of sinigrin /100 g), followed by LDPE (43.74 µmol of sinigrin / 100 g) and PP packaging (48.89 µmol of sinigrin / 100 g). Cauliflower florets packed in HDPE retained significantly higher level of ascorbic acid (43.49 mg / 100 g) and firmness (15.92 N). The electrolyte leakage was found to be lower in HDPE (29.5%), followed by PP (24.7%). The microbial population of minimally processed cauliflower was found to be below the safe level even after 20 days of storage.

KEY WORDS: Cauliflower, Minimal processing, Citric acid, Sodium hypochlorite, Ascorbic acid, Packaging material

Cauliflower (*Brassica oleracea* L. var. *botrytis*) is most important winter vegetable grown in India. This is a power house of health-promoting phytochemicals such as glucosinolates, vitamin C and phenolic compounds. Glucosinolates are unique class of sulphur-containing glycosides responsible for characteristic flavour. These secondary metabolites are known to have cancer chemoprotective activity (Fahey *et al.*, 2001; Kaur and Kapoor, 2001). The risk of cancer can be significantly reduced by an intake of as little as 10 g of cruciferous vegetable per day (Price *et al.*, 1998). Vitamin C is an antioxidant vitamin present in vegetables. Its role in diet is also significant in preventing cancer, heart disease and immune system (Wargovich, 2000, Kaur and Kapoor, 2001). With rapid changes in life-styles in metro cities, people find very less time for cooking of vegetables. Preparatory activities, *viz*. cutting, shredding, washing eliminates non-edible portion that substantially increases the kitchen garbage. In recent times, demand for minimally-processed vegetables is increasing. The main criteria of minimal processing are to keep the produce fresh without losing its nutritional quality. Minimally-processed fresh vegetables are more perishable than unprocessed fresh produce due to tissue damage resulting from processing. The main challenge

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is to combat the rapid quality deterioration in minimally-processed vegetables.

Microflora present in vegetables poses major safety problems and led to use various chemicals to control them. Sodium hypochlorite, a commonly used disinfectant is to be effective in checking microbial population. It is more effective against bacteria than fungi. This is due to oxidizing nature of sodium hypochlorite. Sodium hypochlorite gets hydrolysed in water and spilt into sodium hydroxide and hypochlorous acid. This hypochlorous acid diffuses through cell walls of bacteria and change oxidation-reduction potential of cell. Further, it inactivates triose phosphate dehydrogenase which is essential for digestion of food material (glucose) by microorganism. Thus, ability of microorganisms to function is inhibited.

Citric acid, ascorbic acid and calcium chloride are used to reduce the microbial profile in addition to nutritional and sensory quality improvement in minimally-processed vegetables. Modified atmosphere packaging is being employed as a potential means to extend shelf-life. Modified atmosphere within the bag can be beneficial in maintaining the quality of fresh cut vegetables by reducing the respiration rate, ethylene biosynthesis and its action. Modified atmosphere packaging in combination with refrigeration have profound effect in inactivating the enzymes involved in metabolic reactions and in reducing the microbial load. Although some of these techniques have been in use in different vegetables, these cannot be generalized for all the species and cultivars of vegetables. Therefore, an experiment was conducted to see the effect of different chemicals and packaging materials on quality of minimally-processed cauliflower.

MATERIALS AND METHODS

Freshly harvested curds of var. sweta were obtained from commercial growers from Najafgarh, New Delhi. They were transported to Post-Harvest Technology laboratory, IARI, New Delhi, within 4-5 hours of harvesting. They were washed with tap water and cut into florets (5.5 cm × 3 cm) using a sharp serrated knife. The florets were then washed again and air dried.

To reduce the initial microbial load, florets were dipped in 100 and 150 ppm sodium hypochlorite for 1, 2 and 4 minutes. Then, florets were packed in 500-gauge high-density polyethylene (HDPE) and stored at 5°C in a refrigerator. The treatments were replicated 3 times in a completely randomized design. The observations on bacterial and fungal population were recorded at 0 day and 7 day of storage.

Minimally-processed cauliflower were dipped in Generally Recognized As Safe (GRAS) chemicals such as 1% citric acid, 1% ascorbic acid, 1 % calcium chloride, 0.5% citric acid + 0.5% ascorbic acid, 0.5% ascorbic acid + 0.5% calcium chloride and 0.5% citric acid + 0.5% calcium chloride for 5 minutes. The treatments were replicated 3 times in a completely randomized design. The observations on firmness and bacterial and fungal population were recorded at an interval of 5 days.

Minimally-processed cauliflower were treated with 150 ppm sodium hypochlorite for 2 minutes and 1% citric acid for 5 minutes, packed in 500-gauge high-density polyethylene (HDPE), 100-gauge low-density polyethylene (LDPE) and 100-gauge polypropylene (PP) and stored at 5°C for 20 days. The experiment was laid in a completely randomized design. Observations were recorded at 5 days interval up to 20 days for weight loss, firmness, electrolyte leakage, rate of respiration, ethylene evolution, glucosinolates, ascorbic acid and bacterial and fungal count.

Firmness of cauliflower florets was determined by using Instron universal testing machine (Model 4201, Instron Inc, USA). The electrolyte leakage of cauliflower floret was measured by digital conductivity meter (Model Elico, India). The rate of respiration in cauliflower florets was determined by using Gas Chromatograph (Model Nucon 5500). The rate of ethylene evolution in cauliflower florets was determined by using Gas Chromatograph (Model 5890). Glucosinolates were analyzed as per the method described by Vallego *et al.*, 2002. Total titratable acidity was expressed as per cent citric acid (AOAC, 1984). Ascorbic acid was determined by titrating a known weight of sample with 2, 6-dichlorophenol indophenols dye using metaphosphoric acid as stabilizing agent (AOAC, 1984).

Counting of bacterial and fungal population was done in Standard Plate Count Agar (SPCA) and Martin Rose Bengal Agar (MRBA) medium respectively. Water blanks were prepared by taking 90 ml of water in 250 ml conical flask and were autoclaved. Ten gram of florets from each treatment was weighed and added to 90 ml water blank which gives a dilution of 10-1. From 10-1 dilution, series up to 10-5 were prepared. Dilutions of 10-4 and 10-5 for bacteria and 10-2 and 10-3 for fungi were taken for inoculation. The results were expressed as log cfu / g of floret.

RESULTS AND DISCUSSION

Washing with tap water reduced the bacterial population from 7.25 to 4.30 log cfu / g. It is evident that sodium hypochlorite treatment drastically reduced the bacterial population in minimally-processed cauliflower (Table 1). On the 7 day of storage, sodium hypochlorite dosages of 100 ppm for 4 min or 150 ppm for 2 min or 150 ppm for 4 min were found to be at par with each other having significantly low level of

Table 1. Effect of sodium hypochlorite on bacterial population (log cfu / g) of minimally-processed cauliflower during storage

Treatment	Storage period (days)		
	0	7	Mean
100 ppm + 1 min.	3.50 [a]	5.02 [c]	4.26
100 ppm + 2 min.	3.33 [a]	5.01 [c]	4.17
100 ppm + 4 min.	3.00 [a]	4.97 [b]	3.98
150 ppm + 1 min.	3.42 [a]	5.04 [c]	4.23
150 ppm + 2 min.	2.92 [a]	4.95 [b]	3.93
150 ppm + 4 min.	2.98 [a]	4.90 [b]	3.94
Control	4.30 [b]	6.76 [d]	5.53
Initial population	7.25 log cfu / g		
CD (p=0.05)			
Treatment (T)	0.08		
Storage period (S)	1.23		
T × S	110		

Table 2. Effect of pre-treatment on firmness (N) of minimally-processed cauliflower

Treatment	Storage period (days)			
	0	5	10	Mean
1% CA	18.00	16.72	15.90	16.87
1% AA	18.00	16.63	15.97	16.87
1% $CaCl_2$	18.25	18.01	17.98	18.08
0.05% CA + 0.5% AA	18.00	16.50	15.12	16.54
0.05% CA + 0.5% $CaCl_2$	18.15	16.82	16.72	17.23
0.05% CA + 0.5 $CaCl_2$	18.15	16.58	16.70	17.14
Control	19.00	15.62	14.52	16.38
Mean	18.22	16.69	16.13	-
CD (p=0.05)				
Treatment (T)		0.92		
Storage period (S)		1.55		
T × S		0.01		

Table 3. Effect of pretreatment on bacterial population (log cfu / g) of minimally-processed cauliflower

Treatment	Storage period (days)			
	0	5	10	Mean
1% CA	3.15	3.68	4.13	3.65
1% AA	3.78	3.90	4.55	4.07
1% $CaCl_2$	5.01	6.05	7.09	6.05
0.05% CA + 0.5% AA	3.45	3.79	4.48	3.91
0.05% CA + 0.5% $CaCl_2$	3.92	3.99	4.53	4.15
0.05% CA + 0.5 $CaCl_2$	4.00	4.12	5.12	4.41
Control	5.52	6.62	7.52	6.55
Mean	4.12	4.59	5.34	-
Initial population	6.72 log cfu / g			
CD (p =0.05)				
Treatment (T)		0.40		
Storage period (S)		0.63		
T × S		0.56		

bacterial count compared to other treatments.

The data on firmness revealed that pre-treatment with 1% $CaCl_2$ resulted in maximum retention of firmness (18.08 N) which was at par with only 0.5% citric acid + 0.5% $CaCl_2$ treatment (Table 2).

The pre-treatment with 1% citric acid was found to be highly effective in checking the bacterial population to the least level (3.65 log cfu / g) which was at par with treatment combination of 0.5% citric acid + 0.5% ascorbic acid (3.91 log cfu / g) (Table 3). With respect to fungal population, non-significant difference was observed due to various pre-treatments (Table 4). Food-borne pathogens, *Salmonella typhimurium*, *Shigella flexneri* and *Escherichia coli*, were found to be absent in treated florets even after 10 days.

Minimally processed vegetables are living tissues that are undergoing catabolic activities. Packaging of minimally, processed cauliflower in permeable polymeric film can reduce O_2 concentration and increase CO_2 concentration in package atmospheres, thereby slowing quality changes and increasing product shelf-life.

The weight loss was less in cauliflower stored in HDPE films and it was significantly lower (2.35%) than florets of LDPE (4.64%) film (Fig. 1). This is due to lower water vapour permeability in HDPE than LDPE. The lower weight loss might be partly due to low temperature and high humidity during storage.

The minimally-processed cauliflower stored in HDPE films showed significantly higher values of firmness than that of LDPE packaging (Table 5) due to less ethylene production (Watada *et al.*, 1996). The firmness was in decreasing order with the advancement of storage period and it was non-significant up to 15th days of storage and became significant beyond this period.

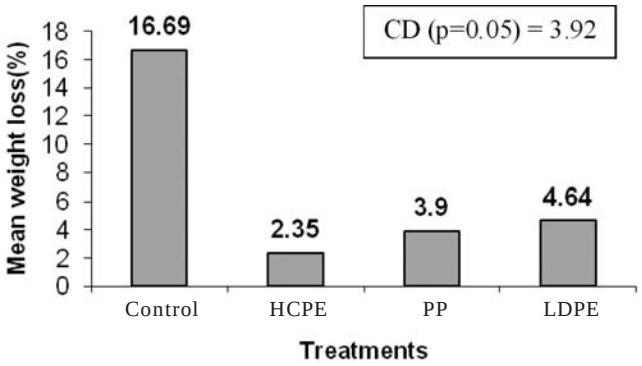

Fig. 1. Effect of packaging material on weight loss (%) during storage of minimally processed cauliflower

Electrolyte leakage is an indirect measure of cell membrane damage. There was lowest loss of electrolytes in HDPE packaging which was at par with PP (Table 6). It also clear from the data that there was a progressive increase in the loss of electrolytes with advancement of storage period and became significant only on 15th day of storage.

Cauliflower immediately after cutting exhibited high respiratory rate and ethylene production. The respiratory rate and ethylene evolution was higher in cauliflower stored in LDPE packaging than HDPE and PP packaging (Figs. 2 and 3). There was a slight increase in respiration during storage period. The similar trend was observed in ethylene production also.

Figure 4 shows that effect of modified atmosphere packaging on the retention of glucosinolate (Sinigrin) content of minimally processed cauliflower on 20th of day of storage. The glucosinolates content was found to be highest in cauliflower packed in HDPE films (55.78 µ mol of sinigrin / 100 g), followed by LDPE (48.89 µ mol of sinigrin / 100g) and PP packaging (43.74 µ mol of sinigrin / 100 g). The lower content of glucosinolates in

Table 4. Effect of pretreatment on fungal population (log cfu / g) of minimally-processed cauliflower

Treatment	Storage period (days)			
	0	5	10	Mean
1% CA	0.24	0.67	1.30	0.73
1% AA	0.24	0.68	1.43	0.78
1% $CaCl_2$	0.24	0.70	1.43	0.79
0.05% CA + 0.5% AA	0.24	0.66	1.35	0.75
0.05% CA + 0.5% $CaCl_2$	0.24	0.69	1.45	0.79
0.05% CA + 0.5 $CaCl_2$	0.24	0.70	1.43	0.79
Control	0.24	1.58	2.90	1.57
Mean	0.24	0.81	1.61	
CD (p =0.05)				
Treatment (T)		0.03		
Storage period (S)		1.38		
T × S		0.77		

Table 5. Effect of modified atmosphere packaging on firmness (N) in minimally-processed cauliflower

Treatment	Storage period (days)				
	5	10	15	20	Mean
HDPE	18.41	17.50	16.20	15.92	17.41
PP	17.93	16.92	15.83	14.90	16.92
LDPE	17.46	16.40	15.00	12.17	16.01
Control	15.12	14.60	11.50	8.20	13.68
Mean	17.23	16.36	14.63	12.79	
Initial value = 19 N					
CD (p =0.05)					
Treatment (T)		2.54			
Storage period (S)		2.92			
T x S		2.78			

Table 6. Effect of modified atmosphere packaging on electrolyte leakage (%) in minimally-processed cauliflower

Treatment	Storage period (days)				
	5	10	15	20	Mean
HDPE	17.15	25.60	29.50	32.50	23.57
PP	19.64	26.13	31.45	34.18	24.90
LDPE	21.17	28.34	33.59	37.68	26.78
Control	20.00	33.00	40.00	49.00	30.82
Mean	19.49	28.27	33.64	38.34	
Initial value = 13.10 %					
CD (p =0.05)					
Treatment (T)		3.13			
Storage period (S)		16.10			
T x S		11.54			

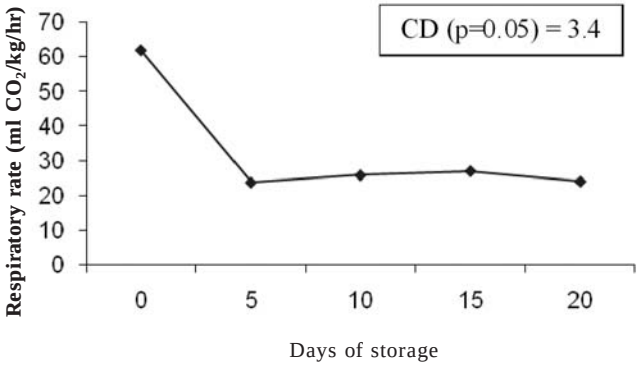

Fig. 2. Effect of packaging material on rate of respiration (ml CO_2/kg/hr) during storage of minimally-processed cauliflower

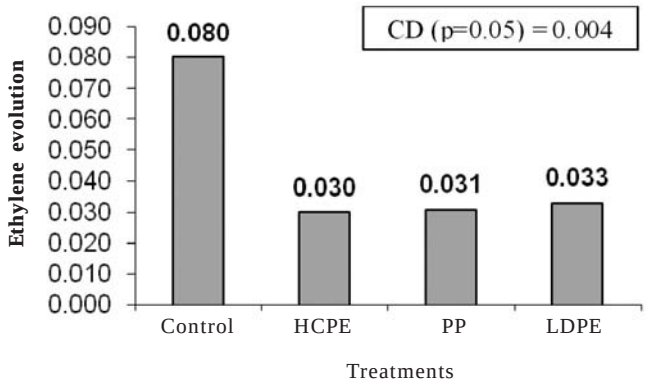

Fig. 3. Effect of packaging material on mean rate of ethylene evolution (µmol/kg/hr) in minimally-processed cauliflower

florets packed in LPDE might be due to cell damage, which is evident from data on firmness and electrolyte leakage. Cell rupture and membrane damage would have paved the way to glucosinolates degrading enzyme, myrosinase to come into contact with glucosinolates (Hansen *et al.*, 1995). In addition to this, comparatively higher concentration of O_2 in LDPE would have been the responsible factor for degradation of glucosinolates (Rangkadilok *et al.* 2002).

Data presented in Table 7 show the effect of modified atmosphere packaging on ascorbic acid content of minimally processed cauliflower. Packaging

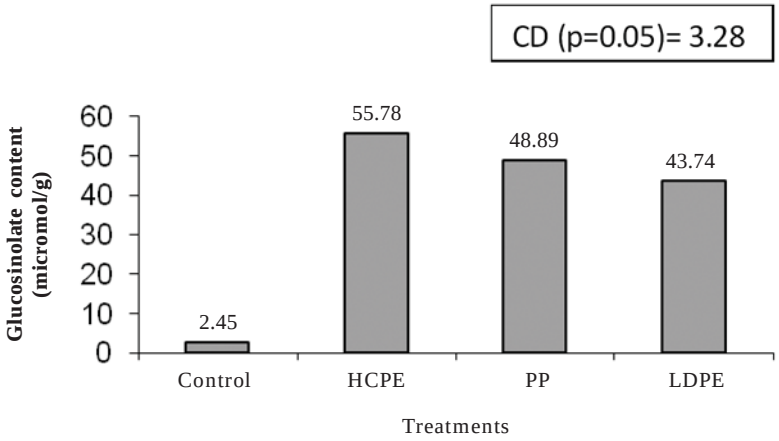

Fig. 4. Effect of packaging material on glucosinolates (micro mol/100g) in minimally-processed cauliflower

in HDPE film resulted in the higher (43.49 mg/100 g) retention which was at par with PP packaging (41.41 mg/100 g). At the end of storage, HDPE film packed cauliflower florets retained the higher level of ascorbic acid due to creation of modified atmospheric condition with reduced level of O_2 in HDPE films.

There was a significant reduction in bacterial population of minimally-processed cauliflower (Table

Table 7. Effect of modified atmosphere packaging on ascorbic acid content (mg/100g) in minimally processed-cauliflower

Treatment	Storage period (days)				
	5	10	15	20	Mean
HDPE	46.32	42.82	39.62	37.71	43.49
PP	44.86	39.60	37.33	34.24	41.41
LDPE	43.47	38.43	36.14	33.13	40.43
Control	42.29	36.17	28.83	20.00	35.68
Mean	44.24	39.26	35.48	31.26	
Initial value = 51 mg/100g					
CD (p =0.05)					
Treatment (T)		1.40			
Storage period (S)		5.45			
T × S		5.12			

Table 8. Effect of modified atmosphere packaging on bacterial population (log cfu/g) in minimally-processed cauliflower

Treatment	Storage period (days)				
	5	10	15	20	Mean
HDPE	2.43	2.90	3.20	3.63	2.84
PP	2.62	3.23	3.31	3.82	3.01
LDPE	3.01	3.45	3.56	3.99	3.21
Control	2.90	3.70	4.80	5.90	3.87
Mean	2.74	3.32	3.72	4.33	
Initial value = 2.06 log cfu/g					
CD (p =0.05)					
Treatment (T)		0.43			
Storage period (S)		0.98			
T × S		1.18			

8). The combined effect of 150 ppm chlorine and 1% citric acid also reduced the initial population of 7.1 log cfu / g to 2.06 log cfu / g. It reduced the microbial activity by lowering the pH of the product (Pal *et al.* 2004). The reduction in microbial population is the combined effect of modified atmosphere, low temperature storage and chemical treatment. The minimally processed cauliflower could be stored up to 15 days in all the packaging material, HDPE, LDPE and PP films. There was no significant difference in fungal population observed irrespective of packaging material and storage period. Food borne pathogens, *Salmonella typhimurium*, *Shigella flexneri* and *Escherichia coli* were found to be absent in treated florets even after 10 days.

The sensory score of minimally-processed cauliflower was significantly higher in florets packed with packaging materials. With respect to storage period, the florets scored above 6 hedonic points in all the combinations and were considered acceptable up to 15 days from marketing point of view.

Thus, minimally processed cauliflower could be stored in HDPE and PP films up to 15 days without significant loss of firmness, electrolyte leakage and ascorbic acid. Minimally processed cauliflower packed in HDPE, PP and LDPE showed significantly lower bacterial population up to 15 days. Ready-to-use cauliflower has the potential of becoming an important product with the changing life styles. Importantly, processing will provide employment opportunities to many people, reduce transportation cost (from field to market), keep the metro cities environment free from pollution and provide convenience to housewives.

REFERENCES

AOAC Washington. 1984. *Official Method of Analysis,* 9th edn. Association of Official Agricultural Chemists, Washington, D.C.

Fahey J W, Zalemann A T and Talalay P. 2001. The chemical diversity and distribution of glucosinolates and isothiocyanates among plants. *Phytochemistry* **56**: 5-51.

Hansen M, Moller P and Sorensen H. 1995. Glucosinolates in Broccoli stored under controlled atmosphere. *J. American Soc. Hort. Sci.* **120**(6): 1069-74.

Kaur C and Kapoor H C. 2001. Antioxidant activity and quality of minimally processed Indian Cabbage (Brassica oleracea var. capitata). *Journal of Food Processing and Preservation* **25**(5): 367-80.

Pal R K, Singh Manoj, Sen Nita, Singh C P and Behera J K. 2004. Utilization of commercially unmarketable bell pepper through minimal processing and modified atmosphere packaging (MAP) technology. *Food and Bioprocess Engineering,* pp. 474.

Price K R, Casuscelli F, Colquhonu I J and Rhodes M J C. 1998. Composition and content of flavonol glycosides in broccoli florets. *(Brassica oleracea)* and their fate cooking. *J. Sci. Food Agri.* **77**: 468-72.

Rangkadilok N, Tomkins B, Nicolas M E, Premier R R, Bennett R N, Eagling D R and Taylor P W. 2002. The effect of post harvest and packaging treatments on glucoraphanin concentration in Broccoli (*Brassica oleracea* var. *italica*). *J. Agric. Food Chem.* **50**: 7386-91.

Vallejo F, Barberan FAT, Bena vente Garcia A G and Garcia Viguera C. 2002. Total and individual glucosinolate contents in inflorescences of eight broccoli cultivars grown under various climatic and fertilization conditions. *J. Sci. Food Agric.* **83**: 307-13.

Wargovich M J. 2000. Anticancer properties of fruits and vegetables. *Hort. Science* **35**: 573-75.

Watada A E, Ko N P and Minott D A. 1996. Factors affecting quality of fresh cut horticultural products. *Post Harvest Biol. Technol.* **9**: 115-25.

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Effect of plant spacing on yield and quality of strawberry *cv.* Festival in West Garo Hills, Meghalaya

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ABSTRACT

An experiment was conducted to see the effect of plant spacing on growth, flowering, fruiting, yield and quality of strawberry *(Fragaria ananassa* L.*) cv.* Festival in West Garo Hills of Meghalaya during 2014-2015. The study area is situated approximately between the longitudes 90° 30' and 89° 40'E, and the latitudes of 26° and 25° 20' N. Runners of strawberry were planted under 9 different spacings, *viz.* T_1, 15 cm × 25 cm × 100 cm; T_2, 15 cm × 30 cm × 100 cm; T_3, 15 cm × 35 cm × 100 cm; T_4, 20 cm × 25 cm × 100 cm; T_5, 20 cm × 30 cm × 100 cm; T_6, 20 cm × 35 cm × 100 cm; T_7, 25 cm × 25 cm × 100 cm; T_8, 25 cm × 30 cm × 100 cm; T_9, 25 cm × 35 cm × 100 cm (plant-to-plant, row-to-row and trench-to-trench). The highest plant height and spread in north-south and east-west direction were recorded with spacing of 15 cm × 25 cm × 100 cm (T_1). The highest TSS: acid ratio in fruits (36.80) was observed in T_7 (25 cm × 25 cm × 100 cm). Plants spaced at 15 cm × 30 cm × 100 cm (T_2) produced highest number of flowers and fruits, fruit setting, larger fruits of higher fruit weight, highest yield / plant and highest productivity and also highest vitamin C content in fruits.

Key Words: Strawberry, Yield, Quality, West Garo Hills, Plant spacing

Strawberry *(Fragaria ananassa* L.*)*, belonging to the family Rosaceae, is most popular soft fruit cultivated in plains as well as in hills up to an elevation of 3,000 m in humid or dry region. Strawberry gives quickest returns in shortest possible time. It is considered as a complete fruit with 98% edible portion and is rich in vitamin C and iron. It provides a good dose of fibre, folic acid, manganese and potassium, and good source of antioxidants. The cultivation of strawberry in India was confined initially to temperate hilly regions, but with the introduction of day neutral cultivars, it has spread to subtropical and tropical regions also. West Garo hills being a subtropical region is favourable for cultivation of its day neutral varieties. However, negligible research has been done on strawberry in this region.

The spacing adopted during planting and cultural practices followed during cultivation like manuring, irrigation, mulching, weeding *etc.* influence vegetative growth, flowering, fruiting and production. Various studies report the effect of planting density on vegetative growth, reproductive development and performance in different strawberry cultivars, suggesting that manipulation of planting density could allow an increase in productivity (Tamiru, 1996; Perez *et al.* 2004). However, response depends upon cultivar, prevailing weather conditions and cultural practices. (Perez *et al.* 2005). According to Wilson and Dixon (1988) and Hancock (1999), optimum spacing depends on force of material and climate. Therefore, an experiment was conducted with 9 spacings to evaluate growth, yield and fruit quality of strawberry.

MATERIALS AND METHODS

The experiment was conducted at the farm of Department. of Rural Development and Agricultural Production, NEHU, Tura Campus, situated in Duragre, Rongram block, West Garo Hills district. It is situated approximately between the longitudes 90° 30' and 89° 40' E, and the latitudes of 26° and 25° 20' N. The runners of strawberry were planted in double row system with 9 spacing treatments and three replications per treatment with 50 plants per replication during 2014-

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2015. The planting was done on 16 September. The treatments were: T_1 : 15 cm × 25 cm × 100 cm; T_2 : 15 cm × 30 cm × 100 cm; T_3 : 15 cm × 35 cm × 100 cm; T_4 : 20 cm × 25 cm × 100 cm; T_5 : 20 cm × 30 cm × 100 cm; T_6 : 20 cm × 35 cm × 100 cm; T_7 : 25 cm × 25 cm × 100 cm; T_8 : 25 cm × 30 cm × 100 cm; T_9 : 25 cm × 35 cm × 100 cm (plant-to-plant, row-to-row and trench-to-trench spacing).

The height, spread and number of crowns were recorded. Flowering and fruiting behaviour were studied with respect to date of first flowering, date of first harvesting, duration from first flowering to first harvesting, number of flowers / plant, number of fruits / plant and percentage of fruit setting. Yield characters like fruit weight, number of fruits / plant, yield / plant (g) and productivity (q / ha) were recorded. The total soluble solids (TSS), acidity, total sugar, vitamin C and TSS: acid ratio were also recorded. The TSS was determined with the help of a Hand Refractometer and expressed in °Brix. Acidity was estimated as per titrating the juice against N / 10 NaOH and expressed as percent citric acid. Total sugar (%) and vitamin C content (mg / 100g pulp) were estimated by the standard procedure of AOAC (2012). The data were statistically analysed (Panse and Sukhatme, 1990).

RESULTS AND DISCUSSION

The highest plant height (21.10 cm) and plant spread in east-west (28.43 cm) and north-south (32.47 cm) directions were observed in T_1 (Table 1). The highest number (4.61) of crowns was recorded in plants of 25 cm × 35 cm × 100 cm spacing (T_9). The duration from first flowering to first harvesting was lowest (29.36 days) in T_6 and highest (43.66 days) in T_8 (Table 1). Fruits of T_8 recorded highest fruit weight (15.26 g) and fruit diameter (29.73 mm). Highest fruit length of 36.57 mm was observed in T_5, followed by 36.36 mm in T_2 and 36.33 mm in T_8 (Table 2). Al- Ramamneh *et al.* (2013) also observed higher fruit weight and size under wider spacing.

Highest number (30.2) of flowers was observed in T_1. Highest number of fruits (22.67) and fruit setting (75.40%) were observed in T_2 (Table 1). The highest yield / plant (342.77 g) was recorded in T_2 (Table 2). The highest productivity (351.55 q / ha) was observed in T_2, followed by 291.26 q / ha in T_1 which were the closest of spacings, followed in our study. The lowest productivity (134.58 q / ha) was noted in T_9 which was the widest spacing, (Table 2). Albregts (1971), Freeman (1981), Nestby (1994), Perez *et al.* (2004), Perez *et al.* (2005), Paranjpe *et al.* (2008) and Laugale *et al.* (2012) also observed that increasing the plant density increased yield per unit area and lower plant densities exhibited lower productivity.

Table 1. Growth, flowering and fruiting behaviour of strawberry *cv.* Festival under different plant spacing

Treatment	Plant height (cm)	Spread E-W (cm)	Spread N-S (cm)	Number of crowns	Date of first flowering	Date of first harvesting	Duration from first flowering to first harvesting	Number of flowers / plant	Number fruits / plant	Fruit setting (%)
T_1	21.10	28.43	32.47	3.00	19 Oct - 15 Dec	14 Nov - 20 Feb	37.02	30.20	18.50	61.25
T_2	18.43	26.07	28.10	2.77	21 Oct - 4 Jan	18 Nov - 17 Jan	31.57	30.06	22.67	75.40
T_3	16.80	25.80	27.23	3.60	20 Oct - 17 Jan	14 Nov - 7 Feb	34.67	19.40	13.07	67.35
T_4	15.40	21.90	26.90	3.13	25 Oct - 19 Jan	24 Nov - 20 Feb	42.27	23.27	14.48	63.14
T_5	16.03	23.67	26.73	3.60	22 Oct - 14 Jan	18 Nov - 14 Feb	35.17	25.51	18.37	72.02
T_6	13.63	17.83	22.13	3.77	18 Oct - 19 Jan	17 Nov - 18 Feb	29.36	27.29	16.74	60.87
T_7	14.07	20.53	20.57	3.00	20 Oct - 15 Jan	14 Nov - 25 Feb	34.80	27.26	19.13	70.20
T_8	13.10	18.17	18.83	3.45	17 Oct - 2 Dec	14 Nov - 7 Feb	43.66	27.33	17.78	63.43
T_9	12.70	16.97	9.62	4.61	16 Oct - 10 Jan	18 nov - 4 Feb	30.17	23.16	16.31	69.86
SEm±	1.22	2.15	2.32	0.29	-	-	2.56	1.42	1.03	2.5
CV (%)	9.5	11.91	12.05	10.27	-	-	8.36	6.73	7.24	4.57
CD at (5%)	2.58	4.57	4.93	0.61	-	-	5.42	3.02	2.19	5.31

However, highest TSS and total sugar were observed in T_5 with 8.50 °Brix and 5.16% respectively. The highest TSS : acid ratio of 36.80 was recorded in T_7 which also showed lowest acidity of 0.21%, while highest vitamin C content of 99.08 mg / 100 g was noted in T_2 (Table 3). Better quality fruits from plants spaced at wider spacing may be due to lesser competition for sunlight, water and nutrition.

The plant height and spread were higher in treatments with higher plant densities (T_1, T_2 and T_3). Fruit weight was also higher in T_1 and T_2. Yield per plant and yield / ha were highest in T_2 (higher density). This implies that spacing adopted in treatments T_1, T_2 and T_3 provide ample amount of nutrients, water and light necessary for proper growth and production. There is further scope for research trials with reduced spacing than considered in our study to accommodate more number of plants in a unit area, thereby improving the chances of further increase in productivity.

Thus, it is concluded that plants spaced at 15 cm ×

Table 2. Fruit weight, size and yield of strawberry *cv*. Festival under different plant spacing

Treatment	Fruit weight (g)	Fruit length (mm)	Fruit diameter (mm)	Yield	
				(g / plant)	(q / ha)
T_1	14.86	35.69	28.62	273.06	291.26
T_2	15.19	36.36	29.17	342.77	351.56
T_3	14.32	35.99	28.46	189.33	186.99
T_4	13.70	35.43	28.89	200.80	160.64
T_5	14.94	36.57	29.32	281.14	216.26
T_6	14.78	35.36	29.47	248.44	184.02
T_7	14.65	34.19	28.52	276.43	176.92
T_8	15.26	36.33	29.73	273.88	168.54
T_9	13.92	36.15	29.24	227.11	134.58
SEm±	0.46	NS	NS	25.49	19.01
CV (%)	0.82	2.57	3.84	12.14	11.20
CD (5%)	0.97	NS	NS	54.04	40.14

NS= Non- significant
T_1:15 cm × 25 cm × 100 cm; T_2: 15 cm × 30 cm × 100 cm; T_3: 15 cm × 35 cm × 100 cm;× 35 × 100 cm (plant-to-plant, row-to-row and trench-to-trench spacing)

Table 3. Fruit quality parameters of strawberry *cv*. Festival under different plant spacing

Treatment	TSS (°Brix)	Acidity (%)	TSS : acid ratio	Sugar (%)	Vitamin C (mg / 100g)
T_1	7.67	0.26	29.48	4.23	73.07
T_2	7.33	0.24	31.74	4.30	99.08
T_3	8.17	0.28	29.36	3.97	76.92
T_4	7.83	0.28	28.20	2.77	64.42
T_5	8.50	0.30	28.72	5.16	68.03
T_6	8.00	0.30	26.80	4.39	75.07
T_7	7.67	0.21	36.80	4.45	89.45
T_8	7.67	0.24	33.03	4.14	56.06
T_9	7.83	0.26	30.12	4.55	78.73
SEm±	NS	0.02	1.39	0.26	3.45
CV (%)	5.89	8.39	5.61	7.62	5.58
CD (5%)	NS	0.04	2.95	0.55	7.3

NS= Non- significant
T_1:15 cm × 25 cm × 100 cm; T_2: 15 cm × 30 cm × 100 cm; T_3: 15 cm × 35 cm × 100 cm;× 35 × 100 cm (plant-to-plant, row-to-row and trench-to-trench spacing)

30 cm × 100 cm showed best performance in terms of number of flowers and fruits, fruit setting (%), larger fruits, higher fruit weight, highest yield / plant, highest productivity and highest vitamin C content.

REFERENCES

Albregts E E. 1971. Influence of plant density on strawberry fruit production. Florida Agricultural Experiment Stations Journal Series No. 4146.http: / / extension.arizona.edu / sites / extension.arizona.edu / files / pubs / az1143 36.pdf Accessed on 1.09.14.

Al-Ramamneh E, Al-Rawashdeh Z, Karajeh M and Abu-Romman S. 2013. Plant Response of Strawberry to Intra-row Spacing and Growing Conditions in South of Jordan. *Asian Journal of Plant Sciences*. **12** : 201-07.

AOAC Washington. 2012. Official Methods of Analysis. Association of Official AnalyticalChemists, AOAC, Washington DC.

Freeman B. 1981. Response of strawberry fruit yields to plant population density. *Australian Journal of Experimental Agriculture and Animal Husbandry* **21**(110) : 349-53.

Hancock J F. 1999. Strawberries. CABI International. Wallingford, UK.

Laugale V, Lepse L, Strautina S, Krasnova I and Seglina D. 2012. Effect of planting density and plastic soil mulch on strawberry plant development, yield and fruit quality. *Acta Horticulturae* **926** : 517-23.

Nestby R. 1994. Effects of bed height, plant spacing and cultivar on strawberry yield and fruit classification. *Norwegian Journal of Agricultural Sciences* **8** : 127-33.

Paranjpe A V, Cantliffe D J, Stoffella P J, Lamb E M and Powell C A. 2008. Relationship of plant density to fruit yield of 'Sweet Charlie' strawberry grown in a pine bark soilless medium in a high-roof passively ventilated greenhouse. *Scientia Horticulture* **115**(2) : 117-23.

Panse V G and Sukhatme P V. 1990. Statistical Methods for Agricultural Workers. I.C.A.R. New Delhi.

Perez De Camacaro M, Carew J and Battey N. 2005. Effect of planting density on vegetative and reproductive growth of strawberry *cv*. *Elsanta. Bioagro* [online] **17**(1) : 11-15.

Perez De Camacaro M E, Camacaro GJ, Hadley P, Dennett M D, Battey N H and Carew J G. 2004. Effect of plant density and initial crown size on growth, development and yield in strawberry cultivars Elsanta and Bolero. *J. Hort. Sci. Biotech* **79**(5) : 739-46.

Tamiru M. 1996. 'The effect of density and initial plant size on radiation absorption, growth and yield in strawberry (Fragaria × ananassa Duch. *cv*. Elsanta)' M.Sc. Thesis, The University of Reading, Reading England.

Wilson F M and Dixon R M. 1988. Strawberry growth and yield related to plant density using matted row husbandry. *Journal of Horticultural Science* **63** : 221-27.

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Bacterial-fungal interaction in stimulation of growth and amelioration of physiological condition of citrus *(Citrus* spp.*)*

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ABSTRACT

An experiment was conducted to find out the effect of arbuscular mycorrhizal fungi *(Glomus intraradices)* and mycorrhizal helper bacteria (phosphorus-solubilising bacteria, *i.e.* PSB containing mixture of *Bacilus subtilis* and *B. megatherium*, *Azospirillum brasilence* and *Providencia* sp. on growth and physiological status of two citrus rootstocks, Troyer citrange *(Citrus sinensix* × *Poncirus trifoliata)* and Cleopatra mandarin *(Citrus reshni)*. The inoculation was done in soil during transplantation of seedlings in plastic containers. Troyer citrange co-inoculated with *G. intraradices* and PSB and Cleopatra mandarin co-inoculated with *G. intraradices* and *A. brasilence* significantly recorded highest root colonization 30, 60 and 120 days after inoculation (DAI). The inoculation of *G. intraradices* along with PSB or *A. brasilence* exhibited highest growth performance in terms of plant height, stem diameter, number of leaves and leaf area in both the rootstocks. The co-inoculation of *G. intraradices* and PSB recorded maximum relative water content, leaf chlorophyll and total carotenoids content in both the rootstocks at all the growth stages. The significantly more content of total phenols and proline were recorded in leaves of both the rootstocks co-inoculated with *G. intraradices* and *A. brasilence* and *G. intraradices* and PSB at all the growth stages, respectively. The soil application of *G. intraradices* along with PSB, *A. brasilence* or *Providencia* sp. recorded maximum total soluble sugars in leaves. Maximum photosynthesis, stomatal conductance and minimum leaf respiration were noticed in both the rootstocks co-inoculated with G. intraradices and PSB.

Key Words: Arbuscular mycorrhizal fungi, Citrus rootstocks, Leaf respiration rate, Mycorrhizal helper bacteria, Proline, Photosynthesis, Stomatal conductance, Total phenols

Mycorrhizal symbiosis is the most important mutualistic association between specific group of fungi from soils and roots of terrestrial plants (Gadkar *et al.*, 2001). Agricultural and horticultural crops have shown benefit from arbuscular mycorrhizal (AM) fungi on a worldwide basis (Mosse, 1973). Ubiquitous AM fungi and plant-growth-promoting bacteria are components of natural systems that benefit the host plant (Barea and Jeffries, 1995). The cooperation of AM fungi and bacteria in nutrient uptake by plants may be due to specific attributes of microorganisms, such as the ability of certain bacteria to stimulate mycorrhizal formation and development. This group of bacteria are referred as mycorrhizal helper bacteria or MHB (Meyer and Linderman, 1986).

Citrus having few and short root hairs, is highly dependent on arbuscular mycorrhizae, because most common mutualistic symbiosis replaces part of the root-hair functions (Graham and Syvertsen, 1985). The study on co-inoculation of AM fungi and MHB is a promising approach either for low-input agricultural technologies or for the reestablishment of natural vegetation in a degraded area. Therefore, an experiment was conducted on two citrus rootstocks to evaluate the effect of AM fungi and mycorrhizal helper bacteria on growth and physiological attributes.

MATERIALS AND METHODS

The study was conducted on two citrus rootstocks, *viz.* Troyer citrange *(Citrus sinensis* × *Poncirus trifoliata)*

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and Cleopatra mandarin *(C. reshni)* collected from the Citrus Germplasm Block of Division of Fruits and Horticultural Technology, Indian Agricultural Research Institute (IARI), New Delhi, during 2009-2011. Seeds of both citrus rootstocks were surface sterilized by immersing in 70% alcohol for five minutes, followed by rinsing with sterile distilled water and then germinated on wet filter paper in Petri dishes at 28°C. After 7 days, these seedlings were transplanted in plastic containers (12 cm × 20 cm) containing a mixture of soil: sand: FYM (2:2:1) under glasshouse conditions. The potting mixture had EC (1:2) 6.35 mSm, $pH_{(1:2)}$ 7.92, HCO_3^- 1.14 gkg^{-1} and Cl^- 5910.75 ppm. The AM fungi, *Glomus intraradices* and three mycorrhizal helper bacterial strains, *viz.* phosphate-solubilising bacteria (*Bacilus subtilis* + *B. megatherium*), *Azospirillum* and *Providencia* were used in the experiment. *Glomus intraradices* and all bacterial strains were purchased from Division of Microbiology, IARI, New Delhi.

The AM fungi (20 g inocula per pot) either alone or in combination with helper bacteria were put 5 cm below the seedlings at transplanting in plastic containers. The seedlings were maintained in glasshouse with day night temperatures ranging from 27±1°C. Day lengths were being extended up to 16 h with cool white fluorescent lights at 630 µmol m^{-2} s^{-1}. Seedlings were watered on alternate days with sterile tap water. After two months, seedlings along with plastic containers were transferred to open conditions at high-density orchard of Division of Fruits and Horticultural Technology, IARI, New Delhi. The seedlings were irrigated at two-day intervals. Water used for irrigation had EC (1:2) 288 µS m^{-1}, $pH_{(1:2)}$ 7.48, HCO_3^- 1.0 meq l^{-1} and Cl^- 110.76 ppm.

The experimental design adopted was completely randomized design with 16 treatments and two replications per treatment. The treatment included T_1 , control troyer citrange; T_2 , troyer citrange inoculated with phosphate-solubilising bacteria (PSB); T_3 , troyer citrange inoculated with *A. brasilens*; T_4 , troyer citrange inoculated with *Providencia* sp.; T_5 , troyer citrange inoculated with *G. intraradices*; T_6 , troyer citrange co-inoculated with *G. intraradices* and PSB; T_7, troyer citrange co-inoculated with *G. intraradices* and *A. brasilense*; T_8, troyer citrange co-inoculated with *G. intraradices* and *Providencia* sp., T_9, control Cleopatra mandarin; T_{10}, Cleopatra mandarin inoculated with PSB; T_{11}, Cleopatra mandarin inoculated with *A. brasilense*; T_{12}, Cleopatra mandarin inoculated with *Providencia* sp.; T_{13}, Cleopatra mandarin inoculated with *G. intraradices*; T_{14}, Cleopatra mandarin co-inoculated with *G. intraradices*; and PSB; T_{15}, Cleopatra mandarin co-inoculated with *G. intraradices* and *A. brasilense*; T_{16}, Cleopatra mandarin co-inoculated with *G. intraradices* and *Providencia* sp. Two-year data were analyzed with SPSS package (SPSS 11.0) and significance of variance was estimated by applying F test at 5% level of significance.

The percentage of AM colonization in roots was analyzed 30, 60 and 120 days after inoculation (DAI) by clearing and staining of roots by the method (Philips and Hayman, 1970). Seedlings were analyzed for plant height, stem diameter, number of leaves and leaf area 60, 120 and 210 DAI. Relative water content (RWC) of fifth fully expanded leaf from the shoot apex was evaluated 60, 120 and 210 DAI by the method of Barrs and Weatherley (1962). Leaf pigments (chlorophyll a, chlorophyll b, total chlorophyll and total carotenoids) were assayed 120 and 210 DAI as per (Hiscox and Israelstam,1979). Total phenol content in leaf was estimated according to Malik and Singh (1980). Leaf proline content and total soluble sugars were estimated. Photosynthesis, leaf respiration and stomatal conductance were measured by using Infra Red Gas Analyser PS System II (Li-Cor 6200) from 9:00 AM to 11:00 AM in youngest fully emerged leaves (third from top) of both the rootstocks.

RESULTS AND DISCUSSION

Root Colonisation

The inoculation of AM fungi increased root colonization (Table 1). There was synergistic increase in root colonization of AM fungi due to MHB at different growth stages in both citrus rootstocks. Troyer citrange co-inoculated with *G. intraradices* and PSB significantly showed higher root colonization (34.37, 59.37 and 71.87%) 30, 60 and 120 DAI, which was statistically at par with troyer citrange co-inoculated with *G. intraradices* and *Azospirillum* (31.25%) and *G. intraradices* and *Providencia* (25.00%) 30 DAI and Troyer citrange co-inoculated with *G. intraradices* and *Azospirillum* (68.75%) 60 DAI, respectively. In Cleopatra mandarin, highest root colonization was found in seedlings inoculated with *G. intraradices* and *Azospirillum* at all growth stages (25.00, 43.75 and 56.25% 30, 60 and 120 DAI, respectively), which was statistically at par with seedlings inoculated with *G. intraradices* and PSB (37.50%) 60 DAI and *G. intraradices* and PSB (56.25%) 120 DAI. Non-mycorrhizal control citrus seedlings or seedlings inoculated with either of bacteria alone did not exhibit any AM fungal inoculation 30 and 60 DAI, but at 120 DAI these seedlings were also infected by native AM fungal strains, while their root colonization was lesser than 13%. Many of the descriptions of bacteria-induced promotion of fungal growth or development involve interactions between fungi and MHB, which aid mycorrhizal development between fungus and plant host (Kobayashi and Crouch,

Table 1. Root colonization of arbuscular mycorrhizal citrus seedlings as influenced by mycorrhizal helper bacteria

Treatment	Root colonization (%)		
	30 DAI	60 DAI	120 DAI
T_1	0.00 (0.72)*	0.00 (0.72)	6.25 (14.48)
T_2	0.00 (0.72)	0.00 (0.72)	9.37 (17.59)
T_3	0.00 (0.72)	0.00 (0.72)	9.37 (17.59)
T_4	0.00 (0.72)	0.00 (0.72)	6.25 (14.48)
T_5	21.87 (27.83)	34.37 (35.87)	46.87 (43.21)
T_6	34.37 (35.87)	59.37 (50.42)	71.87 (58.01)
T_7	31.25 (33.99)	46.87 (43.21)	68.75 (56.01)
T_8	25.00 (29.82)	40.63 (39.58)	62.50 (52.30)
T_9	0.00 (0.72)	0.00 (0.72)	6.25 (14.48)
T_{10}	0.00 (0.72)	0.00 (0.72)	12.50 (20.71)
T_{11}	0.00 (0.72)	0.00 (0.72)	9.37 (17.59)
T_{12}	0.00 (0.72)	0.00 (0.72)	6.25 (14.48)
T_{13}	18.75 (25.35)	28.13 (31.99)	37.50 (37.70)
T_{14}	18.75 (25.66)	37.50 (37.76)	56.25 (48.59)
T_{15}	25.00 (29.82)	43.75 (41.41)	56.25 (48.62)
T_{16}	21.87 (27.83)	34.37 (35.87)	50.00 (45.00)
SE±	3.32	2.30	3.189
CD (0.05)	9.73	6.75	9.365

* Figures in parenthess are arc sin √% transformation data. DAI= days after inoculation

2009). Similar finding was observed by Pivato *et al.* (2009).

Growth parameters

Troyer citrange exhibited significant response in plant height to inoculation of *G. intraradices* alone (13.35 cm) 60 DAI, which was at par with seedlings inoculated with *G. intraradices* and PSB (12.60 cm) and seedlings inoculated with PSB alone (11.40 cm). At 120 and 210 DAI, co-inoculation of *G. intraradices* and PSB recorded highest plant height (61.50 and 111.75 cm, respectively), which was at par with *G. intraradices* and *Azospirillum* (55.60 and 107.55 cm) and G. intraradices and Providencia (106.90 cm) 120 and 210 DAI, respectively. In Cleopatra mandarin, highest plant height was obtained in seedlings inoculated with *Providencia* alone (9.10 cm) 60 DAI, while it was more in seedlings co-inoculated with *G. intraradices* 120 and 210 DAI and PSB (48.50 and 61.20 cm) and seedlings inoculated with *G. intraradices* and *Providencia* (46.80 and 64.10 cm) (Table 2). The maximum stem diameter was recorded in seedlings co-inoculated with *G. intraradices* and PSB 60, 120 and 210 DAI in troyer citrange (3.65, 7.05 and 16.47 mm, respectively) and seedlings inoculated with *Providencia* sp. alone 60 and 120 DAI (3.04 and 5.73 mm, respectively) and co-inoculated with *G. intraradices* and PSB (8.17 mm) at 210 DAI in Cleopatra mandarin (Table 2).

Significantly maximum number of leaves was produced in troyer citrange by inoculation of *Providencia* sp. alone (23.50 and 67.00 at 60 and 120 DAI, respectively). At 210 DAI, co-inoculation of *G. intraradices* and PSB significantly recorded maximum number of leaves (180.50). In Cleopatra mandarin, co-inoculation of *G. intraradices* and PSB significantly produced maximum number of leaves (18.50, 43.50 and 136.50 60, 120 and 210 DAI, respectively) (Table 2). In troyer citrange, significantly higher leaf area was recorded by co-inoculation of *G. intraradices* and PSB (4.13 cm^2), *G. intraradices* and *Azospirillum* (6.97 cm^2) and *G. intraradices* and *Providencia* (10.33 cm^2) 60, 120 and 210 DAI, respectively. For Cleopatra mandarin, co-inoculation of *G. intraradices* and PSB significantly produced maximum leaf area at all the growth stages (13.66, 14.73 and 15.61 cm^2) (Table 2). The response of citrus rootstocks to AM fungi and MHB is in harmony with the findings of Bashan and Levanony (1990) in papaya, Mamatha *et al.* (2002) in mulberry and Jaizme-Vega *et al.* (2004) in banana.

Table 2. Physical parameters of citrus seedlings as influenced by microbial inoculants

Treatment	Plant height (cm)			Stem diameter (mm)			No. of leaves/seedling			Leaf area (cm2)		
	60 DAI	120 DAI	210 DAI	60 DAI	120 DAI	210 DAI	60 DAI	120 DAI	210 DAI	60 DAI	120 DAI	210 DAI
T_1	9.25	20.95	37.20	2.33	3.02	6.01	14.50	40.50	126.50	1.09	1.82	2.96
T_2	11.40	43.45	93.80	2.49	3.17	7.61	19.00	59.00	142.50	1.63	2.60	3.30
T_3	10.00	40.05	86.95	2.74	3.55	7.27	15.50	42.50	102.00	1.77	2.66	3.21
T_4	10.40	49.95	93.45	3.01	5.41	12.24	23.50	67.00	141.00	1.93	3.06	3.94
T_5	13.35	48.60	58.55	3.08	5.18	15.38	12.50	35.00	108.50	2.13	3.44	3.93
T_6	12.60	61.50	111.75	3.65	7.05	16.47	20.50	55.00	180.50	4.13	5.50	7.46
T_7	10.75	55.60	107.55	3.18	6.76	12.76	18.50	45.00	160.00	3.86	6.97	9.44
T_8	9.65	54.25	106.90	2.67	5.79	10.57	13.50	33.00	123.00	3.09	6.86	10.33
T_9	7.35	19.40	26.05	2.31	3.59	3.59	6.50	16.50	42.00	3.78	4.60	5.76
T_{10}	7.70	27.15	36.55	2.84	4.90	6.08	8.50	18.50	54.50	6.03	6.97	7.93
T_{11}	8.85	26.80	42.80	2.91	4.39	7.05	12.50	30.00	88.00	7.15	8.27	9.39
T_{12}	9.10	27.50	40.85	3.04	5.73	6.92	11.50	28.50	77.50	6.12	6.75	7.98
T_{13}	7.25	31.30	32.85	2.68	4.74	4.57	11.00	17.00	47.00	5.79	6.38	8.46
T_{14}	7.55	48.50	61.20	2.89	5.22	8.17	18.50	43.50	136.50	13.66	14.73	15.61
T_{15}	7.30	46.80	64.10	2.59	3.93	5.86	8.50	15.50	73.50	7.06	7.96	9.23
T_{16}	8.90	32.50	48.85	2.45	4.69	5.31	11.00	24.00	90.00	4.79	5.79	7.05
SE±	0.72	2.43	1.93	0.12	0.33	0.60	2.32	3.69	4.46	0.46	0.46	0.48
CD (0.05)	2.10	7.13	5.66	0.34	0.98	1.76	6.81	10.83	13.13	1.34	1.36	1.41

Physiological Parameters

The result revealed that co-inoculation of *G. intraradices* and PSB recorded highest RWC (72.37, 86.09 and 88.19%) in troyer citrange and *G. intraradices* and PSB in Cleopatra mandarin (82.81, 87.56 and 89.18 per cent) 60, 120 and 210 DAI, respectively (Table 3). Higher RWC of citrus seedlings co-inoculated with AM fungi and PSB implied PSB improved the mutualistic interaction of AM fungi with citrus seedlings as stimulation of bacteria can promote the establishment and development of mycorrhizae (Dai *et al.,* 2008).

The co-inoculation of *G. intraradices* and PSB significantly increased chlorophyll a content in troyer citrange (0.89 and 1.76 mg/g fw) and Cleopatra mandarin (0.69 and 1.67 mg/g fw) 120 and 210 DAI, respectively (Table 3). The troyer citrange seedlings inoculated with PSB alone significantly had higher chlorophyll b content at 210 DAI (0.40 mg/g fresh weight). In Cleopatra mandarin, seedlings inoculated with *Azospirillum* alone had highest chlorophyll b content (0.18 mg/g fresh weight) at 210 DAI. The AM fungi *G. intraradices* in association with PSB significantly increased total chlorophyll content (a+b) at 120 and 210 DAI in both troyer citrange (1.37 and 2.22 and mg g^{-1} fresh weight, respectively) and Cleopatra mandarin (0.80 and 1.89 mg g^{-1} fresh weight, respectively) (Table 4). Thus increase in chlorophyll content might be due to changes in plant metabolism, as affected by AM fungi and MHB (Kim *et al.*, 2010).

The result also revealed that *G. intraradices* and PSB significantly increased total carotenoids content in both Troyer citrange and Cleopatra mandarin seedlings 120 DAI (0.32 and 0.27 mg/g fw, respectively). At 210 DAI, highest production of carotenoids was recorded in Troyer citrange seedlings inoculated with PSB alone (0.47 mg/g fw) and Cleopatra mandarin seedlings co-inoculated with *G. intraradices* and PSB (0.43 mg/g fw) and *Azospirillum* alone (0.43 mg/g fw) (Table 4). Similar result was found by Manoharan *et al.* (2010) in Erythrina variegata. The enhancement of photosynthetic rate in co-inoculated plants affects the translocation of soluble sugars to host roots, thus increasing fungal growth and activity in roots, thereby increasing leaf chlorophyll and carotenoid contents.

Plant phenolics are most widespread classes of secondary metabolites known to be involved in plant-microbe interactions. Phenols are important components of plant defence mechanism against diseases. It was found that *Glomus intraradices* along with *A. brasilenc*e had significant effect on total phenols in both Troyer

Table 3. Leaf relative water content of citrus seedlings as influenced by microbial inoculants (Mean of 2009-10 and 2010-11)

Treatment	Leaf relative water content (%)		
	60 DAI	120 DAI	210 DAI
T_1	58.18	68.61	69.39
T_2	66.66	70.76	71.83
T_3	64.56	78.59	79.06
T_4	58.92	74.51	75.96
T_5	65.66	74.41	77.53
T_6	72.37	86.09	88.19
T_7	67.78	82.13	82.84
T_8	61.74	76.52	80.47
T_9	59.12	69.02	71.11
T_{10}	63.43	82.27	82.48
T_{11}	60.85	80.74	81.54
T_{12}	67.99	75.38	76.69
T_{13}	77.52	82.13	82.57
T_{14}	82.81	87.56	89.18
T_{15}	80.12	83.25	84.61
T_{16}	75.54	76.78	81.17
SE±	0.39	0.68	0.59
CD (0.05)	0.121	1.16	2.01

Table 4. Pigment content (mg g^{-1} f w of leaf) of citrus seedlings as influenced by microbial inoculants

Treatment	Chlorophyll a		Chlorophyll b		Total chlorophyll		Carotenoids	
	120 DAI	210 DAI	120 DAI	210 DAI	120 DAI	210 DAI	120 DAI	210 DAI
T_1	0.42	1.37	0.31	0.26	0.73	1.64	0.14	0.32
T_2	0.65	1.53	0.45	0.40	1.10	1.93	0.23	0.47
T_3	0.55	1.40	0.46	0.27	1.01	1.66	0.21	0.35
T_4	0.74	1.55	0.48	0.30	1.22	1.85	0.26	0.40
T_5	0.62	1.51	0.46	0.33	1.07	1.83	0.19	0.37
T_6	0.89	1.76	0.48	0.28	1.37	2.22	0.32	0.43
T_7	0.84	1.60	0.46	0.31	1.30	1.99	0.28	0.42
T_8	0.85	1.53	0.52	0.34	1.36	1.85	0.28	0.40
T_9	0.16	0.88	0.05	0.06	0.20	0.94	0.08	0.30
T_{10}	0.45	1.03	0.08	0.07	0.54	1.10	0.18	0.33
T_{11}	0.35	1.51	0.09	0.18	0.44	1.85	0.16	0.43
T_{12}	0.54	1.29	0.12	0.14	0.66	1.43	0.21	0.40
T_{13}	0.42	1.04	0.10	0.11	0.52	1.15	0.14	0.35
T_{14}	0.69	1.67	0.11	0.14	0.80	1.89	0.27	0.43
T_{15}	0.64	1.56	0.10	0.16	0.74	1.71	0.23	0.41
T_{16}	0.65	1.32	0.15	0.08	0.80	1.40	0.23	0.34
SE±	0.01	0.01	0.02	0.02	0.047	0.03	0.01	0.01
CD 0.05	0.04	0.04	0.07	0.06	0.11	0.09	0.02	0.01

citrange (50.54 and 47.68 mg/100 g fw) and Cleopatra mandarin (49.05 and 47.46 mg 100/g fw) 120 and 210 DAI, respectively (Table 5). Thus, bacteria increased fungal hyphal network in rhizosphere, which in turn increased total phenols content in leaves, which might be one of the factors responsible for increased disease resistance found in mycorrhizal plants (Krishna and Bagyaraj, 1984).

Proline, an important osmolyte in leaf cytoplasm which protects and stabilise macro-molecules from oxidative damage, tend to decrease with passage of time in both Troyer citrange and Cleopatra mandarin seedlings, irrespective of treatments. However, control seedlings recorded lowest proline content in leaf cytoplasm in troyer citrange (97.92 and 62.40 µg/g fw) and Cleopatra mandarin (109.58 and 75.18 µg/g fw) at 120 and 210 DAI, respectively. The highest proline content was recorded in *G. intraradices* and PSB co-inoculated Troyer seedlings (255.20 and 197.38 µg/g fw) and Cleopatra seedlings (262.00 and 189.24 µg/g fw) 120 and 210 DAI, respectively (Table 5). Vivas *et al.* (2003) also reported highest proline content in lettuce seedlings co-inoculated with *G. intraradices* and phosphorus solubilizing bacteria *Bacillus* sp. The greater concentration of amino acids in AM plants may indicate a greater capability for osmotic adjustment through amino acid accumulation in these plants (Auge, 2001).

The total soluble sugars in leaves recorded declining trend with the passage of time in both the citrus plants (Table 5). It was highest in Troyer citrange seedlings co-inoculated with *G. intraradices* and *A. brasilense* 120 and 210 DAI (82.40 and 55.47%, respectively) followed by G. intraradices and PSB and *G. intraradices* and *Providencia* 120 DAI (80.40 and 80.44%) and 210 DAI (51.40 and 50.19%), compared to the control (46.40 and 29.56%) 120 and 210 DAI, respectively. In Cleopatra mandarin, highest total soluble sugars was observed in seedlings co-inoculated with *G. intraradices* and PSB (87.44 and 65.44%) at 120 and 210 DAI, respectively, followed by seedlings co-inoculated with *G. intraradices* and *A. brasilense* (87.02 and 63.21%) and *G. intraradices* and *Providencia* (86.29 and 59.40%) at 120 and 210 DAI, respectively, while the control seedlings significantly recorded lowest leaf total soluble sugars at both the growth stages (56.42 and 31.46%, respectively). The results are in harmony with Selvaraj *et al.* (2009). Phosphorus plays the most important role during the breakdown of carbohydrates and synthesis of polysaccharides. In particular, phosphorus is very effective in the synthesis of starch from glucose (Demir, 2004). Since, AM fungi increase the uptake of phosphorus; they may also increase the synthesis of carbon compounds.

Table 5. Leaf osmolyte contents of citrus seedlings as influenced by microbial inoculants

Treatment	Phenol (mg/100 g fw)		Proline (µg/g fw)		Total soluble sugars (%)	
	120 DAI	210 DAI	120 DAI	210 DAI	120 DAI	210 DAI
T_1	37.36	23.05	97.92	62.40	46.40	29.56
T_2	41.23	24.76	117.72	78.30	47.61	29.39
T_3	42.82	24.44	114.92	73.52	53.62	38.48
T_4	41.32	23.99	125.26	85.71	59.39	37.39
T_5	43.32	36.98	175.36	103.10	78.26	47.42
T_6	49.12	42.83	255.20	197.38	80.40	51.40
T_7	50.54	47.68	238.30	118.00	82.40	55.47
T_8	47.23	43.06	207.10	107.66	80.44	50.19
T_9	30.83	21.10	109.58	75.18	56.42	31.46
T_{10}	31.78	21.84	122.36	82.22	57.40	39.49
T_{11}	32.84	22.78	119.10	78.30	63.35	45.77
T_{12}	32.16	22.16	128.40	90.60	69.50	49.27
T_{13}	41.08	36.48	176.22	107.30	82.47	58.70
T_{14}	48.27	44.41	262.00	189.24	87.44	65.44
T_{15}	49.05	47.46	250.04	129.96	87.02	63.21
T_{16}	44.80	39.70	213.36	113.18	86.29	59.40
SE±	0.33	0.24	0.39	0.34	0.13	0.09
CD 0.05	0.97	0.69	1.15	1.01	0.39	0.26

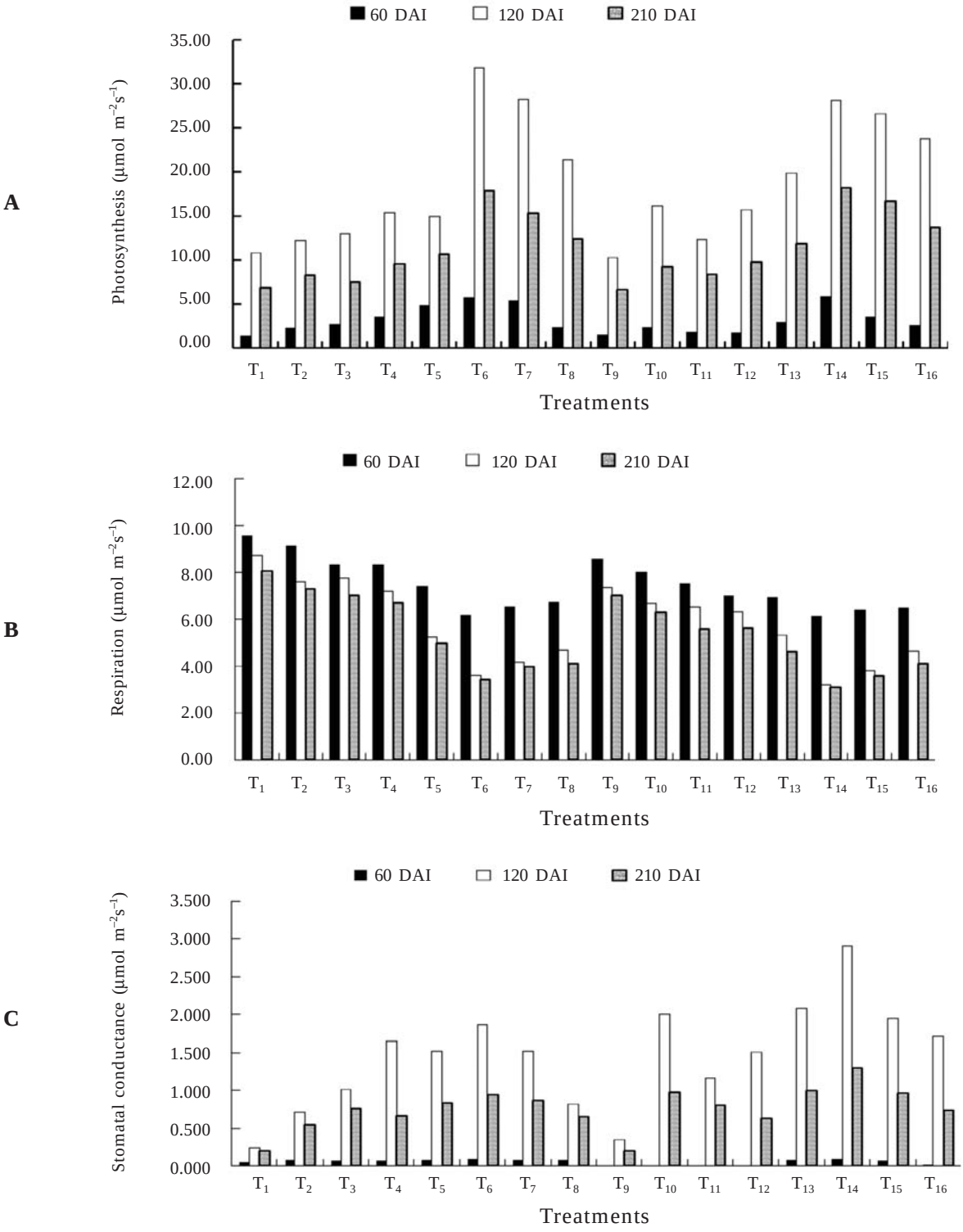

Fig. 1 A, B and C. Response of Troyer citrange and Cleopatra mandarin to AM fungi and MHB on (A). leaf photosynthetic rate, (B). Leaf respiration rate and (C). Leaf stomatal conductance.

Troyer citrange and Cleopatra mandarin seedlings exhibited significant response to co-inoculation of *G. intraradices* and PSB for photosynthetic rate at all the growth stages, as compared to other treatments, while the control seedlings exhibited the lowest photosynthetic rate. For Troyer citrange, *G. intraradices* and PSB had

significantly enhanced the photosynthetic rate at all the growth stages (5.76, 31.82 and 17.82 µmol ms^{-1}, respectively), which was non-significant with co-inoculation of *G. intraradices* and *A. brasilense* (5.48 µmol ms^{-1}) 60 DAI. Cleopatra mandarin also had positive response to *G. intraradices* and PSB for photosynthetic rate at all the growth stages (5.89, 28.19 and 18.19 µmol ms^{-1}, respectively). Irrespective of treatment, photosynthetic rate showed first increasing trend from 60 to 120 DAI and then showed decline 210 DAI in both Troyer citrange and Cleopatra mandarin (Fig. 1A).

The observation recorded for leaf respiration rate of Troyer citrange and Cleopatra mandarin seedlings, as influenced by AM fungi and MHB, revealed that Cleopatra seedlings had comparatively lesser leaf respiration rate than Troyer citrange at different growth stages and respiration rate declined with passage of time, irrespective of any treatment for both the citrus plants (Fig. 1B). At 60 DAI, Troyer citrange and Cleopatra mandarin seedlings co-inoculated with *G. intraradices* and PSB exhibited the lowest leaf respiration rate (6.17 and 6.11 µmol ms^{-1}, respectively), which was non-significant with seedlings co-inoculated with *G. intraradices* and *A. brasilense* and *G. intraradices* and *Providencia* (6.53 and 6.71 µmol ms^{-1}, respectively) in case of Troyer citrange and *G. intraradices* and *A. brasilense* and *G. intraradices* and *Providencia* (6.41 and 6.48 µmol ms^{-1}, respectively) in Cleopatra mandarin. At 120 DAI, G. intraradices + PSB and *G. intraradices* + *A. brasilense* recorded lowest respiration rate for Troyer citrange (3.62 and 4.17 µmol $m^{-2}s^{-1}$, respectively) and Cleopatra mandarin (3.19 and 3.81 µmol $m^{-2}s^{-1}$, respectively). At 210 DAI, co-inoculation of *G. intraradices* and PSB resulted in minimum leaf respiration rate in Troyer citrange and Cleopatra mandarin (3.40 and 3.10 µmol $m^{-2}s^{-1}$, respectively), which was non-significant with dual inoculated treatment of *G. intraradices* + *A. brasilense* (3.96 and 3.59 µmol $m^{-2}s^{-1}$) and *G. intraradices* + *Providencia* (4.10 and 4.11 µmol $m^{-2}s^{-1}$) in Troyer citrange and Cleopatra mandarin, respectively. Co-inoculation of Glomus intraradices and PSB had a synergistic effect for increased stomatal conductance 60, 120 and 210 DAI in both Troyer citrange (0.098, 1.866 and 0.938 µmol $m^{-2}s^{-1}$, respectively) and Cleopatra mandarin (0.095, 2.905 and 1.296 µmol $m^{-2}s^{-1}$, respectively), which was non-significant with co-inoculation of AMF mixed AMF strains and PSB (0.091 µmol $m^{-2}s^{-1}$) 60 DAI and co-inoculation of *G. intraradices* and *A. brasilense* (0.870 µmol $m^{-2}s^{-1}$) and single inoculation of *G. intraradices* (0.833 µmol $m^{-2}s^{-1}$) at 210 DAI in case of Troyer citrange (Fig. 1C). The increase in photosynthesis and stomatal conductance due to inoculation of AM fungi was also reported in chilli (Aguilera-Gomez *et al.* 1999) and maize (Boomsma and Vyn, 2008). Mycorrhiza can increase lateral root formation, which can indirectly increase cytokinin production and also enhances net photosynthetic rate (Davies *et al.*, 1996). These authors attributed the increase in the rate of photosynthesis to increased sink strength resulting from the presence of mycobiont. One of the most common explanations for reduced leaf respiration rate and increased stomatal conductance in microbial inoculated seedlings is the strongly increased absorbing surface caused by soil-growing hyphae combined with the fungal ability to take up water from soils with low water potential (Lehto and Zwiazek 2011).

The application of biofertilizers containing beneficial microorganisms gave a promoting effect on growth of citrus rootstocks and also improved the physiological status. The co- inoculation of AM fungi and MHB resulted in significant improvement in growth performance, chlorophyll content, carotenoids production and accumulation of osmolytes like total phenols. The *Glomus intraradices* and PSB had synergistic effect on higher growth of Troyer citrange and Cleopatra mandarin seedlings, which can be used as biofertilizers in propagation media.

REFERENCES

Aguilera-Gomez L, Davies F T J, Olalde-Portugal V, Duray S A and Phavaphutanon L. 1999. Influence of phosphorus and endomycorrhiza *(Glomus intraradices)* on gas exchange and plant growth of chile ancho pepper (*Capsicum annuum* L. cv. San Luis). *Photosynthetica* **36**(3): 441-9.

Auge R M. 2001. Water relations, drought and vesicular arbuscular mycorrhizal symbiosis. *Mycorrhiza* **11**: 3-42.

Barea J M and Jeffries P. 1995. Arbuscular mycorrhizas in sustainable soil-plant systems. *In: Mycorrhiza: Structure, Function, Molecular Biology and Biotechnology*, pp. 521-61 Varma A and Hock B (Eds). Springer, Berlin-Heidelberg, New York.

Barrs H D and Weatherley P E. 1962. A re-examination of the relative turgidity technique for estimating water deficits in leaves. *Australian Journal of Biological Sciences* **15**: 413-28.

Bashan Y and Levanony H. 1990. Current status of Azospirillum inoculation technology: *Azospirillum* as a challenge for agriculture. *Canadian Journal of Botany* **36**: 591-608.

Boomsma C R and Vyn T J. 2008. Maize drought tolerance: Potential improvements through arbuscular mycorrhizal symbiosis. *Field Crops Research* **108**: 14-31.

Dai M, Wang H X, Yin Y Y, Wu X, Wang M Y and Liu R J. 2008. Effects and mechanisms of interactions between arbuscular mycorrrhizal fungi and plant growth promoting rhizobacteria. *Acta Ecol. Sin*. **28**: 2854-60.

Davies F T, Svenson S E, Cole J C, Phavaphutanon L, Duray S A, Olalde-Portugal V, Meier C E and Bo S H. 1996. Non-

nutritional stress acclimation of mycorrhizal woody plants exposed to drought. *Tree Physiology* **16**: 985-93.

Demir S. 2004. Influence of arbuscular mycorrhiza on some physiological growth parameters of pepper. *Turkish Journal of Biology* **28**: 85-90.

Gadkar V, David-Schwartz R, Kunik T and Kapulnik Y. 2001. Arbuscular mycorrhizal fungal colonization and factors involved in host recognition. *Plant Physiology* **127**: 1493-9.

Graham J H and Syvertsen J P. 1985. Host determinants of mycorrhizal dependency of citrus rootstock seedlings. *New Phytology* **101**: 667-76.

Hiscox J D and Israelstam G F. 1979. A method for the extraction of chlorophyll from leaf tissue without maceration. *Canadian Journal of Botany* **57**: 1332-4.

Jaizme-Vega M C, Rodriguez-Romero A S and Pinero-Guerra M S. 2004. Potential use of rhizobacteria from the *Bacillus* genus to stimulate the plant growth of micropropagated banana. *Fruits* **59**: 83-90.

Kim K, Yim W, Trivedi P, Madhaiyan M, Hari P, Boruah D, Islam R, Lee G and Sa T. 2010. Synergistic effects of inoculating arbuscular mycorrhizal fungi and Methylobacterium oryzae strains on growth and nutrient uptake of red pepper (*Capsicum annuum* L.). *Plant Soil* **327**: 429-40.

Kobayashi DY and Crouch JA. 2009. Bacterial/fungal interactions: from pathogens to mutualistic endo-symbionts. *Annual Review of Phytopathology* **47**: 63-82.

Krishna, KR and Bagyaraj DJ. 1984. Phenols in mycorrhizal roots of *Arachis hypogea. Experientia* **40**: 85-86.

Lehto T, Zwiazek JJ. 2011. Ectomycorrhizas and water relations of trees: a reviews. *Mycorrhiza* **21**: 71-90.

Malik CP and Singh MB. 1980. *In: Plant Enzymology and Histoenzymology*, Kalyani Publishers, New Delhi, pp. 53.

Mamatha G, Bagyaraj DJ and Jaganath S. 2002. Inoculation of field-established mulberry and papaya with arbuscular mycorrhizal fungi and a mycorrhiza helper bacterium. *Mycorrhiza* **12**: 313-6.

Manoharan PT, Shanmugaiah V, Balasubramanian N, Gomathinayagam S, Sharma PM and Muthuchelian K. 2010. Influence of AM fungi on the growth and physiological status of *Erythrina variegata* Linn. grown under different water stress conditions. *European Journal of Soil Biology* **46**(2): 151-6.

Meyer J R and Linderman R G. 1986. Response of subterranean clover to co-inoculation with vesicular arbuscular mycorrhizal fungi and a plant growth promoting bacterium, *Pseudomonas putida. Soil Biology and Biochemistry* **18**: 185-90.

Mosse B. 1973. Advances in the study of vesicular-arbuscular mycorrhiza. *Ann. Rev. Phytopathol* **11**: 171-96.

Philips J M and Hayman D S. 1970. Improved procedure for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. *Trans. Br. Mycol. Soc.* **55**: 158-61.

Pivato B, Offre P, Marchelli S, Barbonaglia B, Mougel C, Lemanceau P and Berta G. 2009. Bacterial effects on arbuscular mycorrhizal fungi and mycorrhiza development as influenced by the bacteria, fungi, and host plant. *Mycorrhiza* **19**: 81-90.

Selvaraj T, Nisha M C and Rajeshkumar S. 2009. Effect of indigenous arbuscular mycorrhizal fungi on some growth parameters and phytochemical constituents of *Pogostemon patchouli* Pellet. *Maejo International Journal of Science and Technology* **3**(1): 222-34.

Vivas A, Marulanda A, Manuel J, Barea J and Azcon R. 2003. Influence of a Bacillus sp. on physiological activities of two arbuscular mycorrhizal fungi and on plant responses to PEG-induced drought stress. *Mycorrhiza* **13**: 249-56.

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Effect of foliar application of chemicals on fruit retention, yield and quality of mango *(Mangifera indica)* cv. Kesar

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ABSTRACT

The field experiment was conducted at Agricultural Experimental Station, Navsari Agricultural University, Paria, District-Valsad, Gujarat, during 2013-14. The experiment included seven treatments consisting of tricontanol @ 500, 750 and 1000 mg/litre, humic acid 0.5, 1.0, and 1.5% and water spray as the control. The experiment was laid out in a randomized block design with four replications. Trees were sprayed three times, *viz*. at flowering, pea size and marble stage of fruits. Among different treatments, tricontanol @ 750 mg/litre enhanced fruit setting at pea and marble stages per panicle; fruit retention at harvesting stage, minimize fruit drop at harvesting stage and number of fruits/tree, while, humic acid @ 1.5%was found effective for fruit yield, physical and quality parameters. Similarly, foliar application of humic acid @ 1.5% gave higher benefit: cost ratio.

Key Words: Tricontanol, Humic acid, Foliar application, Quality, Fruit retention

Mango (*Mangifera indica* L.) belongs to family Anacardiaceae. India is the largest producer and exporter of mango in the world, producing around 17.6 million tonnes with a productivity of 7.13 tonnes/ha. Among different varieties, Kesar having high yield potential, is almost regular-bearer and mid-season variety, It has good consumer acceptance, attractive shape, size, saffron pulp and very good keeping quality. Flowering in mango is preceded by differentiation of flower buds in shoots. The period of differentiation is October - December in Gujarat. In mango, heavy fruit drop is an important factor, contributing to low fruit yield and sometimes only 0.1% of fruits reach up to the maturity. The maintenance of fruit quality is critical, while employing any new technology for increasing production and quality. Thus, fruit setting in mango is crucial event which greatly influence the ultimate fruit yield.

Humic acids (HAs) are main fractions of humic substances (HS) and the most active components of soil and compost organic matter. They exert indirect and direct effect on plants (Chen *et al.*, 2004) and this action of HS is dose dependent and high concentrations of HS are inhibitory for nutrient accumulation (Chen and Avaid, 1990). Tricontanol is a natural plant growth regulator found in epicuticular waxes. It is used to enhance fruit production. Tricontanol can be used for improvement in growth, yield, photosynthesis, protein synthesis, uptake of water and nutrient in various crops (Neem and Khan, 2011). Hence an experiment was conducting to find out the effect of foliar application of chemicals on fruit retention, yield and qualtity of mango.

MATERIALS AND METHODS

The experiment was conducted at the Agriculture Experimental Station, Navsari Agricultural University, Paria, District-Valsad, situated at an elevation of 16 m above mean sea-level on latitude 23°35' North and longitude of 72°35' East. Twelve years old uniform mango trees of Kesar planted at 5m × 5m distance were selected. The experiment was laid out in a randomized block design with 7 treatments and 4 replications, including tricontanol @ 500, 750 and 1000 mg/litre, humic acid 0.5, 1.0, and 1.5% and water spray as the control. Foliar application of chemicals was done at flowering, pea and marble-sized stage of fruit development. The fruits were harvested in June when they were fully mature. The parameters like fruiting

behaviour, yield and physical and chemical characters were observed and statistically analysed.

RESULTS AND DISCUSSION

There were maximum fruit setting at pea and marble-sized stage of fruits per panicle, fruit retention per panicle at harvesting stage with minimum fruit drop at harvesting stage and number of fruits was observed when trees were sprayed with tricontanol @ 750 mg/litre. This might be due to application of tricontanol, attributed to more efficient utilization of food for reproductive growth, flowering and fruit setting, higher photosynthetic efficiency and enhanced source to sink relationship in plants, increased uptake of nutrients and water, reduced transpiration and respiration, enhanced translocation and accumulation of sugar and other metabolites (Chaudhary *et al.*, 2006). The maximum fruit yield (kg/tree) was observed with higher concentration of humic acid @ 1.5% (Table 1). The maximum yield in mango is cumulative effect of fruit length and breadth as well as the average weight of fruits. The positive effect of humic acid in yield of mango may due to enhanced uptake of mineral nutrients and increased cation exchange in soil.

The mango trees sprayed with higher concentration of humic acid (@ 1.5%) resulted in maximum average fruit weight, fruit length, fruit volume and shelf-life (Table 2). The application of organic acids increasing fruit weight by activating hormones like auxin and cytokinin, resulting in high weight of fruits, foliar application of humic acid increased fruit length by increasing cell division and enlargement and resulted in more length of fruits (Mahmoudi *et al.*, 2013). Humic acid stimulated plant enzymes and increased their production. It is known to thicken the cell wall in fruit and prolong the storage as well as or shelf life of fruits (Chen *et al.*, 2004).

Increasing trend of humic acid @ 1.5% was found to be most effective in terms of total sugar and ascorbic acid (mg/100 g), while humic acid @ 1.0% gave maximum total soluble solids (°Brix) and overall acceptability (Table 3). During fruit maturation and at harvesting, humic acids stimulated pigment accumulation, resulting in greener leaves with greater photosynthetic efficiency. In this period, because fruits are strongest sink for carbohydrates and nutrients roots may consequently become less efficient in absorbing mineral nutrients (Hancock 1999). It can be hypothesized that foliar application of humic acids had mostly positive effects on nutrient availability. This new favourable nutritional status, induced by repeated foliar applications, could be the indirect cause of the improvement of fruit characteristics. As a result, fruit quality was significantly increased (Neri *et al.* 2002). Thus, with the application of tricontanol and humic acid, benefit : cost ratio (BCR) was maximum (3.21) under foliar spray of humic acid (@ 1.5%) with the net income of ₹ 3,92,500, followed by tricontanol @ 750

Table 1. Effect of chemicals on fruiting parameters in mango cv. Kesar

Treatment	Fruit setting at pea stage per panicle	Fruit setting at marble stage per panicle	Fruit retention at harvesting stage	Fruit drop (%) at harvesting stage	Number of fruits	Fruit yield (kg/tree)	Fruit yield (tonnes/ha)
T_1 - Tricontanol 500 ppm	13.29	53.21 (46.84)	1.98	85.08 (67.33)	194.75	47.13	18.85
T_2 - Tricontanol 750 ppm	14.30	56.34 (48.64)	2.33	83.62 (66.16)	201.25	49.44	19.78
T_3 - Tricontanol 1000 ppm	12.56	50.58 (45.33)	1.78	85.69 (67.89)	181.0	43.58	17.43
T_4 - Humic Acid 0.5%	12.45	50.27 (45.15)	1.54	87.57 (69.37)	178.0	43.76	17.50
T_5 - Humic Acid 1.0%	12.37	50.47 (45.26)	1.53	87.63 (69.41)	176.7	47.00	18.80
T_6 - Humic Acid 1.5%	12.97	51.31 (45.87)	1.93	85.06 (67.29)	185.0	51.45	20.58
T_7 - Control (Water spray)	10.88	44.93 (42.06)	0.80	92.39 (74.57)	168.0	31.41	12.56
SEm ±	0.40	1.14	0.13	1.35	6.47	2.02	0.81
CD (5%)	1.19	3.34	0.38	4.019	19.23	5.99	2.39
CV (%)	6.31	4.93	15.00	3.92	7.05	8.99	8.99

*Values in parentheses are arc sine transformed value

Table 2. Effect of chemicals on physical parameters of mango cv. Kesar

Treatment	Fruit weight (g)	Fruit length (cm)	Fruit breadth (cm)	Fruit volume (ml)	Pulp: peel ratio	Shelf life (days)
T_1 - Tricontanol 500 ppm	242.50	11.23	6.94	237.73	11.43	15.75
T_2 - Tricontanol 750 ppm	245.75	11.35	7.08	243.60	11.57	16.50
T_3 - Tricontanol 1000 ppm	240.75	11.59	6.95	236.77	11.36	16.00
T_4 - Humic Acid 0.5%	245.75	11.37	7.19	240.08	11.51	16.75
T_5 - Humic Acid 1.0%	266.00	12.16	7.21	255.58	12.07	17.50
T_6 - Humic Acid 1.5%	278.00	12.18	7.20	270.07	11.92	18.25
T_7 - Control (Water spray)	186.75	10.36	6.53	181.02	10.68	15.50
SEm ±	7.44	0.37	0.18	8.04	0.36	0.57
CD (5%)	22.11	1.10	NS	23.89	NS	1.68
CV (%)	6.11	6.45	5.09	6.76	6.21	6.83

Table 3. Effect of chemicals on quality parameters of mango cv. Kesar

Treatment	Fruit firmness (kg/cm^2)	TSS (°Brix)	Acidity (%)	Reducing sugar (%)	Non-reducing sugar (%)	Total sugar (%)	Ascorbic acid (mg/100g)	Overall accepta-bility
T_1 - Tricontanol 500 ppm	3.82	16.31	0.26	3.26	8.16	11.42	15.12	7.15
T_2 - Tricontanol 750 ppm	3.87	16.52	0.26	3.31	8.08	11.39	14.53	7.23
T_3 - Tricontanol 1000 ppm	3.74	16.73	0.27	3.46	8.30	11.76	14.54	7.39
T_4 - Humic Acid 0.5%	3.74	16.94	0.26	3.24	8.04	11.27	15.31	7.10
T_5 - Humic Acid 1.0%	3.90	18.03	0.25	3.59	7.90	11.48	15.72	7.98
T_6 - Humic Acid 1.5%	3.88	17.38	0.26	3.72	8.65	12.36	16.35	7.95
T_7 - Control (Water spray)	3.28	15.58	0.24	3.17	7.95	11.12	14.18	6.70
SEm ±	0.13	0.45	0.01	0.10	0.19	0.24	0.47	0.19
CD (5%)	NS	1.35	NS	0.30	NS	0.71	1.38	0.57
CV (%)	07.16	5.40	6.72	5.98	4.65	4.12	6.16	5.25

mg/litre having a net gain of ₹ 3,74,000 with (benifit : cost ratio) of 3.10.

REFERENCES

Chaudhary BR, Sharma, MD, Shakya, SM and Gautam, DM. 2006. Effect of plant growth regulators on growth, yield and quality of chilli (*Capcicum annuum* L.) at Rampur, Chitwan. *J. Inst. Agric. Anim.* Sci. **27**: 65-68.

Chen Y and Avaid T. 1990. Effect of humic substances on plant growth. *In:* MacCarthy P.; Clapp C. E.; Malcolm R. L.; Bloom P. R. (Eds): *Humic Substances in Soil and Crop Sciences: Selected Readings,* pp.161-86. American Society of Agronomy, Madison, WI, USA.

Chen Y, De Nobili. M and Avaid, T. 2004. Stimulatory effects of humic substances on plant growth. Magdoff F., Eeil R.R. (Eds.): *Soil Organic Matter in Sustainable Agriculture*, pp. 103-29. Magdoff F, and Wiel RR (Eds) CRC Press, New York, USA.

Hancock, JF. 1999. *Strawberry*. University Press, Cambridge, pp 237.

Mahmoudi M, Samavat S, Mostafavi M, Khalighi A and Herati, A. (2013).The effects of proline and humic acid on quantitative properties of kiwi fruit. *Int. Research Journal of Applied and Basic Sci.* **6**(8): 1117-1119.

Neem M and Khan M A. 2011. Tricontanol a potent plant growth regulator in agriculture. *Journal of Plant Interactions* **7**(2): 138-44.

Neri E M, Lodolini G, Savini, P, Sabbatini, G and Zucconi, F. 2002. Foliar application of humic acids on strawberry (cvh Onda). *Acta Hort.* **594**: 297-302.

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Genetic diversity analysis in apple *(Malus domestica)* using yield and yield associating traits as per DUS guidelines

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ABSTRACT

The variability studies undertaken on 56 cultivars of apple (*Malus domestica* Borkh) revealed that lowest st deviation (1.77) was observed for TSS, whereas highest (43.28) for fruit weight. Similarly cv was noted high (57.22%) in yield. The clustering pattern showed that cluster I and cluster IV consist of highest number of cultivars (17 and 12 respectively). Inter cluster distance showed highest(6.09) between cluster IV and Cluster V and lowest (1.98) between cluster I and cluster VI. The cluster IV registered highest mean fruit length, yield/tree and per hectare yield and cluster V showed highest mean for characters days taken to maturity after full blooming. Total principal components for PC 1, PC2 and PC 3 contributed more to total variance (44.75, 19.45 and 15.91% respectively).

Key Words: Diversity analysis, Cultivars, Cluster analysis, Fruit quality

Apple (*Malus domestica* Borkh) is considered as the king of temperate fruits. The virtue of its shelf-life makes it easy to store at ambient temperature even for a month. Most of the apple acreage is at the hill slopes except in Kashmir valley and a limited area in Kullu Valley. Unlike other fruits, apple can be grown successfully under sod, provided additional nitrogen required for growth of grass is provided. In India, apple is mainly grown in Jammu and Kashmir, Himachal Pradesh, Uttarakhand and Arunachal Pradesh and the Nilgiri hills. Total production of apple in India is 24.97 lakh tonnes. Though Jammu and Kashmir is leading in per hectare productivity (10.2 tonnes/ha), followed by Himachal Pradesh (6.9 tonnes/ha) and Uttarakhand (2.6 tonnes/ha) which is very low as compared to 60-84 tonnes/ha in advanced countries in world. More than 10,000 apple varieties are grown. However, only a few are commercially grown.

In our country introduction of new cultivars initiated in 1870 by the European settlers and missionaries, Capt Lee started orcharding in the Kullu Valley and Coutis in Shimla area. S.C. Kashmir was first state to visualize the great potential. Agriculture Mr. M Pychard, a Frenchman introduced many varieties from his country. Further NBPGR and other state government procured a large number of apple varieties and rootstocks for evaluation and popularization time to time. At CITH, Srinagar, a large number of apple varieties were introduced from France, UK, USA, and other European countries. Some of them are quite popular and adapted well. Since, Institute has more than 340 varieties and 15 rootstocks, therefore evaluation of varieties and analysis of genetic divergence were carried out on the basis of DUS test guidelines.

MATERIALS AND METHODS

The studies were undertaken at Central Institute of Temperate Horticulture, Srinagar, during 2009-2011. Apple varieties obtained through Introduction by the NBPGR and other Govermnent agencies were planted during 2002-03 at 5 m × 5 m on seedling apple root-stock. The fruit quality data were recorded at full maturity of fruits. For recording the days taken to fruit maturity from full bloom, date of full bloom (more than 75% bloom) was noted as reference date. The fruit yield after harvesting from individual tree, total fruit weight were worked out from three trees and average was worked out in kg/tree. Whereas yield per hectare was calculated by multiplying the yield per tree with total number of trees/ha. For recording other fruit related observations 15 fruits were randomly collected from all directions of trees. The fruit weight was recorded with the help of digital balance, length, breadth, with the help of digital Vernier caliper (Mitutoyo Inc., Japan).

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Table 1. List of apple cultivars studied

Cultivar	Indigenous/exotic	Cultivars	Indigenous/exotic
Vista Bella	Exotic	Green Sleeves	Exotic
Starkrimson	Exotic	Rome Beauty	Exotic
Mollies Delicious	Exotic	Well Spur	Exotic
Cooper-IV	Exotic	Parlin's Beauty	Exotic
American Apirouge	Exotic	King Hasicus	Exotic
Silver Spur	Exotic	CITH-Apple-13	Indigenous
Golden Delicious	Exotic	CITH-Apple-09	Indigenous
Firdous	Indigenous	CITH-Apple-05	Indigenous
Red Fuji	Exotic	CITH-Apple-04	Indigenous
Gold Spur	Exotic	Starkrimson Gold	Exotic
Red Chief	Exotic	Jonica	Exotic
Shireen	Indigenous	Mayan	Exotic
Oregon spur	Exotic	Tallisare	Exotic
Starking Delicious	Exotic	Lemon Guard	Exotic
Red Gold	Exotic	Wilson Red June	Exotic
Red Delicious	Exotic	Vance Delicious	Exotic
Top Red	Exotic	Pink Lady	Exotic
Tydemans Early	Exotic	Check Ambri	Exotic
Benoni	Exotic	Wealthy Apple	Exotic
Hardiman	Exotic	MC Spur	Exotic
Laxtune Forfune	Exotic	Florina	Exotic
Prima	Exotic	Anna	Exotic
Summer Red	Exotic	Black Ben Davis	Exotic
Rich-a-Red	Exotic	Star Summer Gold	Exotic
Scarlet Gala	Exotic	Red Spur	Exotic
Michal	Exotic	Winter Commercial	Exotic
June Eating	Exotic	Skyline Supreme	Exotic
Lal Ambri	Indigenous (Hybrid)		
Ambri	Exotic		

The TSS was recorded with the help of refractometer and expressed into degree Brix. Fruit firmess was recorded with penetrometer and pressure expressed the reading unit (lb/cm^2).The experiment was laid out in a randomized block design replicated thrice with 2 plants per replication.Genetic diversity was studied using Mahalanobis (1936), generalized distance (D^2) extended by Rao (1952), acreage intra and inter cluster

Table 2. Variability in fruit yield and quality attributes in 56 apple cultivars

Character	Mean	St deviation	Minimum	Maximum	CV (%)
Fruit weight (g)	150.8	43.28	44.47	230.70	28.70
Fruit length (mm)	61.4	8.85	37.39	78.37	14.41
Fruit diameter (mm)	69.65	8.14	47.48	84.60	11.68
TSS (°Brix)	15.39	1.77	11.20	20.24	11.50
Fruit firmness (Lb/cm^2)	46.28	7.01	29.80	61.51	15.14
Yield (Kg/tree)	21.01	12.016	7.32	58.16	57.19
Yield (t/ha)	13.14	7.52	4.57	36.35	57.22
Days taken to maturity after full bloom (days)	141.41	24.94	86.0	171.00	17.63

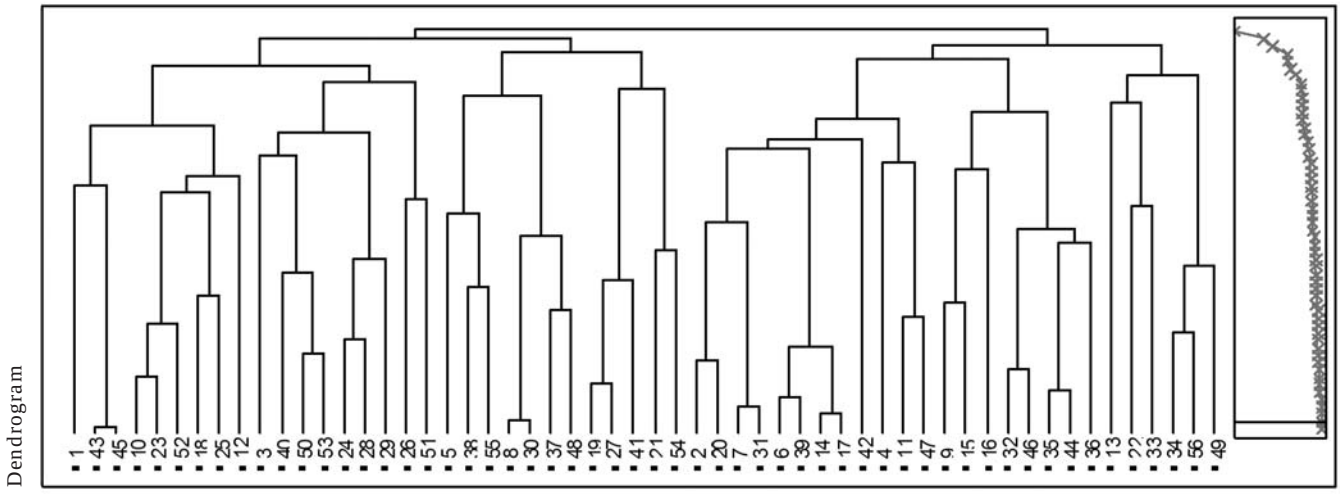

Fig. 1. Dendrogram showing of 56 apple cultivars.

distance using the formulae suggested by Singh and Choudhary (1985). Clustering of cultivars was done according to Tocher's Method (Rao 1952). The two-year data were pooled, analyzed using SAS 9.3 software for conducting the D^2 analysis Ward methods on standard data was used (Table 1).

RESULTS AND DISCUSSION

The variability in each trait was expressed by standard deviation and coefficient of variation. The lowest valve of standard deviation (1.77) was noted in TSS and fruit firmness, while it was highest (43.28) in fruit weight and days taken to maturity (24.94). The coefficient of variation was highest (57.22%) in yield (tonnes / ha), and yield / plant (57.19%), whereas it was lowest for TSS (11.50%) and fruit diameter (11.68%) (Table 2). Yield and quality attribute analysis based on different characters showed high polymorphism with 56 apple cultivars into six distinct clusters (Table 3). Cluster I consisted of maximum 17 cultivars cluster iv (12) cultivars, cluster v, 8, and cluster iii and vi 5, and 6 cultivars respectively.

The highest inter-cluster distance (6.09) was noted between cluster iv followed by (5.57) between cluster 1 and cluster iv. Lowest distance (1.98) recorded between cluster ii and cluster vi and (2.57) between clusterII and clusterIII. The highest inter-cluster distance indicated that cultivars belonging to cluster IV was far away from cluster V, whereas lowest cluster distance revealed that cluster II and cluster VI and cluster II and cluster III are genetically close and similarity in most of the traits (Table 3). Since cluster showed highest inter cluster distances selection of parents from such clusters for hybridization programme would help to develop novel hybrids.

Principal component analysis (PCA) reflects the

Table 3. Grouping of apple cultivars

Cluster number	Number of genotypes	Accession numbers
I	18	Vista Bella, Mayan, Lemon Guard, Florina, CITH-Apple-04, Wealthy Apple, Anna, Mc Spur,Richa Red, Lal Ambri, Ambri, Michel, Gold Spur, Summer Red, Tydeman's Early Worcestor, Scarlet Gala, Shireen
II	7	American Apirouge, Firdous, Winter Commercial, CITH-Apple 13, CITH-Apple-09, Pink Leady, Star Gold
III	5	Black Ben Davis, Starkrimson Gold, Benoni, Laxtone Fortune, June Eating
IV	12	Starkrimson, Hardimann, Golden Delicious, Silver Spur, Starking Delicious, Top Red, CITH-Apple-05, Jonika, Vance Delicious, Skyline Supreme, Cooper IV, Red Chief
V	8	Red Fuji, Red Gold, Red Delicious, Parlins Beauty, King Hasicus, Tallisare, Wilson Red June, Green Sleeves
VI	6	Oregon Spur, Prima, Rome Beauty, Red Spur, Check Ambri

Table 4. Correlation coefficient among different apple characters

Character	Fruit weight (g)	Fruit length (mm)	Fruit diameter (mm)	TSS (°Brix)	Fruit firmness (Lb/cm^2)	Yield (kg/tree)	Yield (tonnes/ha)	Days taken to maturity after full bloom (days)
Fruit weight (g)	1.00	0.88*	0.909	-0.129	-0.077	0.3402	0.3401	0.274
Fruit length (mm)		1.00	0.820	-0.0305	-0.209	0.322	0.3229	0.3794
Fruit diameter (mm)				1.00	-0.0681	-0.231	0.301	0.324
TSS (°Brix)				1.00	-0.260	-0.14	-0.139	0.029
Fruit firmness (Lb/cm^2) Yield (kg/tree)						1.00	1.00	0.401
Yield (tonnes/ha)							1.00	0.402
Days taken to maturity after full bloom (days)								1.00

Table 5. Inter and intra cluster distance among six clusters

Cluster	I	II	III	IV	V	VI
I	0	4.27	3.25	5.57	3.05	3.86
II		0	2.57	2.08	4.52	1.98
III			0	3.57	4.56	3.18
IV				0	6.09	3.28
V					0	3.28
VI						0

importance of largest contributor to the total variation at the each axis of differentiation. The eign values are often used to determine how many factors to retain the sum of eign values are usually equal to number of variables there principal components PC 1 to PC 3 which are extracted from the original data and having latent roots greater than one accounting nearly 80.12% of total variations out of total principal components returned, PC 1, PC 2, and PC 3, with value of 44.75, 19.45 and15.91% respectively contributed more to the total variations. According to Chahal and Gosal (2002), characters with largest absolute value closer to unity within first PC influence the clustering more than those with lower absolute value closer to Zero.

Accordingly, first principal component loading from fruit weight, fruit length to maturity, where as TSS. and fruit firmness had negative loading the positive and negative loading shows the presence of positive and negative correlation trends between components and the variables. Hence, these characters which lead high positively or negatively contributed more to genetic diversity in traits which contributed more positively to principal component 2 are fruit weight, fruit length, diameter and TSS, whereas fruit yield and days taken to maturity contributed negatively to diversity.

In the PC 3, fruit weight, fruit firmness and yield contributed positively, while rest factors had negative load (Table 5). The diversity in cultivars was also evident by the amount of variation among cluster means for different traits (Table 6). The cluster II showed highest cluster means for fruit weight, fruit diameter and fruit firmness. The cluster IV registered highest means of fruit length, yield/tree and yield/hectare where as cluster V showed highest mean for days taken to maturity after full bloom. That these clusters had cultivars with the respective desirable traits hence, for improvement of traits the parents should be selected from their respective clusters showing highest cluster maans.

Some highly significant pearson correlations were observed between characters measured on different quality traits of apple (Table 6). Fruit weight had high positive creation with fruit length and fruit diameter, whereas it is negatively correlated with the TSS and fruit firmness. Higher standrad deviation and coefficient variations for fruit weight, fruit firmness and days taken to maturity showed high degree of diversity among evaluated cultivars. The higher inter cluster distance than the mean intera cluster distance confirms wide genetic diversity among cultivars of different groups than those of the same cluster Srivastava KK *et al.* (2007) also obtained high inter cluster distance in apricot and Dwivedi and Mitra, (1996) in litchi the minimum intra and inter cluster distance indicates the

Table 6. Latent vector for 8 traits of 56 apple varieties

Character	Eign vector		
	PRIN 1	PRIN 2	PRIN 3
Fruit weight (g)	0.44754	0.37246	0.10630
Fruit length (mm)	0.44623	0.33828	-0.04085
Fruit diameter (mm)	0.444192	0.36513	-0.02315
TSS (°Brix)	-0.05964	0.03520	-0.75070
Fruit firmness (Lb/cm^2)	-0.17027	0.15203	0.62511
Yield (kg/tree)	0.37773	-0.52841	0.10009
Yield (tonnes/ha)	0.37782	-0.52847	0.09929
Days taken to maturity after full bloom (days)	0.29549	-0.17515	-0.11091
Eign values	3.58	1.55	1.27
T. variance	44.75	19.45	15.91
Cumulative (%) variance	44.75	64.20	80.12

Table 7. Mean performance of cluster for different fruit quality traits in 56 apple cultivars

Character	Fruit weight (g)	Fruit length (mm)	Fruit diameter (mm)	TSS (°Brix)	Fruit firmness (Lb/cm^2)	Yield (kg/tree)	Yield (tonnes/ha)	Days taken to maturity after full bloom (days)
I	184.96	72.65	77.25	18.8	33.7	26.8	16.75	161
II	198.56	67.61	79.87	15.03	56.33	7.32	4.57	167
III	159.81	65.96	68.58	16.56	41.38	8.46	5.28	96.0
IV	195.8	75.6	77.9	16.0	35.80	58.16	36.35	157
V	127.7	58.61	65.14	12.46	56.0	36.77	22.98	171
VI	44.47	37.39	47.48	15.74	49.38	16.19	10.11	120

close genetic relationship among the studied cultivars the cultivars having high D^2 value can be used for inter varietal hybridization programme cluster means reveal the inner diversity in the material under study. The traits contributing maximum towards the D^2 values should be given priority in choosing a cluster for selection and choice of parents for inter varietal hybridization.

REFERENCES

Chahal GS, Gosal SS. 2002. *Principles and Procedures of Plant Breeding; Biotechnology and Conventional Approaches*. Narosa Publishing House, New Delhi.

SAS. Institute 2011. SAS enterprise guide, *version 9.2*, SAS Inst. Cary, NC, USA.

Srivastava KK, Sharma MK and Sundouri AS. 2007. Estimation of genetic divergence in apricot (*Prunus armeniaca* L.) *Journal of Plant Genetic Resources* **20**(03): 34-236.

Dwivedi AK and Mitra SK. 1996. Divergence analysis of Litchi *(Litchi chinesis)* Sonn cultivars growriun West Bengal. *Indian Journal of Genetics*, **56**: 486-89.

Mahalanobis PC. 1936. On the generalized distance in statistics proe. *National Academy of Sciences.* (India) **2**: 49-55.

Rao CR. 1952. *Advanced Statistical Methods in Biometric Research*, pp. 357-69. John Wiley and Sons, New York.

Singh RK, Choudhary BD. 1985. *Biometrical Methods in Quantitative Genetic Analysis*. Kalyani Publishers, New Delhi, pp. 318.

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Response of indole butyric acid (IBA) on sprouting and shoot growth of stem cuttings of star gooseberry *(Phyllanthus acidus)*

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ABSTRACT

The experiment was conducted to find out the response of IBA on sprouting and shoot growth of stem cuttings of star gooseberry (*Phyllanthes acidus* Skeels) during 2013-14, in polyhouse conditions at Regional Horticultural Research Station, Navsari Agricultural University, Navsari. A experiment was laid out in completely randomized design with factorial concept (FRBD) having 14 treatment combinations, comprising with two factors: (1) types of cutting (hardwood cutting and top cutting) and (2) different concentration of IBA (0, 500, 1000, 1500, 2000, 2500 and 3000 mg/litre) and repeated trice. Among different types of cutting and IBA concentration, hardwood cutting and 2000 mg/litre were individually as well as in their combination most beneficial for early sprouting and maximum sprouting percentage. Similar trend was observed on the growth parameters of shoot like number of shoots, length of longest shoot, fresh and dry biomass of shoot and survival percentage.

Key Words: IBA, Sprouting, Shoot growth, Stem cutting Polyhouse

Star gooseberry *(Phyllanthus acidus* Skeels*)* belongs to family Phyllanthaceae. It has been widely distributed. Its fruit is sour, edible, astringent berry, which is eaten raw, cooked or pickled or is made into jam, chutney, preserve and jelly. The fruits are nutritive and are a rich source of vitamin C and minerals. They are divided into 6-8 segments and single seeded. It is rich in pectin, vitamin C and acid contents. The fruits are non-climacteric, hence attain no respiratory peak at maturity. They have good shelf-life and can be kept for 8-10 days without any deterioration in their quality. It is a heavy bearer and has short fruit maturity period besides being tolerant to biotic and abiotic stresses.

Any living vegetative plant tissue, cambium, epidermis, parenchyma of bark, *etc.* can form roots if there are fulfilled appropriate environmental conditions and if there is a certain level of hormonal contents (growth regulators). The high hormone concentrations provoke the cambium and pericycle cell division from where the process of root formation starts. Roots originated from vegetative plant's parts under the hormone effect are similar to roots formed naturally. For cuttings rooting acceleration can be used growth regulators such are Indole-3-Butyric Acid (IBA) and Naphthalene Acetic Acid (NAA). A higher concentration of rooting hormones is used for woody and lignified parts as well as for those that hardly root, while very high concentrations of rooting hormone inhibit the rooting process.

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MATERIALS AND METHODS

The experiment was conducted during 2013-14, under polyhouse conditions at Regional Horticultural Research Station, ASPEE College of Horticulture and Forestry, Navsari Agricultural University, Navsari, Gujarat, India. An experiment was laid out in completely randomized design with factorial concept (FRBD) having 14 treatment combinations, comprising with two factor: (1) types of cutting (hardwood cutting and top cutting) and (2) different concentration of IBA (0, 500, 1000, 1500, 2000, 2500 and 3000 mg/litre) and repeated rice. The cuttings were taken from healthy mother plants of 10-15 years old, cultivar "Local" in March. The cuttings (15-20 cm long) were generally taken from the matured branches. The top 15-20 cm long shoots were used as top cuttings and the remaining portion was used as hardwood cuttings.

RESULTS AND DISCUSSION

Two types of cuttings (hardwood cuttings and top cuttings) and different IBA concentrations (0, 500, 1000, 1500, 2000, 2500 and 3000 mg/litre) were used. The interaction effect between types of cuttings and IBA concentrations were significant in all the characters the best result was found in hardwood cuttings with IBA 2000 mg/litre (C_1I_5) treatment.

The minimum number of days (7.00 days) required for sprouting were found in C_1I_5 treatment which was at par with C_1I_6 treatment and maximum number of days for sprouting were 18 days in the control (C_2I_1) (Table 1). It might be due to level of maturity of wood, the chemical composition of wood from base to tip and presence of active buds on stem (Kochhar *et al.*, 2008).

Hardwood cuttings having high amount of stored carbohydrate and low to moderate amount of nitrogen which was utilized by cutting to produce shoot system with the help of IBA by hydrolysis, mobilization and utilization of nutritional reserves in region of shoot formation (Kraus and Kraybill, 1918).

Different shoot characteristics, *viz.* number of shoots/cuttings, longest shoot and fresh and dry biomass of cuttings were significantly higher in hardwood cuttings when they were treated with IBA at 2000 mg/litre.

Maximum sprouting percentage (98.33) was found in C_1I_5 treatment and minimum sprouting percentage (26.62) was noted in C_2I_1 (Table 2) because planting material for hardwood cutting was taken from the base and middle portion of the branch and minimum shoots were recorded in the top cutting because of presence of apical portion which resulted in creation of apical dominance. Exogenous application of IBA increased the endogenous level of auxin. Mobilization and utilization of stored carbohydrates due to influence of auxin increased the number of sprouts (Severino *et al.*, 2011).

Thus, portion of branch utilized and mobilization and utilization of carbohydrate due to increase in indigenous auxin had significant effect on maximum number of shoots recorded. The same reasons were recorded by Singh *et al.* (2014) in mulberry.

Maximum shoot length (27.10 cm) was observed in C_1I_5 treatment (Table 3), while, minimum (6.47 cm) shoot length was found in C_2I_1 which was at par with C_1I_1 (6.73 cm).

The number of leaves, leaf area and length of longest shoot were maximum in hardwood cuttings. It

Table 1. Effect of type of cuttings and IBA concentration on number of days required for sprouting.

IBA/cutting	(C_1)	(C_2)	Mean
I_1 - 0 ppm	16.67	18.00	17.33
I_2 - 500 ppm	11.33	13.00	12.17
I_3 - 1000 ppm	11.00	11.00	11.00
I_4 - 1500 ppm	10.00	14.00	12.00
I_5 - 2000 ppm	7.00	9.00	8.00
I_6 - 2500 ppm	7.67	10.33	9.00
I_7 - 3000 ppm	8.00	11.00	9.50
Mean	10.24	12.33	
	S.Em±	**CD at 5 %**	**CV %**
C	0.32	0.92	
I	0.60	1.73	4.32
C * I	0.28	0.82	

Table 2. Effect of type of cuttings and IBA concentrations on sprouting percentage.

IBA/cutting	(C_1)	(C_2)	Mean
I_1 - 0 ppm	38.33	26.67	32.50
I_2 - 500 ppm	76.67	75.00	75.83
I_3 - 1000 ppm	81.67	76.67	79.17
I_4 - 1500 ppm	86.67	85.00	85.83
I_5 - 2000 ppm	98.33	88.33	93.33
I_6 - 2500 ppm	86.67	83.33	85.00
I_7 - 3000 ppm	90.00	83.33	86.67
Mean	79.76	74.05	
	S.Em±	**CD at 5 %**	**CV %**
C	1.67	4.85	
I	3.13	9.09	3.32
C * I	1.48	4.28	

Table 3. Effect of type of cuttings and IBA concentrations on longest shoot length (cm) (120 DAP).

IBA/cutting	(C_1)	(C_2)	Mean
I_1 - 0 ppm	6.73	6.47	6.60
I_2 - 500 ppm	14.33	11.00	12.67
I_3 - 1000 ppm	19.00	13.70	16.35
I_4 - 1500 ppm	21.00	15.57	18.28
I_5 - 2000 ppm	27.10	20.10	23.60
I_6 - 2500 ppm	22.97	17.13	20.05
I_7 - 3000 ppm	18.19	14.95	16.52
Mean	18.47	14.13	
	S.Em±	**CD at 5 %**	**CV %**
C	0.35	1.02	
I	0.65	1.90	3.29
C * I	0.30	0.90	

Table 4. Effect of type of cuttings and IBA concentration on number of shoots / cutting (30, 60, 90 and 120 DAP).

	30 DAP			60 DAP			90 DAP			120 DAP		
	(C_1)	(C_2)	Mean	(C_1)	(C_2)	Mean	(C_1)	(C_2)	Mean	(C_1)	(C_2)	Mean
I_1 - 0 ppm	1.00	1.00	1.00	2.00	1.00	1.50	3.00	1.00	2.00	3.00	1.00	2.00
I_2 - 500 ppm	4.00	1.00	2.50	3.00	1.00	2.00	2.00	1.00	1.50	2.00	1.00	1.50
I_3 - 1000 ppm	6.00	1.00	3.50	5.00	1.00	3.00	3.00	1.00	2.00	3.00	1.00	2.00
I_4 - 1500 ppm	6.00	1.00	3.50	4.00	1.00	2.50	3.00	1.00	2.00	2.00	1.00	1.50
I_5 - 2000 ppm	9.00	1.00	5.00	7.00	1.00	4.00	5.00	1.00	3.00	3.33	1.00	2.17
I_6 - 2500 ppm	6.33	1.00	3.67	4.00	1.00	2.50	2.87	1.00	1.93	2.33	1.00	1.67
I_7 - 3000 ppm	7.00	1.00	4.00	4.67	1.00	2.83	3.00	1.00	2.00	3.00	1.00	2.00
Mean	5.62	1.00		4.24	1.00		3.12	1.00		2.67	1.00	
	S.Em±	**CD at 5%**	**CV %**	**S.Em±**	**CD at 5%**	**CV**	**S.Em± %**	**CD at 5%**	**CV %**	**S.Em±**	**CD at 5%**	**CV %**
C	0.10	0.29		0.10	0.29		0.06	0.16		0.06	0.16	
I	0.19	0.55	4.66	0.19	0.55	5.89	0.10	0.30	4.17	0.17	0.31	4.76
C * I	0.09	0.26		0.09	0.26		0.45	0.14		0.50	0.14	

may be due to more reserved carbohydrates present in cuttings, thickness of cuttings and cuttings taken from the base of the stock branch (Severino *et al.* 2011).

Exogenous IBA application affected significantly on shoot parameters because it enhance hydrolysis of nutritional reserves under the influence of exogenous auxin (Kochhar *et al.* 2008).

Maximum number of shoots at 30, 60, 90 and 120 DAP (9.00, 7.00, 5.00, and 3.33 respectively) (Table 4) were found in hardwood cuttings when they were treated with IBA 2000 mg / litre (C_1I_5). Minimum numbers of shoots (1.00) were found in C_1I_1 and C_2I_1 to C_2I_7 treatments at 30 DAP and C_2I_1 to C_2I_7 at 60, 90, and 120 DAP. Because of maturity of wood more number of active buds present on hardwood cuttings as the apical portion is removed. In top cuttings, apical portion is not removed so because of the apical dominancy only one shoot is active. As the cuttings are getting older the numbers of shoots are decreased because of the suppression of the growth because of over crowing of shoots and competition between shoots.

Maximum fresh (280.77 g) and dry (96.52 g) biomass were recorded in C_1I_5 treatment and minimum fresh (2.73 g) and dry (0.90 g) biomass were recorded in C_2I_1 which was at par with C_1I_1 (5.70 and 1.36 g, respectively) (Tables 5 and 6).

The increased in production of of leaves and leaf area ultimately increased the photosynthesis, relative growth rate and growth of lateral branching of shoots which increased fresh and dry biomass of shoot and root : shoot ratio.

Thus, combined effect of cutting type and different IBA concentration significantly acted on different shoot parameters. The hardwood cutting with combination with IBA at 2000 mg / litre recorded maximum number of leaves, leaf area and shoot length. More or less similar results were also reported by Singh *et al.* (2011) in pear; Thakur *et al.* (2014) in olive.

Table 5. Effect of type of cuttings and IBA concentrations on fresh biomass of shoot (g) (120 DAP).

IBA / cutting	(C_1)	(C_2)	Mean
I_1 - 0 ppm	5.70	2.73	4.21
I_2 - 500 ppm	56.60	36.80	46.70
I_3 - 1000 ppm	80.67	53.50	67.08
I_4 - 1500 ppm	108.93	64.40	86.67
I_5 - 2000 ppm	280.77	167.17	223.97
I_6 - 2500 ppm	170.50	100.13	135.31
I_7 - 3000 ppm	168.80	87.33	128.07
Mean	124.57	73.15	
	S.Em±	**CD at 5 %**	**CV %**
C	1.25	3.61	
I	2.33	6.76	1.93
C * I	1.10	3.18	

The maximum survival percentage was recorded when hardwood cuttings were treated with IBA at 2000 mg / litre (Table 7). This might be due to overall performance in relation to growth parameters of root and shoots which were significantly better in this treatment and ultimately increased the survival percentage. These observations are in close conformity with findings of Tu *et al.* (1991) in kiwi; Kracikova

Table 6. Effect of type of cuttings and IBA concentrations on dry biomass of shoot (g) (120 DAP).

IBA / cutting	(C_1)	(C_2)	Mean
I_1 - 0 ppm	1.36	0.90	1.13
I_2 - 500 ppm	17.65	13.16	15.40
I_3 - 1000 ppm	26.49	18.13	22.30
I_4 - 1500 ppm	37.49	22.70	30.10
I_5 - 2000 ppm	96.52	46.90	71.71
I_6 - 2500 ppm	56.85	32.01	44.43
I_7 - 3000 ppm	55.17	26.41	40.79
Mean	41.65	22.89	
	S.Em±	**CD at 5 %**	**CV %**
C	0.97	2.82	
I	1.82	5.29	4.62
C * I	0.86	2.49	

Table 7. Effect of type of cuttings and IBA concentrations on survival percentage (120 DAP).

IBA / cutting	(C_1)	(C_2)	Mean
I_1 - 0 ppm	20.00	18.33	19.17
I_2 - 500 ppm	68.33	65.00	66.67
I_3 - 1000 ppm	80.00	73.33	76.67
I_4 - 1500 ppm	78.33	68.33	73.33
I_5 - 2000 ppm	93.33	81.67	87.50
I_6 - 2500 ppm	71.67	68.33	70.00
I_7 - 3000 ppm	73.33	71.67	72.50
Mean	69.29	63.81	
	S.Em±	**CD at 5 %**	**CV %**
C	1.67	4.85	
I	3.13	9.08	3.84
C * I	1.48	4.28	

(1996) in plum, Das *et al.* (2006) in olive. This might be due to overall performance in relation to growth parameters of root and shoots which were significantly better and ultimately increased the survival percentage.

REFERENCES

Das B, Tantry F A and Srivastava K K. 2006. Rooting response of olive stem cuttings under zero energy environment. *Indian J. Hort.* **63**(2): 209-12.

Kochhar S, Singh S P and Kochhar V K. 2008. Effect of auxin and associared biochemical changes during clonal propagation of the biofuel plant *Jatropha curcas*. *Biomass and Bioenergy* **32**: 1136-43.

Kracikova M. 1996. Selection of plum rootstocks for economic propagation by hardwood cuttings. *Vedecke Prace Ovocnarske* **15**: 41-49.

Kraus E J and Krybill H R. 1918. Vegetation and reproduction with special reference to tomato. *Oregon Sta. Bull.* **149**: 1-190.

Severino L S, Lima R L S, Lucena A M A, Freire M A O, Sampaio L R, Veras R P, Medeiros K A A L, Sofiatti V and Arriel N H C. 2011. Propagation by stem cuttings and root system structure of *Jatropha curcas. Biomass and Bioenergy* **35**: 3160- 66.

Singh K K, Chaudhary T and Kumar A. 2014. Effect of various concentrations of IBA and NAA on the rooting of stem cuttings of mulberry (*Morus alba* L.) under house condition in garhwal hill region. *Indian J. Hill Farming* **27**(1) 125-31.

Singh S K, Singh R K, Kumar M, Chaubey S K and Prasad K K. 2011. Studies on Pear Propagation through Cutting Treated with Indole Butyric Acid under Ranchi Condition. *Prog. Hort.* **43**(1).

Thakur M, Sharma D D and Singh K 2014. Studies on the effect of girdling, etiolation and auxin on rooting of olive (*Olea europaea* L.) cuttings. *International J. Farm Sci.* **4**(2): 39-46.

Tu C Q, Jiang H R and Tu Y S. 1991. Rapid propagation of the chinese kiwifruit, Actinidia chinensis, using cutting in full sunshine. *New Zealand J. Crop Hort. Sci.* **19**(4): 355-59.

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Effect of Aluminium sulphate and citric acid on post-harvest qualities of spray chrysanthemum *(Dendranthema × grandiflora)*

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ABSTRACT

An experiment was conducted on five spray cultivars of Chrysanthemum, (*Dendranthema* × *grandiflora* Ramat) *viz.* Kundan, Maghi Yellow, Maghi White, Corcon Small and Jubilee, to see the effect of aluminium sulphate and citric acid on post-harvest attributes and vase-life. The experiment was laid out in factorial completely randomized block design with five varieties and seven treatment combinations replicated thrice. Among different varieties maximum vase-life (19.64 days), solution uptake (40.93ml), floret opening (89.25%) were observed in Kundan, however, minimum weight loss (17.46%) was observed in Corcon Small. Amongst chemical treatments, maximum average vase-life (13.60 days) and minimum weight loss (28.84%) were recorded with 150 ppm citric acid +1000 ppm aluminium sulphate +2% sucrose. Maximum solution uptake (37.83 ml) was recorded with 300 ppm citric acid + 1000 ppm aluminium sulphate + 2% sucrose. However, maximum flower size (3.83 cm) and bud opening (89.13%) were observed under the control.

Key Words: Aluminium sulphate, Citric acid, Sucrose, Post-harvest, Vase-life, Spray cultivars

Chrysanthemum (*Dendranthema* × *grandiflora* Ramat) is native of Asia and north-eastern Europe. It is also called Queen of East and mostly used for cut flower, loose flowers and pot purposes. In India, it is cultivated in 16.63 thousand ha area with a total production of 179.37 metric tonnes (Saxena *et al.* 2015). Fresh flowers lose their freshness and quality both during handling and transportation. Due to high perishability, flower and foliage parts are vulnerable to large post-harvest losses. To preserve best quality of flowers after harvesting and to make them tolerant to fluctuations in environmental conditions treatment with floral preservatives is recommended. Effect of different holding solutions on standard cultivars of chrysanthemum has been reported (Kofranek and Halevy, 1972, Marousky, 1969; 1971; Talukdar *et al.* 2004) but the information on spray cultivars is meagre. Therefore, an experiment was conducted to see the effect of aluminium sulphate and citric acid on post-harvest handling of spray cultivars.

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MATERIALS AND METHODS

The experiment was conducted at Division of Floriculture and Landscaping, IARI, New Delhi. The cut flowers of spray type of cultivars, *viz.* Kundan, Maghi Yellow, Maghi White, Corcon Small and Jubilee were harvested in morning hours. After harvesting, flowers were immediately placed in the bucket containing water and brought to the laboratory. The stems were cut to a uniform length of 30 cm and were dressed by removing lower one-third leaves. The cut stems were kept in vases containing distilled water (T_1) 150 ppm citric acid + 500 ppm $Al_2(SO_4)_3$ (T_2), 150 ppm citric acid + 500 ppm $Al_2(SO_4)_3$ + 2% sucrose (T_3), 150 ppm citric acid + 1000 ppm $Al_2(SO_4)_3$ (T_4), 150 ppm citric acid + 1000 ppm $Al_2(SO_4)_3$ + 2% sucrose (T_5) 300 ppm citric acid + 500 ppm $Al_2(SO_4)_3$ + 2% sucrose (T_6) and 300 ppm citric acid + 1000 ppm $Al_2(SO_4)_3$ + 2% sucrose (T_7).

The experiment was laid out in a factorial completely randomized design, replicated thrice with three stems per replication. Observations on vase-life, flower diameter, solution uptake, per cent floret opening

and per cent weight loss were recorded and data was subjected to analysis of variance (Panse and Sukhatme 1985). Analysis of variance for factorial completely randomized design (FCRD) was carried out using a statistical package OPSTAT version 6.1 computer section, Chaudhary Charan Singh Haryana Agriculture University.

RESULTS AND DISCUSSION

The data shows that out of various cultivars maximum vase-life (19.64 days) was observed in Kundan and it was significantly different from all other cultivars while minimum vase-life (6.81 days) was recorded in Jubilee (Table 1). This difference in vase-life of cultivars may be attributed to their genetic make-up. The variation among cultivars might be due to difference in senescing behaviour by producing higher amount of ACC, ethylene forming enzymes and ethylene (Acati and Jona, 1989). However, Zamski *et al* (1991) stated that the variation in cultivars could be attributed to differences in number of thick-walled supporting cells in xylem element and phloem fibers and presence and absence of a complex ring of secondary thickening in flower peduncles. Comparison of different preservative solutions reveals that maximum vase-life (13.60 days) was observed in cut flowers held in 150 ppm citric acid + 1000 ppm $Al_2(SO_4)_3$ + 2% sucrose solution (T_4) which was at par with T_2, T_3 and T_5, whereas minimum vase-life (11.96 days) was observed in the control. Similarly, Rajagopalan and Abdul Khader (1993) observed that in chrysanthemum, CO1 and CO2 use of aluminium sulphate as a floral preservative retard bacterial growth, thus enhancing the vase-life of cut flowers. Use of holding solution containing citric acid increased vase-life of chrysanthemum cv. Cat Eye's by 9 days compared to the control (Yuniarti *et al,* 2007). Interaction between cultivars and treatments reveals that maximum vase-life (18.78 days) was observed in cv. Maghi White with 150 ppm citric acid + 500 ppm $Al_2(SO_4)_3$ + 2% sucrose solution (T_3) and was significantly superior over all other treatments; however minimum vase-life (5.56 days) was observed in cv. Jubilee under the. Similar to our findings Jain *et al.* (2009) reported that holding cut flowers of chrysanthemum cv. Kargil, Shyamal and Ravi Kiran in solution containing 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ + 2% sucrose had maximum vase-life. The present findings also corroborate with the findings of Banik *et al*. (1999) who recorded maximum vase-life in Chandrama with 0.1% $Al_2(SO_4)_3$ and 3% sucrose. Moreover, presence of sugar in vase solution must have acted as a source of energy. Similarly, Van Doorn (2004) reported that sugar starvation is a cause of petal senescence. Jain *et al.* (2014) also reported that keeping flowers of chrysanthemum cv. Thai Chen Queen in 150 ppm citric acid + 500 ppm aluminium sulphate + 2% sucrose resulted in maximum vase-life (25.41 days).

The effect of cultivars was found significant in flower diameter. The maximum flower diameter (5.26 cm) was observed in cv. Jubilee, while it was minimum (2.81 cm) in cv. Maghi White and at par with cv. Kundan

Table 1. Effect of aluminium sulphate and citric acid on vase-life (days) of chrysanthemum cultivars

Treatment	Cultivars					
	Kundan	Maghi Yellow	Maghi White	Corcon Small	Jubilee	Mean
T_1, Control (distilled water)	12.60	11.78	15.33	14.55	5.56	11.96
T_2, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$	19.33	12.22	16.78	8.78	7.89	13.00
T_3, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose	20.11	12.33	18.78	8.78	6.67	13.33
T_4, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	21.77	12.89	16.67	9.88	6.78	13.60
T_5, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	23.33	12.78	16.33	8.22	6.78	13.49
T_6, 300 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose	20.22	13.22	16.33	8.44	6.67	12.98
T_7, 300 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	20.11	13.63	15.57	5.33	7.33	12.39
Mean	19.64	12.69	16.54	9.14	6.81	
CD (0.05)						
	Treatments (T)=0.78		Cultivars (C)= 0.66		T x C= 1.75	

Table 2. Effect of aluminium sulphate and citric acid on flower diameter (cm) of chrysanthemum cultivars

Treatment	Cultivars					
	Kundan	Maghi Yellow	Maghi White	Corcon Small	Jubilee	Mean
T_1, control (distilled water)	2.98	2.97	2.78	5.03	5.38	3.83
T_2, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$	3.09	2.85	2.99	4.59	5.46	3.80
T_3, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$ + 2% sucrose	3.01	2.74	3.02	4.53	4.79	3.62
T_4, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ + 2% sucrose	2.99	2.89	2.93	4.46	5.17	3.69
T_5, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ + 2% sucrose	2.91	2.77	2.88	4.73	5.23	3.70
T_6, 300 ppm citric acid +500 ppm $Al_2(SO_4)_3$ + 2% sucrose	2.96	2.78	2.85	4.50	5.32	3.68
T_7, 300 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ + 2% sucrose	2.65	2.70	2.91	4.50	5.44	3.64
Mean	2.94	2.81	2.91	4.62	5.26	
CD (0.05)						
	Treatments (T)=NS		Cultivars (C)= 0.36		T x C= NS	

and Maghi Yellow (Table 2). The effect of various chemical preservatives and their interaction was found to be non-significant in flower diameter.

The data shows that cut flowers of cv. Kundan showed maximum solution uptake (40.93 ml) and minimum uptake was observed in cv. Corcon Small (18.34 ml) and was significantly different from all other treatments (Table 3). Among different chemical preservatives; maximum solution uptake (37.83 ml) was recorded in flowers kept in a solution containing 300 ppm citric acid + 1000 ppm $Al_2(SO_4)_3$ + 2% sucrose (T_7), while minimum (20.98 ml) under the control. The interaction between cultivars and treatments showed that maximum solution uptake (49.70 ml) was observed in 150 ppm citric acid + 500 ppm $Al_2(SO_4)_3$ + 2% sucrose (T_4) in cv. Jubilee. Similarly, Jain *et al.* (2009)

Table 3. Effect of aluminium sulphate and citric acid on solution uptake (ml) of chrysanthemum cultivars.

Treatment	Cultivars					
	Kundan	Maghi Yellow	Maghi White	Corcon Small	Jubilee	Mean
T_1, control (distilled water)	29.11	21.44	18.67	14.89	20.78	20.98
T_2, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$	33.00	23.22	34.78	19.67	21.89	26.51
T_3, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose	34.77	21.77	34.55	18.11	21.78	26.20
T_4, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	35.44	23.11	37.44	18.56	49.70	32.85
T_5, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	41.33	22.45	40.44	21.22	29.45	30.98
T_6, 300 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose	35.44	25.89	38.33	19.23	18.00	27.38
T_7, 300 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	77.41	40.75	34.12	16.67	20.22	37.83
Mean	40.93	25.52	34.05	18.34	25.97	
CD (0.05)						
	Treatments (T)=1.69		Cultivars (C)= 1.39		T x C= 3.78	

Table 4. Effect of aluminium sulphate and citric acid on floret opening (%) of chrysanthemum cultivars

Treatment	Cultivars					
	Kundan	Maghi Yellow	Maghi White	Corcon Small	Jubilee	Mean
T_1, control (distilled water)	80.30 (71.06)	79.62 (75.16)	84.17 (77.67)	93.40 (20.22)	83.68 (6.56)	84.23 (50.13)
T_2, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$	86.81 (33.27)	83.43 (41.26)	79.99 (21.73)	79.78 (53.76)	70.55 (39.25)	9.35 (44.63)
T_3, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose	93.20 (68.76)	84.16 (32.85)	85.18 (63.19)	84.03 (34.10)	75.06 (63.43)	4.33 (52.47)
T_4, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	89.37 (39.68)	87.51 (63.28)	75.20 (29.02)	87.42 (57.13)	75.44 (33.39)	2.99 (44.50)
T_5, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	93.71 (75.10)	79.61 (46.18)	76.14 (66.60)	79.83 (39.92)	5.44 (67.37)	80.95 (59.04)
T_6, 300 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose	91.92 (35.20)	84.08 (66.43)	78.17 (20.51)	83.76 (60.04)	78.91 (33.42)	3.37 (43.12)
T_7, 300 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	80.30 (71.06)	84.77 (51.14)	77.63 (69.40)	90.17 (41.26)	65.06 (60.12)	79.57 (58.59)
Mean	89.25 (56.31)	95.00 (32.80)	79.50 (49.73)	85.48 (43.78)	74.88 (51.93)	
CD (0.05)	Treatments (T)=1.55		Cultivars (C)= 1.31		T x C= 3.47	

Figures in parentheses are transformed values

reported that holding flowers of cv. Kargil in 150 ppm citric acid + 500 ppm $Al_2(SO_4)_3$ + 2% sucrose resulted in maximum solution uptake. Rakesh *et al.* (2004) also reported maximum vase-life and highest water uptake in cv. Flirt with 4% sucrose + 0.2% aluminium sulphate + 0.02% cobalt sulphate. This increase in solution uptake may be attributed to the fact that the presence of citric acid and aluminium sulphate in preservative solution might have reduced the solution pH, ultimately reduced vascular blockage due to bacteria and hence increased uptake.

There was maximum floret opening (89.25%) in cv. Kundan while minimum (74.88%) was observed in cv. Jubilee (Table 4). Among various chemical treatments maximum floret opening (84.33%) was recorded under 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose (T_3), while minimum (79.35%) was recorded with 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$ solution (T_2) and was at par with T_7. Interaction between treatments and cultivars reveals that maximum floret opening (93.71%) was recorded in cv. Kundan with 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose (T_5) and was at par with T_1, T_3 and T_7, while minimum floret opening (65.06%) was observed in cv. Jubillee flowers held in 300 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose (T_7). This is because, the addition of germicide inhibited microorganisms growth in solution and apparently promoted solution uptake. Hussain *et al.*, (2001) reported that sucrose in combination with citric acid or aluminium sulphate maintains endogenous levels of soluble sugars and soluble proteins which in turn provide energy as well as required osmaticum for floret development and longevity. Moreover, flowers fed with sucrose solution have increased opening as the addition of sucrose allows the flower to develop fully which is not possible with water. Jowkar *et al.*, (2012) reported that considering different aspects of biocide application i.e. microbial control, solution uptake, relative fresh weight, flower longevity, and appearance etc. aluminum sulfate was an efficient treatment.

The effect of cultivars, treatments and interaction between treatments and cultivars was significant for weight loss (Table 5). Among cultivars, minimum weight loss (17.46%) was recorded in cv. Corcon Small, while maximum (37.00%) was observed in Maghi White which was significantly different from all other treatments. Among preservatives, minimum weight loss (28.84%) was recorded in flowers held in a preservative solution containing 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose (T_4) and was it par with T_2, whereas maximum weight loss (33.97%) was observed under the control. Interaction data between treatment × cultivar shows that minimum weght loss (3.52%) was recorded in cv. Corcon small in 150 ppm citric acid

Table 5. Effect of aluminium sulphate and citric acid on weight loss (%) of chrysanthemum cultivars

Treatment	Cultivars					
	Kundan	Maghi Yellow	Maghi White	Corcon Small	Jubilee	Mean
T_1, control (distilled water)	42.34 (42.22)	43.53 (69.21)	30.16 (32.00)	13.76 (60.30)	40.06 (32.83)	3.97 (47.31)
T_2, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$	43.52 (75.73)	31.46 (40.58)	40.81 (63.19)	23.57 (35.91)	30.33 (60.75)	33.94 (55.23)
T_3, 150 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose	29.40 (36.47)	41.21 (63.29)	33.28 (29.68)	12.33 (60.30)	30.38 (44.81)	9.32 (46.91)
T_4, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	29.46 (73.64)	12.00 (34.48)	45.19 (66.54)	28.12 (40.42)	29.45 (62.14)	28.84 (55.45)
T_5, 150 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	25.51 (38.88)	34.44 (66.22)	35.39 (23.81)	24.55 (62.67)	49.70 (36.94)	33.92 (45.71)
T_6, 300 ppm citric acid +500 ppm $Al_2(SO_4)_3$ +2% sucrose	35.12 (63.64)	42.07 (30.31)	39.45 (65.99)	16.34 (34.38)	6.16 (61.82)	33.83 (51.23)
T_7, 300 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ +2% sucrose	52.10 (36.50)	31.94 (71.76)	35.43 (10.79)	3.52 (66.20)	32.74 (34.88)	31.15 (44.03)
Mean	36.78 (52.44)	33.81 (53.69)	37.10 (41.71)	17.46 (51.45)	35.55 (47.74)	
CD (0.05)	Treatments (T)=2.61		Cultivars (C)= 2.21		T x C= 5.85	

Figures in parentheses are transformed values

+500 ppm $Al_2(SO_4)_3$ + 2% sucrose (T_3) and was at par with T_4, while maximum weight loss 52.10% was recorded in cv. Kundan with 300 ppm citric acid +1000 ppm $Al_2(SO_4)_3$ + 2% sucrose (T_7). Similarly, Jain *et al.* (2009) observed that holding cut flowers of chrysanthemum cv. Shyamal in 300 ppm citric acid +500 ppm $Al_2(SO_4)_3$ + 2% sucrose solution resulted in minimum weight loss. Park *et al.* (2000) also reported that fresh weight of cut chrysanthemum was higher in 250 ppm $Al_2(SO_4)_3$ and 3% sucrose solution. The possible reason for minimum weight loss might be low transpirational losses. The presence of aluminium sulphate in vase solution resulted in partial closure of stomata and hence reduced transpiration loss of water.

Thus, it is concluded that keeping flowers of cultivars Kundan, Maghi Yellow and Jubilee in 150 ppm citric acid + 500 ppm $Al_2(SO_4)_3$ +2% sucrose increased the vase-life and improved other post-harvest qualities, however cv. Corcon Small performed best under distilled water.

REFERENCES

Accati E G, and Jona R, 1989.Parameters influencing gerbera cut flower longevity. *Acta Horticulturae,* **261**: 63-68

Banik A K, Dutta P and Barman D. 1999. Effect of aluminium sulfate and sucrose on vase-life of Chrysanthemum morifolium Ramat. var Chandrama. *Environment and Ecology*. **17**(1): 248-49

Hussain C T S, Misra R L, Bhattacharjee S K and Voleti S R. 2001. Changes in soluble carbohydrates and proteins in cut gladiolus. *Journal of Ornamental Horticulture.* **4**(2): 83-86.

Jain Ritu, Prasad K V and Singh O P. 2009. Effect of different floral preservative solutions on vase life of few chrysanthemum spray cultivars. *Journal of ornamental Horticulture*. **12**(4): 245-50.

Jain Ritu, Janakiram T, Singh K P and Kumawat G L. 2014. Reduction of foliage discoloration and extending vase-life of chrysanthemum *(Dendranthema × grandiflora)* with different floral preservatives. *Indian Journal of Agricultural Sciences*. **84**(10): 1250-3.

Jowkar Mohammad Mahdi, Ahmad Mohsen Kafi, Khalighi and Nader Hasanzadeh. 2012. Evaluation of aluminum sulfate as vase solution biocide on postharvest microbial and physiological properties of 'Cherry Brandy' rose. *Annals of Biological Research.* **3**(2): 1132-44

Kofranek A M and Halevy A M. 1972. Conditions for opening cut chrysanthemum flower buds. *Journal of American Society for Horticultural Science*. **97**(5): 578-584.

Marousky F J. 1969. Influence of various commercial floral preservatives and 8-hydroxy quinoline citrate plus sucrose on development and lasting ability of flower buds of several chrysanthemum cultivars. *Proceedings of Florida State Horticultural Society*, **82**: 381-385.

Marousky F J. 1971. Handling and opening bud cut chrysanthemum flowers with 8-hydroxy quinoline citrate and sucrose. Marketing Research Report, United States Development of Agriculture, **905**: 14.

Panse V G and Sukhatme P V. 1985. Statistical methods for agricultural workers. Published by, ICAR, New Delhi.

Park Y Y, Kim H K and Cho M S. 2000. Effect of Sucrose, Germicides and ABA in the preservatives on postharvest quality of Cut Chrysanthemum 'Kyoungsubang'. Korean *Journal of Life Science.* **10**(6): 591-597

Rajagopalan A and Abdul Khader M. 1993. Effect of pulsing treatments on vase life of chrysanthemum cut flowers. *South Indian Horticulture*. **41**(1): 61-63

Rakesh Singhrot R S, Gupta A K and Moond A K. 2004. Studies on vase life of cut flowers in chrysanthemum. *Haryana Journal of Horticultural Science*. **33**(1&2): 78-80

Saxena M, Bhattacharya S and Malhotra S K. 2015. Overview: Crop-wise Area and Production of Horticultural Crops, 2012-13 to 2014-15. *In: Horticulture Statistics at a Glance* 2015, NHB, Ministry of Agriculture & Farmers Welfare, GOI, Oxford University Press, New Delhi, p. 17.

Talukdar M C, Sharma B J, Das S and Mahanta S J. 2004. Effect of pulsing on post harvest quality of chrysanthemum (*Dandranthema grandiflora,* Tzevelve.) under different storage conditions. *Journal of Ornamental Horticulture*. **7**(2): 1333-140.

Van Doorn W G. 2004. Is petal senescence due to sugar starvation? *Plant Physiologica*. **134**: 35 42.

Yuniarti P E, Prahardini R and Santoso P. 2007. Research on lengthening the shelf life of Cat Eye's chrysanthemum cut flower. *Acta Horticulturae.* **755**: 109-112

Zamski E, Starkman F and Zieslin N. 1991. Mechanical strength and anatomical structure of the peduncles of rose *(Rosa × hybrid)* flowers. *Israel Journal of Botany*. **40**(1): 1-6.

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Studies of pollen-grains in Heliconia *(Heliconia* spp.*)* for improvement

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ABSTRACT

The studies were undertaken on 30 selected species and cultivars of Heliconia having good cut flower qualities and popularity in the market. A thorough understanding of pollen characters is a fundamental requirement for any successful breeding. The interspecific hybrids recorded lower fertility percentage, while other species and cultivars recorded higher fertility. Pollen shape varied within same species in different cultivars.

Key Words: Pollen grains, cut flower, Inter specific hybrid, Pollen shape.

The cultivation of Heliconia (*Heliconia* spp.) is rapidly expanding in tropical areas throughout the world. It is one of the most important cut flower and landscape plants. A thorough knowledge of floral biology is essential for its improvement. Pollen is a useful character for identifying and classifying members of the genus. Most pollen features are best used to distinguish broad groups of species rather than individual species (Kress and Stone, 1983). Therefore, studies were undertaken on morphology and fertility of pollen-grains of some commercially significant Heliconias in which development of novel hybrids have immense scope in our country.

MATERIALS AND METHODS

The studies were undertaken on Heliconia *(Heliconia spp.)*" at the Department of Pomology and Floriculture, College of Agriculture, Vellayani, during 2010-2012. Thirty species and cultivars of Heliconia with good cut flower qualities and popularity in the market were selected. Pollen-grains were observed under binocular microscope, images were taken and pollen diameter measurements were made using software Motive 2 Plus. Pollen-grains were selected from fully-opened flowers and stained in 2% glycerin: acetocarmine solution in the ratio of 1:1. Diameter of 10 normal shaped and well-stained pollen-grains was measured and the mean diameter was recorded in microns. The pollen fertility of cultivars was estimated using acetocarmine stain. For the estimation of pollen fertility flower buds were collected at the time of anthesis and pollen-grains were stained with 1:1 glycerin - acetocarmine (2%) and viewed under 10 × magnifica-tions. Three slides were prepared for each variety and from each slide five microscopic fields were scored and the data recorded. All the pollen-grains that were well filled and stained were counted as fertile and others as sterile. The pollen fertility was calculated as,

$$\text{Pollen fertility} = \frac{\text{Number of well filled and uniformly stained pollen-grains}}{\text{Total number of pollen-grains}} \times 100$$

RESULTS AND DISCUSSION

The largest pollen diameter was recorded in cultivar *Heliconia collinsiana* × *Heliconia bourgaeana cv.* Pedro Ortiz with mean value 82.53 µm, which was on a par with *Heliconia stricta cv.* Dorado Gold (81.15 µm), *Heliconia stricta cv.* Iris (80.33 µm), *Heliconia wagneriana cv.* Red (80.13 µm). Lowest pollen diameter was recorded in cultivar T_7 *Heliconia psittacorum* × *Heliconia spathocircinata cv.* Keanae Red with mean value 39.13 µm, which was on a par with *Heliconia psittacorum cv.* Andromeda (47.07 µm), *Heliconia psittacorum* × *Heliconia spathocircinata cv.* Alan Carle (48.50 µm) and *Heliconia psittacorum cv.* Lena (49.65 µm).

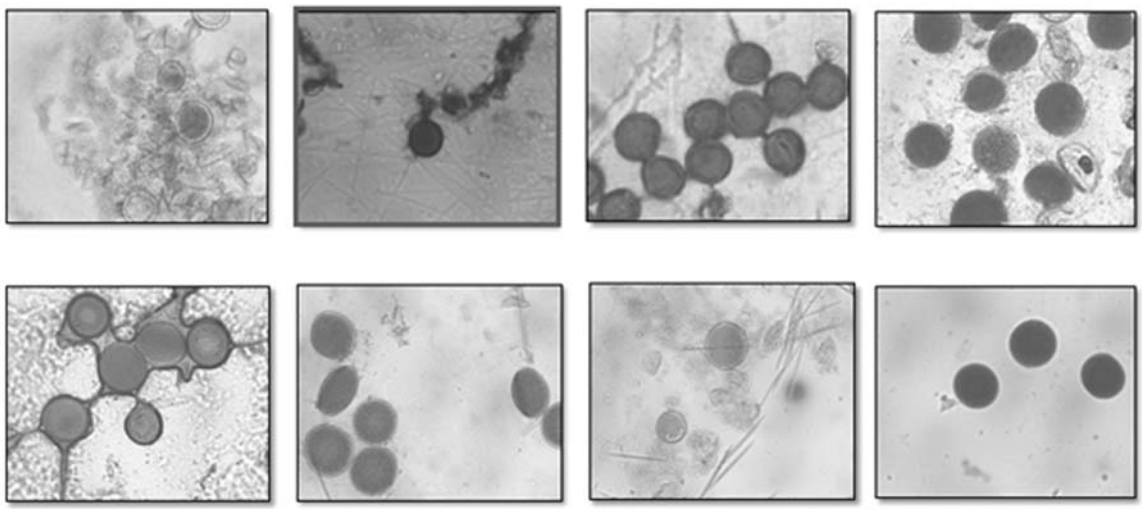
Highest fertility was recorded in cultivar T8 *Heliconia psittacorum cv.* Lena, *i.e.* 100%, which was on a par with *Heliconia rostrata* (99.50%), *Heliconia psittacorum cv.* Pascal (98.00%), *Heliconia psittacorum cv.*

Andromeda (98.00%), *Heliconia psittacorum cv*. St. Vincent Red (98.25%) and *Heliconia stricta cv*. Iris (98.00%). Lowest fertility percentage was recorded in *Heliconia psittacorum* × *Heliconiaspathocircinata cv*. Keanae Red, *i.e.* 15.25 %, which was on a par with *Heliconia psittacorum* × *Heliconia spathocircinata cv*. Tropics (16.75%).

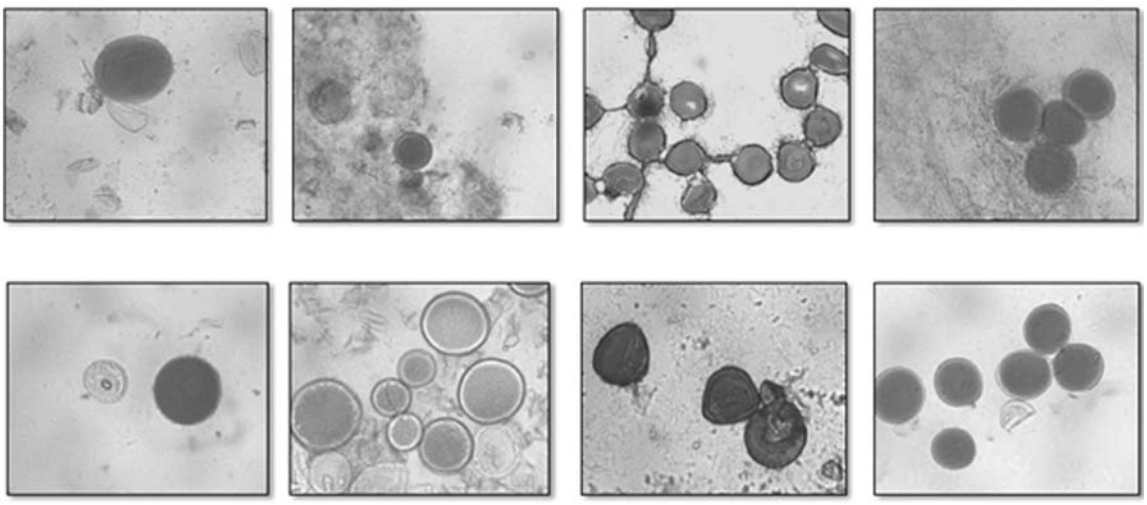
Understanding pollen biology and assessment of pollen viability is critical for monitoring pollen vigour during storage, gene bank maintenance, incompatibility and fertility studies and evaluation of pollen germination after exposure to certain conditions. The largest pollen diameter was recorded in *Heliconia collinsiana* × *Heliconia bourgaeana cv*. Pedro Ortiz and lowest pollen diameter was recorded in *Heliconia psittacorum* × *Heliconia spathocircinata cv*. Keanae Red.

It is observed that cultivars Golden torch; Guyana, deRooij and Alan Carle which are all interspecific hybrids produce round shape pollen-grains of different size but *Psittacorum* cultivars like Lena, Pascal and Strawberry produced perfect spheroid shape and uniform-sized pollen-grains. Petra and Sassy produced mix type of pollen having oval and round shape. Lady Di and Andromeda produced pollen-grains of irregular in shape. St. Vincent Red produced slightly oval shape pollen-grains. These results are therefore in agreement with those of Kress and Stone (1983). They observed large, variable shaped pollen grains in *Heliconia stricta*.

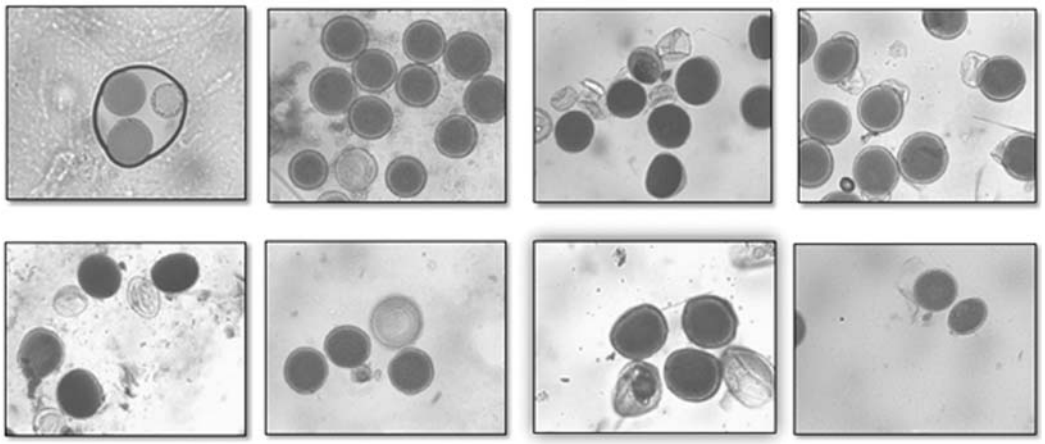
Cultivar Sexy pink and *Heliconia rostrata* produced round and uniform size pollen-grains. *Heliconia wagneriana* cv. Red produced spherical shape pollen with different size, whereas *Heliconia wagneriana* cv.



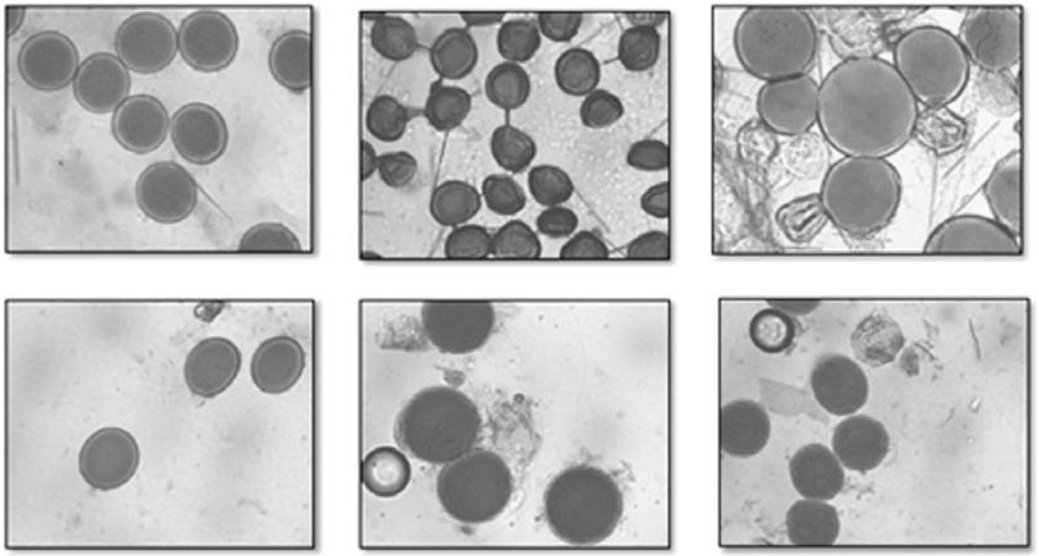
Top row: Golden Torch, Guyana, Parakeet and Latispatha
Bottom row: deRooij, Petra, Keanae Red and Lena



Top row: Tropics, Alan Carle, Lady Di and Rostrata
Bottom row: Wagneriana Red, Christmas Red, Wagneriana Yellow and Sexy Pink



Top row: Orange gyro, Pascal, Jacquinii and Pedro Ortiz
Bottom row: Sunrise, Mathiasiae, Dorado Gold and Fire flash



Top row: Strawberries, Andromeda and Sassy
Bottom row: St. Vincent Red, Iris and Lingulata

Yellow produced typical pollen shape with a spine like protuberance at one end. *Heliconia angusta* cv. Christmas Red produced round shaped pollen-grains of different size. *Stricta* species produced mixed typed of pollen-grains, showing shapes ranging from irregular to circular. Higher proportions of normal pollen-grains indicate higher degrees of chromosome homology.

Self-and cross-compatibility problems was reported in *Heliconia* which hinders the seed production of various varieties by selfing or crossing (Lee *et al.* 1994). In agreement with the above observation to the pollen fertility ranges from 33-100%. Highest fertility percentage was recorded in *Heliconia psittacorum* cv. Lena (100%). Lowest fertility percentage was recorded in *Heliconia psittacorum* × *Heliconia spathocircinata* cv. Keanae Red. In hybrid plants differences in pollen production vary with the genetic constitution. Fluctuation in percentages of good and bad pollen in pure species is probably not influenced by external factors but by the physiological adjustmentsmade to flowering and senescence.

Interpsecific hybrids of *psittacorum* species recorded lowest percentage for fertility, whereas *psittacorum* species recorded highest percentage of fertility. This shows that sterility of pollen might be one reason for poor seed set in some varieties. Similar results were reported by Sheela *et al.* (2005) and Sanjeev (2005).From the present study, it is evident that considerable variability exists among, different Heliconias studied regarding pollen characteristics. Hence suitable cultivars should be selected with caution considering their compatibility and fertility characteristics for utilizing

Table 1. Means value of pollen-grains size (µm) and pollen fertility (%)

	Accession	Average pollen size in (µm)	Pollen fertility (%)
T1	*Heliconia psittacorum* × *Heliconia spathocircinata* cv. Golden Torch	50.28	35.75
T2	*Heliconia psittacorum* × *Heliconia spathocircinata* cv. Guyana	51.70	39.75
T3	*Heliconia psittacorum* cv. Parakeet	54.35	92.75
T4	*Heliconia latispatha*	64.05	53.75
T5	*Heliconia psittacorum* × *Heliconia marginata* cv. deRooij	65.74	49.50
T6	*Heliconia psittacorum* cv. Petra	66.33	96.50
T7	*Heliconia psittacorum* × *Heliconia spathocircinata* cv. Keanae Red	39.13	15.25
T8	*Heliconia psittacorum* cv. Lena	49.65	100.00
T9	*Heliconia psittacorum* × *Heliconia spathocircinata* cv. Tropics	74.70	16.75
T10	*Heliconia psittacorum* × *Heliconia spathocircinata* cv. Alan Carle	48.50	33.00
T11	*Heliconia psittacorum.* cv. Lady Di	65.18	81.25
T12	*Heliconia rostrata*	65.73	99.50
T13	*Heliconia wagneriana* cv. Red	80.13	73.75
T14	*Heliconia angusta* cv. Christmas Red	65.98	40.00
T15	*Heliconia wagneriana* cv. Yellow	79.50	62.50
T16	*Heliconia chartacea* cv. Sexy Pink	69.10	84.75
T17	*Heliconia latispatha* cv. Orange gyro	50.15	53.00
T18	*Heliconia psittacorum* cv. Pascal	63.90	98.00
T19	*Heliconia caribaea* × *Heliconia bihai* cv. Jacquinii	70.95	50.00
T20	*Heliconia collinsiana* × *Heliconia bourgaeana* cv. Pedro Ortiz	82.53	65.50
T21	*Heliconia sunrise*	71.93	45.00
T22	*Heliconia mathiasiae*	64.95	63.50
T23	*Heliconia stricta* cv. Dorado Gold	81.15	80.00
T24	*Heliconia densiflora Verlot* cv. Fire Flash	63.23	63.50
T25	*Heliconia psittacorum* cv. Strawberries	63.43	63.50
T26	*Heliconia psittacorum* cv. Andromeda	47.07	98.00
T27	*Heliconia psittacorum* cv. Sassy	79.70	85.75
T28	*Heliconia psittacorum* cv. St. Vincent Red	70.45	98.25
T29	*Heliconia stricta* cv. Iris	80.33	98.00
T30	*Heliconia lingulata* cv. Fan	66.20	34.75
	CD (5%)	13.07	3.54

for further breeding programmes.

Mean pollen diameter values of 30 selected specie and cultivars are presented in Table 1.

REFERENCES

Kress W J and Stone D. 1983. Morphology and phylogenetic significance of exine-less pollen of *Heliconia* (Heliconiaceae). *Syst. Bot.* **8**(2): 149-67.

Lee Y H, Ng, N Y and Goh C J. 1994. Pollen formation and fruit set in some cultivars of *Heliconia psittacorum*. *Sci. Hort.* **60**: 167-72.

Sanjeev S J. 2005. 'Floral biology and compatibility studies in *heliconia*'. M.Sc (Hort) thesis, Kerala Agriculture University, Thrissur, p. 107.

Sheela V L, Rakhi R, Nair C S J and George T S. 2005. Genetic Variability in *heliconia. Journal of Ornamental Horticulture* **8**(4): 284-286.

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Evaluation of Noni (*Morinda citrofolia*) as a mixed crop in coconut garden under South Gujarat condition

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ABSTRACT

A field experiment was conducted on integrated cropping system in coconut to study the effect of noni *(Morinda citrifolia* L.*)* as mixed crop in coconut, during 2010-11 to 2015-16 at Regional Horticultural Research Station, ASPEE College of Horticulture and Forestry, Navsari Agricultural University, Navsari. Twenty-five each of tissue culture plantlets and seedlings were planted as non replicated trial under old coconut garden. The growth and yield performance of noni as well as coconut were recorded with economics of the cropping system during 2015-16. The results revealed that, functional leaves on crown of coconut increased from 26.2 to 27.5 & annual leaf production per palm increased from 11.10 to 12.20 during the years 2011-12 to 2015-16. In case of yield of coconut, the initial yield was recorded as 60 nuts / palm which was increased to 74 nuts / palm in the year 2015-16. However, maximum yield of *Morinda citrifolia* was recorded in seedling plants (12.79 kg / plant) than in tissue culture plants (9.52 kg / plant) in the year 2015-16. Whereas, the juice percentage of *Morinda citrifolia* are recorded 60% with TSS of 6.5ºB. The study indicated that inter cropping of *Morinda citrifolia* increased productivity of coconut as well as economics of *Morinda citrifolia* as inter crop indicated profitability of cultivation in coconut than monocrop.

Key Words: Mixed crop, Integrated cropping, Tissue culture, Plantlets, Seedilings

Coconut (*Cocus nucifera* L.) is an important perennial oil-yielding crop of humid tropics and is mainly grown in Kerala, Karanataka, Tamil Nadu, Andhra Pradesh, coastal districts of Maharashtra and Gujarat. Being a small holders crop in India, when grown as monocrop, it does not provide adequate income and employment to dependent families. The adult palm of sole crop of coconut, spaced at 7.5 m × 7.5 m apart effectively uses only 22.3% of land area, while average air space utilization by canopy is 30% and solar radiation interception is 45-50% (Bavappa *et al.* 1986). Thus, coconut gardens offers excellent opportunities for inclusion of compatible component crops in inter spaces, for effective utilization of natural resources. Unlike in annuals, the potential for increasing productively per unit area of land, time and inputs is considerably higher in perennial crops (Bavappa and Jacob, 1982). The coconut-based crop systems evolved in response to the pressure of shrinking land resource base coupled with high population density which necessitated a conscious attempt on the part of farmers to achieve their goals by living within biophysical, ecological and economic constraints (Maheswarappa *et al.* 2013). Noni (*Morinda citrifolia*) is compatible perennial medicinal plant in coconut based cropping system. Its juice has antioxidant properties and targeted the digestive, intestinal, respiratory and immune systems (www. en.wikipedi. org / wiki / *Morinda citrifolia*). Hence study was initiated to evaluate performance of *Morinda citrifolia* as a mix crop in coconut.

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MATERIALS AND METHODS

Field trial on "Performance of *Morinda citrifolia* as mixed crop in coconut garden under South Gujarat region" was conducted at Regional Horticultural Research Station, ASPEE College of Horticulture and Forestry, Navsari (Gujarat) during 2010-11 to 2015-16. The experiment was conducted on 40 years old West Coast Tall garden spaced at 7.5 m × 7.5 m. The Regional Horticultural Research Station is situated on 20°57'

North latitude and 72° 57' East longitudes and has an altitude of 12 m above mean sea-level. The mean annual rainfall during 2010-2015 was 1600-1900 mm. The mean maximum temp is 35°C, while minimum temperature is 20°C. *Morinda citrifolia* was planted in June-2010 as a mixed crop in single hedge system at 3.75 m plant to plant distance at the centre of two rows of coconut palm. Twenty five each of tissue culture plantlets and seedlings were planted as non replicated trial. The experiment plot of coconut mixed crop with *Morinda citrifolia* was maintained as per the recommendations. Mulching with coconut leaves were followed in the summer months. Drip irrigation was followed after rainy season *i.e.* October-May for both coconut and Noni. The growth & yield observations of coconut *i.e.*, average no. of functional leaves on the crown, annual leaf production per palm, no. of nuts per palm and no. of nuts per hectare were recorded in 2011-12 to 2015-16. The growth & yield observations of Noni *viz.* plant height (cm), no. of branches per plant, no. of fruits per plant, weight of fruits per plant were recorded during 2015-16. Juice percentage and T.S.S (°Brix) were also recorded on both planting material. The economics of cropping system was calculated including labour cost, input cost, irrigation, other miscellaneous charges for both planting materials and converted into economics per ha.

RESULTS AND DISCUSSION

The growth and yield performance of coconut as influenced by mix crop of *Morinda citrifolia* is presented in Table 1. The average functional leaves on the crown increased from 26.20 to 27.50 whereas, annual functional leaf production per palm showed same trend and increased from 11.10 to 12.20 leaves / palm due to mix crop of *Morinda citrifolia*. Mean five year yield data indicated that nut yield increased 23.33% over pre experimental yield (2011-12 to 2015-16). The yield data revealed that average initial yield of coconut was 10800 nuts / ha which increased to 13320 nuts / ha. due to mixed crop of *Morinda citrifolia*. Results analogous to these finding were also reported by Nair and Balakrishnan (1976) in coconut mixed crop with cocoa. The additional increase in yield of coconut under mix cropping of *Morinda* could be due to synergistic effect of crop combination (Khandekar *et al.* 2014).

The growth and yield performance of *Morinda citrifolia* as mix crop in coconut is presented in Table 2. Maximum plant height (5.68m.), stem girth (16.75cm) and number of branches per plant (24.91) were found in seedling than tissue culture plants. This may be due to faster growth of seedlings than tissue culture plants. However, number of fruits per plant (325), total fruit weight per plant (11.15kg), and yield of fruits (2.78t / ha) was also recorded maximum in seedlings than tissue culture plants (2.38). The juice percentage was similar (60%) in tissue culture to seedling plants with 7.0% T.S.S. The maximum yield in seedling plants than tissue culture plants could be due to faster growth rate of seedling plants and more production of food material over tissue culture plants. Similarly, more biomass production per plant (3.90 kg) was recorded in seedling plants than tissue culture plants (2.00 kg). Similar findings also reported by Khandekar *et al.* 2014 in coconut mixed crop with noni.

The economics of production of *Morinda citrifolia* mix crop in coconut is presented in Table 3. Maximum gross returns were recorded in coconut + seedling

Table 1. Growth and yield of coconut as influenced by mix crop of *Morinda citrifolia*

Treatment	Functional leaves on crown		Annual leaf production / palm		Yield (nuts)				Increase over initial yield (%)
					2008-09 to 2010-11		2011-12 to 2015-16		
	2008-09 to 2010-11	2011-12 to 2015-16	2008-09 to 2010-11	2011-12 to 2015-16	Nuts / palm	Nuts / palm	Nuts / palm	Nuts / palm / ha	
Coconut + noni	26.2	27.5	11.1	12.2	60	10800	74	13320	23.33

Table 2. Growth, yield and quality parameters of *Morinda citrifolia* as mix crop in coconut

Planting material	Plant height (m)	Stem girth (cm)	No. of branches / plant	No. of Fruits / plant	Total fruit weight / plant (kg)	Yield (tonnes / ha)	Juice %	T S S (ºB)	Biomass production / plant (kg)
Tissue cultured plants	4.85	14.82	22.87	190	9.52	2.38	60%	7.0	3.90
Seedlings	5.68	16.75	24.91	325	11.15	2.78			2.00

Table 3. Economics of *Morinda citrifolia* as mix crop under coconut garden

Crop	Yield / ha	Cost of production (₹ / ha)	Gross return (₹ / ha)	Net return (₹ / ha)	Benefit:Cost ratio
Coconut (mono)	13,000 nuts	30,440	91,000	60,550	1.98
Coconut + noni tissue cultured plants	13,320 nuts+2,380 kg noni	50,500	1,88,440	1,37,940	2.73
Coconut + noni seedling	13,320 nuts+2,780 kg noni	45,500	2,04,440	1,58,940	3.49

Selling price: coconut= ₹ 7 / nut and noni fruits (avg. price) = ₹ 40 / kg

plants (Rs. 204440.00) than coconut + tissue culture plants (Rs. 188440.00) and maximum net return of Rs. 158940.00 with 3.49 B:C ratio. Growing of both tissue culture plants and seedling plants increase in yield than pre experimental yield of coconut. Thus by utilizing same land, resources like space, light, irrigation facility, *Morinda Citrifolia* is a suitable mix crop under cropping system in coconut.

REFERENCES

Bavappa K V A and Jacob V J. 1982. High intensity multispecies cropping- A new approach to small scale farming in the tropics. *World Crops* **34**(2): 47-50.

Bavappa K V A, Abdul Khader K B, Biddappa C C, Khan H H, Kasturi K V, Ramadasan A, Sundaragu P, Bopaiah B M, Thomas G V, Misra L P, Balasimha D, Bhat N T and Shama Bhat K. 1986. Coconut and arecanut based high density multi-species cropping systems. *Journal of Plantation Crops* **14**(2): 74-87.

Khandekar R G, Nagwekar D D, Sawant V S, Gurav S S and Haldankar P M. 2014. Performance of Morinda citrofolia as mixed crop in coconut under Konkan region of Maharashtra. *International Journal for Research in Emerging Science and Technology* **1**(4): 63-65.

Maheswarappa H P, Dhanapal R, Subramanian P and Palaniswami C. 2013. Division of Crop Production-Evaluation of coconut based high density multi-species cropping system under organic and integrated nutrient management. *Journal of Plantation Crops* **41**(2): 130-35.

Nair P K R and Balakrishnan T K. 1976. Pattern of light interception by canopies in a coconut + cocoa crop combination. *Indian Journal of Agricultural Science* **46**(10): 453-62.

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Survey for purple blotch [*Alternari porri* (Ellis) cif.] on onion *(Allium cepa)* in northern parts of Karnataka

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ABSTRACT

Onion (*Allium cepa* L.) is most important commercial vegetable crop cultivated extensively in India, accounting for 90% of the exported vegetables from India. The production of bulbs and seeds is limited by certain diseases. The most serious is purple blotch caused by *Alternaria porri* (Ellis) Cif. The disease causes extensive damage to bulbs as well as seed crop and also a major limiting factor in its cultivation. Therefore, investigation was conducted through survey to know the disease incidence or severity and collection of infected samples. A survey was conducted during *kharif* 2013-2014 in onion-growing areas of northern Karnataka, *viz.* Bijapur, Bagalkot, Gadag and Dharwad districts. The highest per cent disease index was noticed in Ilkal village of Bagalkot district. While, lowest per cent disease index was noticed in Kerur village of Bagalkot district. Among the districts, severity of disease was more in Bijapur and less in Gadag. Isolation was made from onion leaves showing typical purple blotch symptoms. Pure culture of *A. porri* was obtained and its pathogenicity to onion plants was proved. On the basis of isolation and morphological studies, the pathogen was identified as *Alternaria porri* (Ellis) Cif.

Key Words: Onion, Purple blotch,Alternaria blight, PDI and Pathogenicity

Onion (*Allium cepa* L.) a member of family slliaceae, is an important vegetable crop grown in India. Onion is cultivated and used around the world. India is a traditional grower and assumes second positionin onion production with 86.34 million tonnes from 4.36 million ha area (FAOSTAT 2013). Onion is cultivated round the year throughout the country. Onion is susceptible to numerous pests and diseases throughout growing period under field conditions. Alternaria blight is one of the most devastating disease (Marmath *et al.*, 2013). The most important factors for low productivity are the diseases like purple blotch, downy mildew, stem phylium blight, basal rot and storage rots and non-availability of varieties resistant to biotic and abiotic stresses. Among foliar diseases, purple blotch is one of the most destructive diseases, commonly prevailing in almost all onion-growing pockets of the world. Losses ranging from 30-100 per cent. The disease may reach epidemic states during favourable conditions of high relative humidity (80-90%) and optimum temperature (24 ± 1°C) (Yadav *et al.*, 2013).

The name "Purple blotch" for this disease was proposed by Nolla (1927). He named the causal organism as *Alternaria alli* which was later amended to *Alternaria porri*.

Alternaria infection of onion is widespread, particularly in rainy season or high moisture conditions. Survey and surveillance form the basis for any successful plant protection strategy. Successful plant protection depends upon early detection of the disease severity followed by timely adoption and application of preventive measures (Sudarshan Rao, 1975). However, systemic survey on the distribution and severity in Northern parts of Karnataka is lacking. There is a need to undertake systemic survey to identify hot spots for the disease in Northern parts of Karnataka. Keeping in view, the present investigation was undertaken to know the disease severity in northern parts of Karnataka.

MATERIALS AND METHODS

A roving survey was conducted to know the per cent disease index of purple blotch disease in districts of Northern Karnataka during *kharif* 2013 when the

crop was at physiological maturity. The survey was carried out from onion growing districts, *viz.* Bijapur, Bagalkot, Gadag and Dharwad. The purple blotch severity was scored by following 0-5 scale as given by Sharma (1986).The details of scales are as shown below: 0 no disease symptoms; 1 a few spots towards tip covering 10 per cent leaf area; 2 several dark purplish brown patch covering upto 20 per cent leaf area; 3 several patches with paler outer zone covering upto 40 per cent leaf area; 4 leaf streaks covering upto 75 per cent leaf area or breaking of the leaves from center; 5 complete drying of the leaves or breaking of the leaves from centre.

Further, per cent disease index (PDI) was worked out by using following formula proposed by Wheeler (1969).

$$PDI = \frac{\text{Sum of individual ratings}}{\text{Total no. of leaves observed}} \times \frac{100}{\text{Max. Grade}}$$

The pathogen *(Alternaria porri)* from the purple blotch infected leaf samples collected from different areas of Northern Karnataka were isolated separately by following tissue isolation technique. The infected leaves along with healthy portions were cut into small bits and surface sterilized with 1:1000 mercuric chloride solutions for 30 seconds and washed three times in sterile distilled water before transferring them to potato dextrose agar. The plates were incubated at room temperature (28 ± 1°C) and observed periodically for fungal growth. The colonies which developed from the tissue bits were transferred to PDA slants.

A 10 ml of clear sterilized water agar of two per cent strength was poured into Petri plates and allowed to solidify. Dilute spore suspension was prepared using sterile distilled water from 12 days old culture. One ml of suspension was spread uniformly in Petri plates over which two per cent agar was poured aseptically and allowed to solidify. Then the plates were examined under low power objective (10 xs) of compound microscope to locate the conidia. Single isolated conidium was then marked under the microscope field with ink on the surface of the plate. Those marked agar areas were cut and transferred to PDA slants with the help of Cork borer (2 mm)under aseptic conditions and incubated at temperature of 28 ± 1°C.

Onion seedlings were raised in earthen pots, size 6" × 5", filled with sterilized soil. Plants were thoroughly cleaned with sterilized distilled water using moist cotton. Later, plants were sprayed with distilled water. They were covered with polythene bags for 24 hours. The inoculum suspension from ten day old culture was prepared in sterile distilled water and sprayed on to the plants. Similarly control plants were sprayed with sterile distilled water for comparison.

The seedlings were covered with polythene bags and were incubated for 120 hours to ensure successful penetration of the pathogen into the tissue. The polythene bags were removed after five days and seedlings were kept under greenhouse conditions. Observations were made regularly for the appearance and development of symptoms. After appearance of disease symptoms, re-isolation was made from the diseased tissues of artificially infected plants. The isolate obtained was compared with the original culture for confirmation of fungus under study.

RESULTS AND DISCUSSION

During the present investigation a field survey was conducted to gather information on the severity of purple blotch of onion from onion-growing districts of Northern Karnataka. A roving survey was carried out for recording the severity of purple blotch disease of onion during *kharif* 2013 in four major onion-growing districts of Northern Karnataka, *viz.* Bijapur, Bagalkot, Gadag and Dharwad. The survey for symptomatology, severity, distribution and spread was carried out at physiological maturity and the data pertaining to survey work is presented in Table 1 and Fig. 1.

Fig. 1. Karnataka map showing district-wise average incidence of purple blotch of onion during kharif 2013-14.

Table 1. Survey for purple blotch of onion in Northern parts of Karnataka

District	Taluka	Village name	Stage of the crop	Crop grown condition	Per cent Disease Index (PDI)
Bijapur	Bijapur	Hitnalli	Physiological maturity	Rainfed	25.89
		Jumnal	Physiological maturity	Irrigated	31.16
		Utnal	Physiological maturity	Rainfed	22.43
	Basavana Bagewadi	Telagi	Physiological maturity	Irrigated	33.03
		Golasangi	Physiological maturity	Irrigated	30.86
		Yatnal	Physiological maturity	Rainfed	28.16
Mean					**28.58**
Bagalkot	Hunagund	Hunagund	Physiological maturity	Rainfed	29.12
		Kudalasangam	Physiological maturity	Irrigated	32.76
		Ilkal	Physiological maturity	Irrigated	36.23
	Badami	Badami	Physiological maturity	Rainfed	19.38
		Kerur	Physiological maturity	Rainfed	18.02
		Kerakalmatti	Physiological maturity	Irrigated	33.07
Mean					**28.09**
Gadag	Naragund	Naragund	Physiological maturity	Rainfed	28.80
		Konnur	Physiological maturity	Rainfed	19.68
		Kelakeri	Physiological maturity	Rainfed	29.01
Mean					**25.83**
Dharwad	Navalgund	Navalgund	Physiological maturity	Irrigated	33.12
		Annigeri	Physiological maturity	Rainfed	22.21
		Timmapur	Physiological maturity	Rainfed	26.82
Mean					**27.38**

The survey revealed that prevalence of disease in all locations and disease severity ranged from 18.02 to 36.23 per cent disease index (PDI) in different parts of the districts surveyed. The highest severity (36.23 PDI)of purple blotch was noticed in fields of Ilkal village in Bagalkot district (Fig. 2b), whereas least severity (18.02 PDI) of the disease was recorded at Kerur village in Bagalkot district (Fig. 2a). The average severity of 28.58 per cent disease index was recorded in Bijapur district followed by Bagalkot (28.09 PDI) and Dharwad (27.38 PDI). The lowest disease severity of 25.83 per cent disease index was recorded in Gadag district. The Purple blotch of onion was severe in Bijapur district compared to Gadag district. This could be because of favorable environmental conditions and initial inoculum prevailed in this rejion might have helped in the rapid development of the disease in *kharif*.

Working on survey of *Alternaria* leaf blight and other diseases of onion, Patil and Patil (1991) concluded that it is the most predominant and severe disease in the onion growing areas of Maharashtra. Srivastava *et al.* (1994) in their report on status of field diseases and insect pest of onion in India also indicated that purple blotch incidence was high in both rainy and post-rainy seasons when high humidity prevailed. The present findings are in accordance with the results of Chethana (2000) who conducted survey in Northern parts of Karnataka during *kharif* 1999 also revealed that incidence of purple blotch of onion was noticed in all districts of Northern Karnatakaand recorded highest per cent of disease incidence in Ronihal village (Basavanabagewadi taluk) of Bijapur district and lowest in Wadullur village of Raichurtaluk.

Survey carried during *kharif* 2006 revealed that purple blotch was severe in six districts of Northern Karnataka, *viz.* Dharwad, Bagalkot, Bijapur, Belgaum, Gadag and Haveri. Isolation and morphological studies revealed *A. porri* and *A. alternata* as pathogens (Pramod Kumar, 2007). Survey during *kharif* 2012-13 revealed that purple blotch was found in all parts of Northern Karnataka and was severe in Haveri district (Vinamrata Patil Kulkarni, 2013).

During survey various symptoms of the disease were noticed on leaves and also on bulbs. At initial stages, leaves were with circular to oval water-soaked areas which later on, as the disease progressed, became oblong and a fresh zone of discoloured tissue was formed around the spots. Initially spots were white, but later turned pinkish or purple. The change in colour started from the center and gradually progressed towards the periphery, where it changed into light purplish. The transition of colour was marked by concentric rings clearly visible to the naked eye.The older leaves were more susceptible than younger leaves and were relatively more susceptible when they reach close to bulb maturity. The symptoms of the disease

Fig. 2a. Severity of purple blotch of onion at Kerur (Badami Taluk)

Fig. 2b. Severity of purple blotch of onion at Ilkal (Hunagund Taluk)

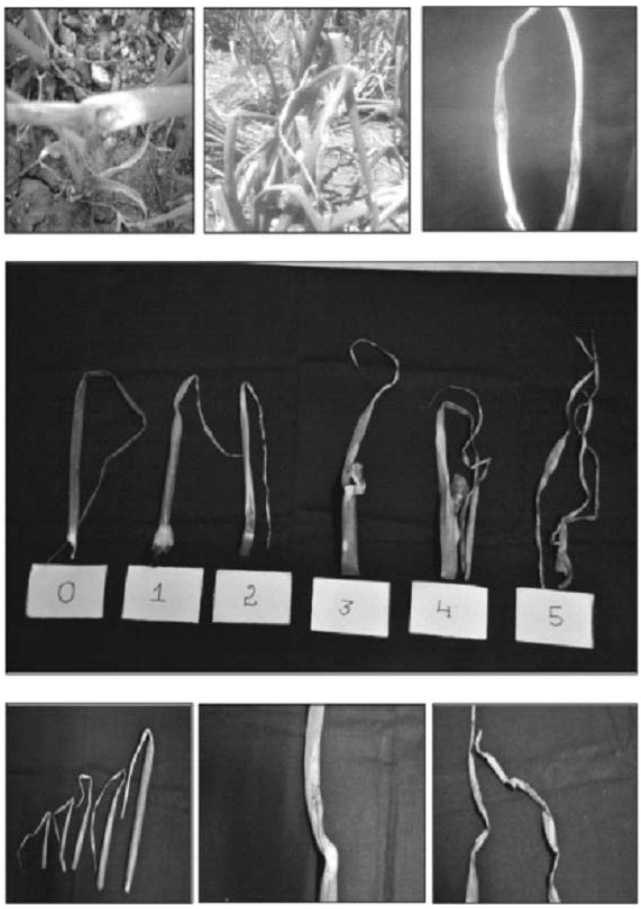

Fig. 3. Symptoms and disease grading of purple blotch of onion

were photographed and are presented (Fig. 3).

Isolation

Isolation of pathogen was made from onion leaves showing typical symptoms of the disease. Leaves with such symptoms were collected for isolation purpose. Standard tissue method was followed after surface disinfection as described in materials and methods and further isolation brought into pure culture by single spore isolation. The pure culture of fungus was obtained after eight days of inoculation which showed whitish growth at initial stage turning later to ash gray color. Such pure culture obtained was again sub cultured on potato dextrose agar slants and kept in the refrigerator at 5°C for further studies (Fig. 4). Dhiman and Chadha (1986) obtained pure culture of fungus using tissue isolation method and described it as a new technique for inoculum preparation and concluded that spore or conidial suspension is the most effective inoculum.

Identification of pathogen

Identification of fungus was carried out based on the morphological characters of fungus isolated. The fungus in present study produced septate mycelium. Later it produced conidiophores arising singly or in small groups. The conidiophores were straight or flexuous, sometime geniculate, septate, pale or mid brown in color and measured upto 120 µm long and 6-10 µm thick, with one or several conidial scars.

A mature conidiophore usually produced solitary conidium but occasionally it also produced conidia with very short chains, straight or curved, rostrate, beak generally equal to the length of body of conidium, pale brown to mid golden brown in colour. Overall length of conidia ranged from 100-300 µm, 15-20 µm thick inthe broadest part with 7-12 transverse and zero to several longitudinal septa, beak flexuous, pale, 2-4 µm thick and tapering. The typical conidium is photographed and is shown (Fig 4). All these characters agreed with those of *A. porri* described by Cifferi (1930) with minor variation in shape and dimension which may be either, due to host or environmental factor and hence were considered to fall within the limits for species. Chethana (2000), who worked on purple blotch of onion also indicated *A. porrias* the causal agent of the

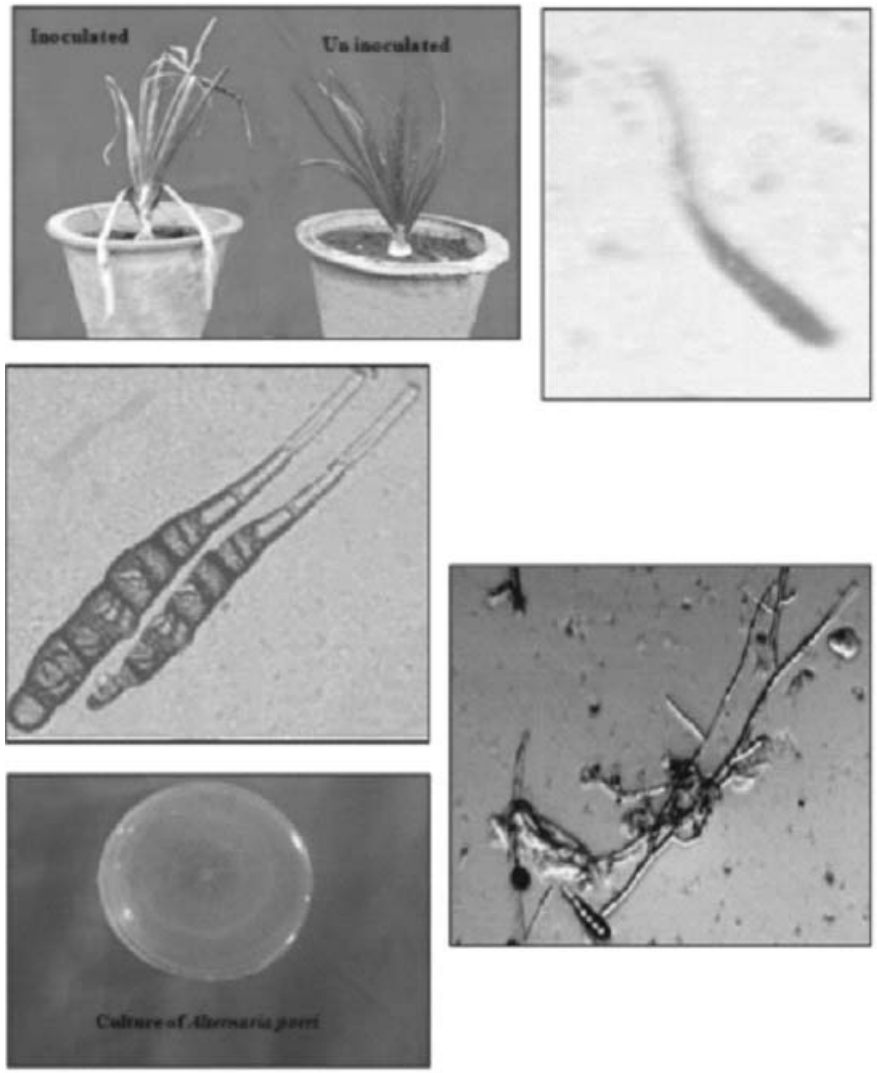

Fig. 4. Photographs showing pathogenecity, conidia and culture of *A. Porri*.

disease and the description is in line with the present investigation.

Pathogenicity test

For proving pathogenicity on host, the pathogens were artificially inoculated on the leaves of onion plants as described in material and methods. After ten days of inoculation, the leaves exhibited symptoms of infection. Earlier infection symptoms could be seen as a small, water soaked lesions which appeared on leaves. Later, these spots started to enlarge and became sunken and purplish in color, with yellow halo. However, this complete expression of the disease symptoms was clearly noticed after 60 days of inoculation. The typical symptoms like purplish zonate spots were noticed on leaves of the artificially inoculated plants. The symptoms were photographed and are presented (Fig. 4).

In the present study, symptoms of disease inoculation technique were found to be in agreement with typical symptoms of the disease described earlier by many workers (Ponnappa, 1974; Utikar and Padule, 1980; Patil and Patil, 1992; Chetana, 2000) who proved pathogenicity of onion by spraying conidial suspension on the host surface. The pathogen was re-isolated from such leaves and the morphological character of the re-isolated organism was compared with the original culture of the pathogen which was similar in all respects. Hence, the causal agent of the disease was confirmed as *Alternaria porri* (Ellis) Cif.

REFERENCES

Chethana B S. 2000, 'Studies on *Alternaria* leaf blight of onion (*Allium cepa* L.)'. M.Sc. (Agriculture) Thesis. University of Agricultural Science Dharwad (India).

Cifferi R. 1930, Phytopathological survey of Santo Domingo.

1925-1929. *J. Porto Rico.*, Department of Agriculture (USA). **14**: 5-44.

Dhiman J S and Chadha M L. 1986, Studies on artificial creation of purple blotch epiphytotics in onion for identifying source of resistance. *Veg. Sci.,* **13**: 293-99.

Marmath K K, Giri P, Sharma S, Taj G and Kumar A. 2013, In-silico interaction studies of *Alternaria brassicae* toxin destruxin B and potential partners of MAPK4 cascade. *International J Agric Environ Bio-tech* **l6**(2): 203-10.

Nolla J A B. 1927, A new *Alternaria* Disease of onions *(Allium cepa).* Phytopathol **17**: 115-37.

Patil A O and Patil B C. 1992, Leaf blight of onion caused by *Alternariastate* of *Pleosporainfectoria. J. Maharashtra Agric. Uni.* **17**: 353.

Patil A O and Patil B C. 1991, Varietal reaction against Alternarialeaf blight of onion (*Allium cepa* L.). *Maharashtra J. Hort.* **5**: 70-71.

Ponnappa K M. 1974, Leaf blight of onion *(Allium cepa)* caused by *Alternariacepulae* Ponnappa and Deshpande, *BlihefteZur Nova Hadwigia.* **47**: 547-64.

Pramod Kumar T. 2007, Biological Management of *Alternaria* blight of onion. M.Sc. (Agricultural) Thesis. Uni. Agric. Sci. Dharwad, Karnataka (India).

Sharma S R. 1986, Effect of fungicidal sprays on purple blotch and bulb yield of onion. *Indian Phytopath.* **39**: 78-82.

Srivastava P K, Bharadwaj B S and Gupta P P. 1994, Status of field diseases and selected pests of onion in India. *News letter Nation. Hort. Res. Devel. Found.* **14**(2): 11-14.

Sudarshan Rao A N. 1975, Survey, surveillance and forecast in plant protection. *Kissan World* **2**: 43-44.

Utikar P G and Padule D N. 1980, A virulent species of *Alternaria* causing leaf blight of onion, *Indian Phytopath* **33**: 335-336.

Vinamrata Patilkulkarni G. 2013, Organic management of purple blotch of onion caused by *Alternaria porri* (ellis) cif. M.Sc. (Agric.) Thesis. Uni. Agric. Sci. Dharwad. Karnataka (India).

Yadav P M, Rakholia K B, Pawar K B. 2013, Evaluation of bioagents for management of the onion purple blotch and bulb yield loss assessment under field conditions. *The Bioscan* **8**(4): 1295-98.

Short Note

Current Horticulture **4**(1): 61–63, January–June 2016

Effect of IPNM packages on quality parameters of bottle gourd *[Lagenaria Siceraria]*

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An experiment was conducted to study the change in the quality of bottle gourd [Lagenaria siceraria (Mol.) Standl.], under IPNM different packaging materials at progressive farmer' field located at Village-Khajua, Post-Mahsanw, Distriet.- Rewa (Madhya Pradesh) during winter seasons of 2013-14 and 2014-15. The bottle gourd variety cv. Puna Naveen was used The highest total soluble solids (TSS) content, titratable acidity, ascorbic acid contents and lowest moisture content under T11 (100% RDF of NPK +FYM@ 5 tonnes / ha + Vermicompost @10 tonnes / ha) followed by T9 (100% RDF of NPK +FYM@ 5 tonnes / ha + Vermicompost @10 tonnes / ha). The minimum TSS, titratable acidity, ascorbic acid contents and highest moist was recorded T16 (Azospirillum@ 2 kg / ha) during both the years respectively.

Bottle gourd [*Lagenaria siceragia* (mol.) Standl.] belonging to the family Cucurbitace, is cultivated in tropics and subtropics. It can be considered as nutrition rich fruit vegetable. It contains considerable amount of water 96.1 g, carbohydrates 2.5 g, protein 0.2 g, fat 0.1 g, minerals 0.5 g, fibre 0.6 g, riboflavin 0.023 mg, vitamin 'A' 10 IU, Vitamin 'c' 11 mg, calcium 16 mg, Iron 0.4mg, phosphorus 14 mg and energy 12 K cal per 100 g of edible fruit (USDA National Nutrient database, 2016). Externally the pulp is applied as poutice and cooling application to the saved head delirium and also rubbed on the flat of the feat and hands to diminish the effect of heat. It helps against constipation, cough, night blindness and function as antidote against certain positions. Among the cucurbits, bottle gourd (*Lagenaria siceraria* (mol.) standl.) is one of the most important and widely grown vegetable crops of India. The fruits in green and tender stage are used as vegetable and for preparation of sweets, raiyta, pickles and different dishes. The application of pre harvested treatments like organic and inorganic production system may also play important role in improving the fruit quality (Mishra *et al.,* 1999) further attributed that different sources of nutrients such as organic / inorganic and integrated had significant effect on the physiological and biochemical changes in bottle gourd fruits.

The experiment was conducted at progressive farmer, field located at Village-Khajua, Post-Mahsanw, District Rewa (Madhya Pradesh) during winter seasons of 2013-14 and 2014-15. The bottle gourd variety was *cv*. Pusa Naveen. The experiment on the effect of different 16 IPNM treatments consisting of T_1 : normal dose of NPK 120: 60: 60 kg / ha; T_2: FYM@20 tonnes / ha; T_3: vermicompost @ 10 tonnes / ha; T_4: poultry manure @ 5 tonnes / ha; T_5: 50% RDF of NPK + FYM@20 tonnes / ha; T_6: 100% RDF of NPK + FYM@10 tonnes / ha + vermicompost @ 5 kg / ha; T_7 : 50% RDF of NPK + vermicompost @ 2.5 tonnes / ha + poultry manure @ 5 tonnes / ha; T_8 : 100% RDF of NPK + FYM @ 5 tonnes / ha + Azospirillum @ 1 kg / ha; T_9 : 100% RDF of NPK + FYM @ 5 tonnes / ha + vermicompost @ 10 kg / ha; T_{10} : 100% RDF of NPK + FYM @ 5 tonnes / ha + vermicompost @ 2.5 tonnes / ha + Azospirillum @ 1 kg / ha; T_{11} : 100% RDF of NPK + FYM @ 10 tonnes / ha + vermicompost @ 5 tonnes / ha + poultry manure @2.5 tonnes / ha; T_{12} : 100% RDF of NPK + FYM@5 tonnes / ha + vermicompost @ 2.5 tonnes / ha + Poultry manure @ 1.25 tonnes / ha; T_{13} : 50% RDF of NPK + vermicompost @ 10 tonnes / ha; T_{14} : 100% RDF of NPK + vermicompost @ 5 tonnes / ha; T_{15} : 100% RDF of NPK + vermicompost@ 2.5 tonnes / ha; T_{16} : Azospirillum @ 1 kg. The extracted juice in two layered muslin cloth was subjected to measurement of TSS with hand refractometer (0-32%). Vitamin 'C' content of freshly harvested and stored

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Table 1. Effect of different IPNM packages on quality parameters of bottle gourd

Treatment	TSS (°Brix)		Ascorbic acid (mg/100 g of dried flesh of fruit)		Acidity		Moisture content	
	2013	2014	2013	2014	2013	2014	2013	2014
T_1 : Normal dose of NPK 120: 60: 60 kg/ha	2.05	2.31	6.07	6.15	0.34	0.32	98.56	98.73
T_2 : FYM@ 20 tonnes/ha	2.07	2.73	6.09	6.17	0.38	0.37	98.32	98.41
T_3 : Vermicompost@10 tonnes/ha	2.31	3.01	6.39	6.48	0.35	0.34	97.67	97.85
T_4 : Poultrymanure@5 tonnes/ha	2.14	2.48	6.13	6.20	0.37	0.35	97.91	98.25
T_5 : 50% RDF of NPK + FYM @20 tonnes/ha	2.12	2.44	6.11	6.13	0.40	0.39	98.21	98.36
T_6 : 100% RDF of NPK + FYM@10 tonnes ha + vermicompost @5 tonnes/ha	3.01	3.41	7.79	7.83	0.44	0.45	90.15	90.39
T_7 : 50% RDF of NPK + vermicompost@2.5 tonnes ha + poultrymanure@1.25 tonnes/ha	2.27	2.61	6.21	6.30	0.37	0.37	97.80	97.88
T_8 : 100% RDF of NPK + FYM@5 tonnes/ha + Azospirillum@1 kg/ha	2.35	2.77	6.87	6.93	0.41	0.41	97.41	97.45
T_9 : 100% RDF of NPK + FYM@5 tonnes/ha + vermicompost@10 tonnes/ha	3.03	3.72	8.28	8.31	0.46	0.46	88.24	89.16
T_{10} : 100% RDF of NPK + FYM@5 tonnes/ha + vermicompost@2.5 tonnes/ha + Azospirillum@1 kg/ha	2.78	2.98	7.30	7.10	0.42	0.41	93.34	94.14
T_{11} : 100% RDF of NPK + FYM@10 tonnes/ha + vermicompost@5 tonnes/ha + Poultry manure@2.5 tonnes/ha	3.18	3.95	8.69	8.74	0.46	0.47	86.23	87.41
T_{12} : 100% RDF of NPK + FYM@5 tonnes/ha + vermicompost@2.5 tonnes/ha + poultry manure@1.25 tonnes/ha	2.76	3.07	7.27	7.25	0.43	0.42	92.43	92.87
T_{13} : 50% RDF of NPK+ vermicompost@10 tonnes/ha	2.62	2.99	6.52	6.82	0.36	0.34	95.46	95.87
T_{14} : 100% RDF of NPK + vermicompost@5 tonnes/ha	2.64	2.98	6.88	6.89	0.39	0.38	93.54	93.82
T_{15} : 100% RDF of NPK + vermicompost@2.5 tonnes/ha	2.59	2.94	7.17	7.84	0.40	0.39	96.32	97.53
T_{16} : Azospirillum@2 kg/ha	2.04	2.18	6.07	7.06	0.34	0.31	98.56	98.50
SEm	0.03	0.04	0.09	0.10	0.34	0.32	0.84	0.85
CD (P=0.05)	0.09	0.11	0.25	0.28	0.38	0.37	2.44	2.46

fruits was calculated by the reduction of 2, 6 Dichloro-indophenol dye as described by Ranganna (1986).

The highest total soluble solids (TSS) content, titratable acidity, ascorbic acid contents and lowest moisture content under T_{11} (100% RDF of NPK + FYM @ 5 tonnes / ha + vermicompost@10 tonnes / ha) followed by T_9 (100% RDF of NPK +FYM@ 5 tonnes / ha + vermicompost @10 tonnes / ha). The minimum TSS, titratable acidity, ascorbic acid contents and highest moist was recorded T_{16} (Azospirillum@ 2 kg / ha) during both the years respectively (Table 1). The present results are in accordance with the earlier findings of *Mostakin et al.* (2000). Higher content of TSS, ascorbic acid, and percentage of acidity and lowest percentage of moisture content was observed in fruit where plot receiving 100% RDF of NPK + FYM@ 10 tonnes / ha + vermicompost @ 5 tonnes / ha + Poultry manure @ 2.5 tonnes / ha) under treatment T_{11}. The organically managed crop have usually higher TSS, ascorbic acid and acidity than the conventional fertilized crop because when a plant exposed with more N, it increases protein production and reduces carbohydrates synthesis.

Since TSS, ascorbic acid and acidity is synthesized from carbohydrates, its levels are also reduced. In case of organically managed soil plants is generally exposed with comparatively lower amount of N and several plant nutrients are released slowly over time. Therefore, organic crop would be expected to contain higher value of these quality traits and carbohydrates and less protein. Furthermore, soil micro-organism affects soil dynamics and plant metabolisms and ultimately results in plant composition and nutrition quality. *Worthington* (2001) and *Bahadur et al.*, (2003) are also of the similar view. Increased in ascorbic acid and TSS content of fruit in these treatments could be attributed to combined application of organic, inorganic fertilizers along with the bio-fertilizers (Azospirillum) which helped in better uptake of NPK nutrients including micronutrients which inturn influence the quality traits in bottle gourd.

REFERENCES

Bahadur A, Singh J, Upadhyay A K and Singh K P. 2003. Effect of organic manures and biofertilizers on grwoth, yield and quality attributes of broccoli (*Brassica oleracea* L. var. italica Plenck.) *Veg. Sci.* **30**(2): 192-94.

Mishra O P, Verma O P and Sharma A K. 1999. Sustainable agriculture a need of the day. *Farmer and Pariliament* **34**: 9-10.

Mostakin M, Ahmad, M F and Singh, D B. 2000. Effect of Azotobacter and spacing onion yield. *App. Bio. Res* **2**(1-2): 35-37.

Nutrient content in bottle gourd, USDA National Nutrient database, 2016.

Ranganna S. 1986. *Hand Book of Analysis and Quality Control for Fruit and Vegetable Products*. Tata Mc Graw-Hill Publishing Company Ltd, New Delhi, pp. 9-10.

Worthington, V. 2001. Nutritional quality of organic versus convention fruits vegetables and grains. *J. Altern Compl. Med.* **7**(2): 161-73.

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