Evaluation of Garlic Genotypes Under Farmer's Field Condition of Jumla, Nepal

ABSTRACT

On-farm trial on different garlic genotypes selected from Advance Yield Trial was carried out at Patrashi Rural Municipality of Jumla (2430 masl) for two consecutive years 2017/18 and 2018/19 to evaluate garlic genotypes suitable for the high hills of Karnali region of Nepal. Twelve different garlic genotypes: ARM 01, ARM 02, ARM 03, ARM 04, ARM 05, ARM 07, ARM 09, Mugu Local, Kathmandu Local, Chinese, Holeri and Malikabota were tested on Randomized Complete Block Design (RCBD). Each treatment was replicated three times. Compost was used at 20t/ha at the time of land preparation. There was no use of chemical fertilizer. Selected cloves of garlic were planted with 15 cm x 10 cm spacing of row to row and clove to clove respectively. Planting and harvesting were done on the third week of September and first week of July respectively. Tested genotypes differed significantly for vegetative as well as yield parameters. The genotype ARM01 produced the bulbs with the highest (50.49 mm) diameter followed by ARM 04 (45.14 mm), Kathmandu Local (34.52 mm) and Chinese (33.44 mm) respectively. The highest bulb yield (29.73 t/ha) was recorded from genotype ARM 01 followed by Kathmandu Local (19.4 t/ha), ARM 04 (18.51 t/ha) and Chinese (17.59 t/ha). Based on the average result of both years, genotypes ARM 01, Kathmandu Local, ARM 04, and Chinese showing above mentioned average productivity ranging from 17.59 t/ha to 29.73 t/ha, were identified as the promising genotypes for commercial cultivation in the high hills of Karnali region of Nepal.

Key words: *On-farm Trial, Genotypes, Cloves, Yield*

1. INTRODUCTION

"Garlic (*Allium sativum* L.) belongs to family Alliaceae and it is second most widely cultivated bulb crop after onion in the world and is propagated vegetatively" [1]. "It is monocotyledonous diploid (2n = 16) plant with rigid asexual life cycle, primarily grown for its cloves and mostly used as a food flavouring condiment. The fresh leaves and cloves of garlic are widely used raw, cooked in culinary globally and It is also consumed as powder form after processing. Garlic has multiple uses; it is used for its aroma, as well as a traditional medicine" [2]. "In tropical areas, consumption of immature bulbs for salad is also popular" [3]. Garlic is an indispensable commodity of Nepalese kitchen. Its demand and consumption are increasing day by day however the production is almost stagnant far behind the domestic demand. Nepal is a net spice importing country and garlic is being imported every year from neighboring countries in a huge amount, accounting a large sum of money outflow from the economy. Though garlic cultivation started years ago, lack of quality genotypes and appropriate package of practices (POP) demonstrates little applicability in commercial cultivation in Nepal. None of the garlic genotypes has been recommended, registered or released from NARC for cultivation in Nepal. There is subsistence cultivation of few land races and few other exotic germplasms introduced from adjoining country at different dates. Very few studies have been carried out in garlic.

"None of the variety is characterized and standard farming technology is yet to be developed. Garlic is grown globally but China is leading country in production followed by India, Bangladesh, South Korea, Egypt and Spain, sequentially and Nepal stands in 22nd position" [4]. In Jumla, cultivated area under garlic is 55 hectare (ha) and the production is 330 ton (t) with productivity of 6 t/ha. Similarly, in Nepal, garlic has been cultivated under an area of 9784 ha with a total production of 72490 t [5] with productivity of 7.41 t/ha. However, the productivity of garlic in Jumla is 6 t/ha whereas the national productivity is 7.41 t/ha which is below than the national average. Similarly, total area occupied by garlic in Jumla is only 55 ha and production is 330 t. This suggests that identifying suitable genotypes and a package of practices

could lead to both vertical and horizontal expansion of garlic cultivation in this region. The demand for spices peaks in October due to the festive season, and garlic is in short supply. However, the agro-climatic conditions in high hills allow for production and supply to low-lying areas during this peak period, which commands a high price. Furthermore, because it is non-perishable, it can be easily stored under normal conditions with minimal postharvest losses.

In the past, little work on garlic was done, almost entirely in the terai and mid hills. In spite of great production potential of high hills, garlic imported from Tibet to Kathmandu reaches the Karnali region via airways and vehicle incurring very high cost. This is a significant misfortune for the region. There are several land races and a few exotic germplasms in cultivation, and there is a lot of variation between them. Improvements in qualitative and quantitative traits can be achieved through careful genotype selection and improved crop husbandry. In the context of no recommendation, registration or release of garlic variety, an elaborated study of the same crop is necessary for germplasm selection, improvement and release as a new variety along the appropriate POP for garlic commercialization. This may contribute to mass production leading to import substitution and export promotion of the same commodity. To address the above-mentioned issues, this study was carried out.

2. MATERIALS AND METHODS

On-farm experiment was conducted by Horticulture Research Station (HRS), Rajikot, Jumla at Urthuchautara settlement of Patrashi Rural Municipality in Jumla district. Experimental area is situated at 29°18'10"N and 29°13'42"E with the altitude of 2430 meters above mean sea level; with temperate climate. August-September is the main planting season of garlic in Jumla, which soil is sandy loam in nature. Maximum and minimum average temperature of garlic growing season in 2017/18 were 17 ^{0}C to 27 ^{0}C and -6 ^{0}C to 17 ^{0}C. Total rainfall during this year was 468 mm [6]. Similarly, during 2018/19 maximum and minimum average temperature were 11 ^{0}C to 26 ^{0}C and -5 ^{0}C to 16 ^{0}C and total rainfall was 715 mm [7].

Twelve different garlic genotypes collected from different districts of Nepal were tested under farmer's field trial after on station trial during 2017/18 and 2018/19. The farmer's field trial was conducted at Urthuchautara settlement of Patrashi Rural Municipality, Jumla. Germplasms: ARM 01, ARM 02, ARM 03, ARM 04, ARM 05, ARM 07, ARM 09, Mugu Local, Kathmandu Local (Ktm local), Chinese, Holeri and Malikabota were tested. The plot size was assigned 0.75 m^2 (1m x 0.75 m). The plots were fertilized with 20 t compost/ha. There was no use of chemical fertilizer. Selected cloves were planted in 15 cm x 10 cm spacing. Randomized Complete Block Design (RCBD) was used and each treatment (genotype) was replicated three times. Planting and harvesting were done on the third week of September and first week of July respectively. Observations were collected for plant height (cm), length of leaf (cm), width of leaf (cm), leaf number per plant, bulb diameter (mm), bulb height (mm), bulb color, number of cloves per bulb, number of bulbs, total yield and taste of cloves (pungency level and bitterness). Plant height was measured from the base of the leaf sheath to the tip of the longest leaf of five randomly selected plants and mean was calculated in cm.

The number of leaves per plant was counted from five randomly selected plants. The mean number of leaves was calculated by dividing total number of leaves observed from five plants by 5. Length of leaf was measured from the base to top of the leaf of five randomly selected leaves in centimeter and mean was calculated. Diameter at the widest part of 5 randomly selected bulbs was measured in millimeter and then average diameter of each bulb was calculated. Height of bulb of five randomly selected bulbs was measured from base to tip of the bulb in millimeter and then average diameter of each bulb was calculated. The cloves were counted from five plants and their average was taken as a number of cloves per plant. The yield of bulb per plot was taken in kilogram (kg) by harvesting all the bulbs of each plot

after removing the roots and pseudo stem. Per plot yield of bulb was converted into yield per ha and expressed in t. The data recorded for on-farm varietal trial were managed in a spreadsheet and analyzed using Genstat version 18 software for windows [8]. Analysis of variance (ANOVA) was used to determine statistically significant differences between means. The data for 2017/18 and 2018/19 were analyzed separately, and the results of two years are presented. The significant mean data of observed variables were separated using Duncan's Multiple Range Test (DMRT) at 5% level of probability. Least significant differences (LSD) were determined for all significant data. Coefficient of Variation (CV), the ratio of standard deviation to mean which indicate the quality of sample data on estimating the dispersion of population, is expressed in percentage. Lower the value of CV, the more precise the estimate of mean.

3. RESULTS AND DISCUSSION

Most of the recorded parameters (plant height, number of leaves per plant, length of leaf, width of leaf, bulb diameter, bulb height, number of cloves per bulb, number of bulbs per hectare and bulb yield) were found statistically significant among the tested garlic genotypes.

3.1 Plant height

The tallest plants (81.1 cm) were measured from genotype ARM 01 followed by ARM 04 (72.6 cm), Chinese (59.5 cm), Malikabota (57.37 cm), Holeri (54 cm) and Kathmandu Local (52.27 cm) whereas the dwarfest (45.6 cm) plant height was measured from genotype ARM 02 followed by ARM 05 (47.5 cm) and ARM 03 (47.73 cm). Statistically significance difference of genotypes for plant height was due to genetic difference **(Table 1).** Some researchers [9, 10, 11] reported significant variation for plant height due to the difference between genotypes i.e. variation for plant height due to varietal difference. Similar types of results were reported by different researchers [12, 13, 14, 15].

3.2 Number of leaves per plant

Number of leaves per plant were counted maximum from ARM 01 (8.43) followed by ARM 04 (7.83), Holeri (7.5), ARM 07 (7.43), Malikabota (7.43) and Kathmandu Local (7.2) whereas the minimum (5.53) number of leaves per plant were counted from Chinese followed by ARM 05 (6.63) and ARM 03 (6.6) **(Table1).** Significant variation for number of leaves per plant was due to the difference between genotypes. Similar types of results were reported by researchers [16, 17].

Table 1: Effect of garlic genotypes on plant height and number of leaves per plant in On-farm Varietal Trial at Patrashi Rural Municipality, Jumla during 2017/18 and 2018/19

SN	Genotypes	Plant height (cm)			No. of leaves per plant		
		2017/18	2018/19	Average	2017/18	2018/19	Average
1	ARM-01	56.6 a	105.6 a	81.1 a	8.87 a	8 ab	8.43 a
2	ARM-02	43.87 c	47.33 f	45.6 f	6.53 c	7.06 bcd	6.8 c
3	ARM-03	45.27 c	50.2 ef	47.73 ef	6.67 c	6.53 cd	6.6 c
4	ARM-04	56 ab	89.2 b	72.6 b	8.53 ab	7.13 abcd	7.83 ab
5	ARM-05	46.2 c	48.8 f	47.5 ef	7 c	6.27 d	6.63 c
6	ARM-07	53.53 abc	52.67 def	52.1 de	7.13 bc	7.73 ab	7.43 bc
7	ARM-09	46.08 c	50.73 ef	48.77 ef	6.93 c	7.2 abcd	7.06 bc
8	Mugu Local	49.33 abc	55.67 def	52.5 de	7.06 c	7.2 abcd	7.13 bc
9	Kathmandu Local	44.73 c	59.8 cde	52.27 de	6.53 c	7.87 ab	7.2 bc
10	Chinese	57.13 a	61.87 cd	59.5 c	6.4 c	4.67 e	5.53 d
11	Holeri	51.47 abc	56.53 def	54 cde	7.46 abc	7.53 abc	7.5 bc
12	Malikabota	48 bc	66.73 c	57.37 cd	6.67 c	8.2 a	7.43 bc
	Mean	49.74	62.09	55.92	7.15	7.12	7.13
	F test	**	**	**	*	**	**
	CV (%)	8.8	8.7	6.2	11.1	8.1	7.3
	LSD (0.05)	7.39	9.15	5.84	1.34	0.97	0.88

Note: NS=Non Significant *= Significant at ***P***<0.05 **=Significant at ***P***<0.001 LSD=Least Significant Difference CV= Coefficient of Variation

3.3 Length of leaf

Garlic genotype Chinese produced the longest leaves (23.87 cm) followed by ARM 01 (23.08 cm), ARM 04 (21.68 cm), Malikabota (20.73 cm), Mugu Local (19.9 cm), Holeri (19.52 cm) and Kathmandu Local (19.04 cm) and ARM 02 produced the shortest leaves (16.59 cm) followed by ARM 09 (16.91 cm) and ARM 03 (17.03 cm) **(Table 2).** The longest leaves were observed from genotypes with the maximum diameter and height of bulbs. This is because large-sized bulbs have more reserve food materials which ultimately supports to produce larger leaves as compared to small bulbs. Similar type of result was also mentioned by [18, 19] different researchers who reported that availability of more food reserves in bulbs allowed vigorous in growth and development.

3.4 Width of leaf

Width of leaves were maximum (2.24 cm) in genotype ARM 01 followed by ARM 04 (2.05 cm), Kathmandu Local (1.06 cm), Chinese (1.05 cm) and Malikabota (1.05 cm) whereas the narrowest leaves (0.87 cm) were recorded from ARM 02 and Holeri followed by Mugu Local (0.90 cm), ARM 03 (0.92 cm) and ARM 05 (0.92 cm) **(Table 2).** Researchers reported [16, 20] significant variation in width of leaves was due to the difference between genotypes.

Table 2: Effect of garlic genotypes on Length of leaf and width of leaf in On-farm Varietal Trial at Patrashi Rural Municipality, Jumla during 2017/18 and 2018/19

SN	Genotypes	Length of leaf (cm)			Width of leaf (cm)		
		2017/18	2018/19	Average	2017/18	2018/19	Average
1	ARM-01	15.94 ab	30.22 a	23.08 ab	1.97 a	2.51 a	2.24 a
2	ARM-02	10.7 e	22.48 e	16.59 f	0.82 c	0.92 b	0.87 b
3	ARM-03	11.73 de	22.41 e	17.07 ef	0.85 c	1 b	0.92 b
4	ARM-04	14.73 bc	28.63 abc	21.68 abc	1.73 b	2.37 a	2.05 a
5	ARM-05	11.79 de	24.07 cde	17.93 def	0.89 c	0.95 b	0.92 b
6	ARM-07	12.88 cde	22.93 de	17.9 def	0.9 c	1.14 b	1.02 b
7	ARM-09	11.11 e	22.7 de	16.91 ef	0.81 c	1.1 b	0.95 b
8	Mugu Local	12.06 de	27.74 abc	19.9 cd	0.84 c	0.96 b	0.90 b
9	Kathmandu Local	10.7 e	27.37 abcd	19.04 cdef	0.89 c	1.23 b	1.06 b
10	Chinese	17.96 a	29.89 a	23.87 a	0.98 c	1.11 b	1.05 b
11	Holeri	14.07 bcd	24.96 bcde	19.52 cde	0.81 c	0.94 b	0.87 b
12	Malikabota	12.02 de	29.44 ab	20.73 bc	0.83 c	1.28 b	1.05 b
	Mean	12.97	26.07	19.52	1.03	1.29	1.16
	F test	**	**	**	**	**	**
	CV (%)	10.1	9.8	7.4	9.9	18	11.3
	LSD (0.05)	2.23	4.34	2.45	0.17	0.39	0.22

Note: NS=Non Significant *= Significant at ***P***<0.05 **=Significant at ***P***<0.001 LSD=Least Significant Difference CV= Coefficient of Variation

3.5 Diameter of bulb

The result for the mean bulb diameter of genotype showed highly significant (p<0.01) difference. The data indicated that the genotype ARM01 produced the bulbs with the highest (50.49 mm) diameter followed by ARM 04 (45.14 mm), Kathmandu Local (34.52 mm) and Chinese (33.44 mm) respectively. The lowest diameter of bulb (29.06 mm) was obtained from ARM 02 followed by ARM 05 (29.18 mm) **(Table 3).** Similar type of variation of bulb diameter of different genotypes was also reported by Researcher [21].

3.6 Height of bulb

The result for the mean bulb height of the tested genotype also showed highly significant (p<0.01) difference. The highest bulb height (42.17 mm) was recorded from genotype ARM01 followed by ARM 04 (39.97 mm), ARM 03 (39.1 mm) and ARM 07 (35.4 mm) respectively. The lowest height of bulb (29.26 mm) was obtained from Malikabota followed by ARM 05 (31.99 mm) **(Table 3).** Statistically, significant difference for bulb height was due to the difference between genotypes [16].

Table 3: Effect of garlic genotypes on bulb diameter and bulb height in On-farm Varietal Trial at Patrashi Rural Municipality, Jumla during 2017/18 and 2018/19

SN	Genotypes	Bulb diameter (mm)			Bulb height (mm)		
		2017/18	2018/19	Average	2017/18	2018/19	Average
1	ARM-01	41.98 a	59 a	50.49 a	42.33	42 a	42.17 a
2	ARM-02	22.9 c	35.22 bc	29.06 d	36.8	32 bc	34.4 bcde
3	ARM-03	31.4 bc	34.67 bc	33.03 cd	43.53	34.67 b	39.1 abc
4	ARM-04	34.4 b	55.89 a	45.14 b	39.83	40.11 a	39.97 ab
5	ARM-05	25.47 c	32.89 c	29.18 d	34.87	29.11 c	31.99 de
6	ARM-07	25.67 c	38.44 bc	32.06 cd	36.8	34 bc	35.4 bcd
7	ARM-09	23.63 c	36.33 bc	29.98 cd	32.67	33.89 bc	33.28 de
8	Mugu Local	28.2 bc	38.33 bc	33.27 cd	35	31.11 bc	33.06 de
9	Kathmandu Local	29.16 bc	39.89 b	34.52 c	34.4	32.22 bc	33.81 cde
10	Chinese	27.11 bc	39.78b	33.44 cd	37.6	32.67 bc	35.13 bcd
11	Holeri	26.97 bc	38 bc	32.48 cd	39.67	31.67 bc	35.67 bcd
12	Malikabota	23.55 c	40.33 b	31.94 cd	27.07	31.11 bc	29.26 e
	Mean	28.37	40.73	34.55	36.71	33.82	35.27
	F test	**	**	**	NS	**	**
	CV (%)	15.5	8.3	6.9	15.7	7.9	8.4
	LSD (0.05)	7.43	5.74	4.04		4.54	5

Note: NS=Non Significant *= Significant at ***P***<0.05 **=Significant at ***P***<0.001 LSD=Least Significant Difference CV= Coefficient of Variation

3.7 Number of cloves per bulb

Statistically highly significant (p<0.001) difference on number of cloves per compound bulb was found among the genotypes. The maximum number of cloves per bulb was obtained from Malikabota (10.3) followed by Mugu Local (10.06). Lowest number of cloves (3.7) was recorded from genotype ARM 01 followed by ARM 04 (3.9) **(Table 4).** Generally, the lowest number of cloves per bulb was recorded from medium to large sized cloves. Similar observations were made by different researchers [17, 10, 21] who reported that clove size had significant effects on the number of cloves per bulb.

3.8 Number of bulbs

Number of bulbs per ha were counted maximum from genotype ARM 07 (546667) followed by ARM 03 (533333), ARM 05 (468889), Chinese (468889) whereas lowest (36000) from Mugu Local followed by ARM 04 (386667). Total number of bulbs was related to mergence of cloves. That means higher number of bulbs per ha means more emergence percentage of that genotype of garlic **(Table 4).**

Table 4: Effect of garlic genotypes on no. of cloves per bulb and no. of bulbs per ha in On-farm Varietal Trial at Patrashi Rural Municipality, Jumla during 2017/18 and 2018/19

SN	Treatment	No. of cloves per bulb			No. of bulbs per ha		
		2017/18	2018/19	Average	2017/18	2018/19	Average
1	ARM-01	3.26 cde	4.13 e	3.7 f	644444 a	448889	546667 a
2	ARM-02	2.2 de	8.67 cd	5.43 cdef	360000 cd	395556	377778 bc
3	ARM-03	1.86 e	7.73 d	4.8 def	635556 a	431111	533333 a
4	ARM-04	3.2 cde	4.6 e	3.9 ef	408889 bcd	364444	386667 bc
5	ARM-05	4.26 bcde	8 cd	6.13 bcd	533333 ab	404444	468889 ab
6	ARM-07	3.13 cde	8.67 cd	5.9 bcde	475556 bc	377778	426667 bc
7	ARM-09	4.2 bcde	10.46 bc	7.33 bc	417778 bcd	444444	431111 bc
8	Mugu Local	8.33 b	11.8 b	10.06 a	404444 bcd	315556	360000 c
9	Kathmandu Local	5.2 bcd	10.33 bc	7.76 b	448889 bc	360000	404444 bc
10	Chinese	6.47 ab	9.06 cd	7.76 b	440000 bcd	497778	468889 ab
11	Holeri	5.13 bcd	8.93 cd	7.03 bc	426667 bcd	422222	424444 bc
12	Malikabota	6.13 abc	14.46 a	10.3 a	293333 d	506667	400000 bc
	Mean	4.45	8.91	6.68	457407	414074	435741
	F test	**	**	**	**	NS	**
	CV (%)	34.9	14.9	16.5	17.4	16.3	12.2
	LSD (0.05)	2.63	2.24	1.87	134974		89986

Note: NS=Non Significant *= Significant at ***P***<0.05 **=Significant at ***P***<0.001 LSD=Least Significant Difference CV= Coefficient of Variation

3.9 Bulb Yield and characteristics

Different genotypes of garlic showed statistically highly significant *(p<0.001)* differences on yield of bulb per ha. ARM 01 gave the highest bulb yield (29.73 t) per ha followed by Kathmandu local (19.4 t/ha), ARM 04 (18.51 t/ha) and Chinese (17.59 t/ha). The lowest bulb yield per ha (12.34 t/ha) was obtained from ARM 02 followed by ARM 09 (12.58 t/ha) and Holeri (13.09 t/ha) (Table 5). Different researchers [18, 9, 20, 21] reported similar type of yield variation of different genotypes of garlic due to genetic differences.

Color of bulb was found white in ARM 02, ARM 05 and Holeri. Light violet color was observed in Chinese, ARM 03, ARM 07 and ARM 09. Violet color was recorded in Malikabota, Mugu local and Kathmandu local. Similarly, creamy color was observed in ARM 01 and ARM 04.

ARM 01 and ARM 04 were found slightly bitter and more pungent, ARM 09, Malikabota and Kathmandu local were found more pungent. Similarly, Chinese, ARM 07 and ARM 08 were pungent where as ARM 02, ARM 03, ARM 05, Mugu local, were less pungent.

Table 5: Effect of garlic genotypes on bulb yield and and bulb skin color in On-farm Varietal Trial at Patrashi Rural Municipality, Jumla during 2017/18 and 2018/19

SN	Treatment	Bulb yield (t/ha)			Bulb skin color
		2017/18	2018/19	Average	
1	ARM-01	23.15 a	36.31 a	29.73 a	Creamy
2	ARM-02	13.07 cde	11.61 d	12.34 e	White
3	ARM-03	19.34 abc	11.76 d	15.55 bcde	Light Violet
4	ARM-04	17.91 abcd	19.11 b	18.51 bc	Creamy
5	ARM-05	19.02 abc	10.87 d	14.94 bcd	White
6	ARM-07	18.72 abcd	12.03 cd	15.37 bcd	Light Violet
7	ARM-09	12.51 de	12.66 cd	12.58 e	Light Violet
8	Mugu Local	17.64 abcd	12.09 cd	14.86 bcde	Violet
9	Kathmandu Local	19.87 ab	18.93 b	19.4 b	Violet
10	Chinese	17.86 abcd	17.32 bc	17.59 bcd	Light Violet
11	Holeri	13.69 bcde	12.5 cd	13.09 de	White
12	Malikabota	10.36 e	18.25 b	14.3 cde	Violet
	Mean	16.93	16.12	16.52	
	F test	**	**	**	
	CV (%)	19.4	17.8	15.1	
	LSD (0.05)	5.57	4.85	4.23	

Note: NS=Non Significant *= Significant at ***P***<0.05 **=Significant at ***P***<0.001 LSD=Least Significant Difference CV= Coefficient of Variation

4. CONCLUSION

Based on the results of Advance Yield Trial carried out at the station, twelve garlic genotypes were selected and tested under farmer's field during 2017/18 and 2018/19. During both of the years, genotypes ARM 01, Kathmandu local, ARM 04 and Chinese performed better with respect to productivity ranging from 17.59 t/ha to 29.73 t/ha which is two to three times higher than the national productivity (7.41 t/ha). This result clearifies their genetic potentiality to perform better under high hill conditions and uplift the stagnant low productivity of the garlic. Moreover, these genotypes performed better with respect to vegetative parameters too. All these vegetative parameters are associated with photosynthetic efficiency which ultimately resulted in more yield. Though the yield potentiality of ARM 01 and ARM 04 is high, these genotypes may gather lesser consumer appeal for table purpose due to slight bitterness present on them. However, they might be the best fit for industrial purpose owing to the lesser number of large sized cloves which go quite easy on processing. Kathmandu local and Chinese are better for kitchen/spice purpose and are promising garlic genotypes for Jumla and similar locations of Karnali Region of Nepal.

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COMPETING INTERESTS

Authors declare that there is no conflict of interest related to the publication of this manuscript.

AUTHORS' CONTRIBUTIONS

This trial was carried out in close collaboration among the authors. All the authors read the first draft paper, commented, suggested and approved the final paper for submission.

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