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# Effect of Nitrogen on the Growth and Yield of Carrot (Daucus carota L.)

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## ABSTRACT

This study illustrates how nitrogen levels and carrot variety selection influence growth and production. The experiment was conducted in Kashiadanga, Rajpara Thana, Rajshahi, Bangladesh, from October 2019 to March 2020, evaluating the effects of four nitrogen levels (0 kg N/ha, 70 kg N/ha, 140 kg N/ha, and 210 kg N/ha) on the growth and yield of two carrot varieties, New Kuroda and Pusa Kesar. The study was conducted with three replications, employing a Randomized Complete Block Design as the experimental setup. Significant variations in carrot growth and yield were observed across the treatments. Notably, the Pusa Kesar cultivar and 140 kg N/ha demonstrated superior performance. The application of 140 kg N/ha with Pusa Kesar resulted in the highest values for various growth parameters, including plant length (80.00 cm), root length (23.03 cm), diameter (36.04 mm), dry root weight (23.54 g per 100 g of fresh root weight), the number of leaves (12.44) at 90 days after sowing (DAS), and yield (27.03 t/ha). Nitrogen dose and variety combinations had considerable effects on growth and yield, underscoring the necessity of selecting appropriate varieties and applying judicious amounts of nitrogen fertilizer for satisfactory carrot production in Bangladesh and elsewhere.

# Introduction

Carrots (*Daucus carota* L.) originated in Asia. Carrots are members of the Apiaceae family, the genus Daucus, and the species Carota, which has chromosome number 2n=18. Carrots are annuals for root production and biennials for seed production. Carrots have a 'Compound Umbel' flower, and the edible component is a modified

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root (conical shape) that grows in soil (Pal et al., 2019). This ancient crop is widely grown in Asia, Europe, North Africa, North and South America (Thompson and Kelly, 1957). Carrot root contains moisture 86%, protein 0.9 g, carbohydrate 10.6 g, fat 0.2 g, fiber 1.2 g, energy 48 kilo calorie, mineral 1.1 g, iron 2.2 mg, carotene 1890 mg, thiamine 0.04 mg, riboflavin 0.02 mg, niacin 0.5 mg, vitamin-C 3 mg, folic acid 15 mg, calcium 80 mg, and phosphorus 30 mg per 100 g of edible portion (Bose et al., 2003). Carrots are an essential and

popular root vegetable used in salads and cooked dishes (Kadam et al.,1998), preserved pickles, preserves, sweets (particularly Gajar halwa), carrot powders, kanji, an appealing drink, and so on. Aside from being a food, carrots have medicinal value since they improve resistance to blood and eye illnesses (Kumawat et al., 2018).

In Bangladesh, carrots are an essential component of the agricultural landscape, contributing significantly to both subsistence and commercial farming. The area of carrot cultivation in Bangladesh is 5085 acres with a total production of 19246 MT (BBS, 2020).

Comparatively, the yield of carrots in our country is very low. Successful growing of highquality roots is attributed to a variety of factors, one of which is the prudent application of fertilizers (Moniruzzaman et al., 2013). Because fertilization determines crop quality and quantity as well as the physical, biological, and physiochemical characteristics of soil, as well as the quality of air, ground, and surface water, it is crucial to crop productivity. From the perspective of the producer, fertilization has a major impact on the cost of production (Sikora et al., 2020). One of the most crucial elements that plants use to restrict how much they produce is nitrogen. The type of nitrate fertilizer applied, as well as nitrogen rates, variety, environment, harvesting date, and other agronomical parameters, all have an impact on nitrate accumulation (Boskovic-Rakocevic et al., 2012). Significant differences in nitrogen uptake may be pointed to various soil types, nutrient concentrations, climates, and welldeveloped root systems that allow plants to effectively absorb nitrogen from the soil (Warncke, 1996). In the same way, variety is crucial to the production of carrots. There is no suggested carrot variety in Bangladesh. Carrot seeds are produced by the majority of seed firms worldwide to fit their specific climates; if the seeds are planted without first undergoing an adaptation test, producers risk financial loss (Osei et al., 2020). Here, carrot seed output is significantly influenced by varietal selection. The yield of carrots per hectare can be greatly increased by employing high-yielding variety seeds. The gap in understanding the specific effects of nitrogen on carrot cultivation, particularly in conjunction with varietal diversity, is a critical area requiring attention. While nitrogen's influence on plant growth and yield is welldocumented across various crops, such as cereals and legumes, research dedicated specifically to carrots is limited. Additionally, although studies have identified diverse carrot varieties with varying yield and quality attributes, few have systematically explored how these varieties interact with different nitrogen levels. Consequently, there is a notable research gap in comprehensively understanding the interplay between nitrogen levels and varietal diversity in carrot production, hindering the development of tailored fertilization strategies and cultivar recommendations essential for sustainable carrot cultivation.

The objectives of this study encompass evaluating the influence of different nitrogen levels on carrot growth parameters, assessing the impact of varied carrot varieties on yield metrics and quality attributes, investigating potential combinations between nitrogen levels and varietal diversity, and providing practical for optimizing nitrogen recommendations fertilization practices. The novelty of this research lies in its systematic exploration of the combination between nitrogen levels and varietal diversity in carrot cultivation. By conducting comprehensive experiments across varieties under varying nitrogen regimes, this study aims to offer fresh insights into optimizing yield and quality while minimizing environmental impact.

# Materials and methods

# Experimental site

The research work was conducted at Kashiadanga, Rajpara Thana, Rajshahi, Bangladesh during the period from October 2019 to March 2020.

# **Climate and Soil**

The experimental area is situated in the subtropical zone, characterized by heavy rainfall during the Kharif season and scanty rainfall in the Rabi season. The Rabi season is characterized by plenty of sunshine. The experimental area belonged to the High Ganges River Floodplain and AEZ 11. The soil was silt loam. Soil samples were randomly collected from a depth of up to 30 cm of the experimental plot, and analyses showed nitrogen at 0.075%, phosphorus at 13 ppm, exchangeable potassium at 0.20 me/100g soil, and organic carbon at 0.82%.

#### Design of the experiment

The seeds of these varieties were collected from Siddique Bazar, Dhaka. The two-factor experiment was laid out in a Randomized Complete Block Design with three replications. The experiment consisted of two factors: Factor A: Variety, V<sub>1</sub>= New Kuroda, V<sub>2</sub>= Pusa Kesar, and Factor B: Nitrogen level, with treatments T<sub>1=</sub> 0 kg N/ha; T<sub>2=</sub> 70 kg N/ha; T<sub>3=</sub> 140 kg N/ha; T<sub>4=</sub> 210 kg N/ha. The total area was divided into 24 beds with a spacing of 20 cm x 15 cm and a depth of 1.5 cm in each bed. The size of each unit plot was 1.0 m × 1.0 m.

#### Seed Soaking and Treatment

Carrot seeds were soaked in water for 12 hours and then wrapped with a piece of thin cloth before sowing. They were then spread over a polythene sheet for two hours to dry. The seeds were treated with Vitavex-200 @ 3g/100g seed.

## Seed Rate and Seed Sowing

Seeds were sown at a rate of 3 Kg/ha, as narrated by Rashid (1993), equivalent to 59 g for the experimental area. Before seed sowing, Savin 85 WP @ 2 kg/ha was applied to each plot as a precautionary measure against ants and worms infestation to the seed and seedlings.

#### Land preparation

The selected land for the experiment was first ploughed on October 20, 2019, by disc plough and was exposed to the sun for seven days prior to the next ploughing. The land was ploughed six times by a tractor to obtain a vigorous tilth. Laddering was done to break the soil clods and pieces with each ploughing. All weeds and stubbles were removed, and the land was finally prepared through the addition of basal doses of manure and fertilizers. Plots were prepared according to design and layout. Finally, soil of each plot was treated with Seven 80 WP @ 2 kg/ha to protect the young plant from the attack of mole cricket, cutworm, and ants. Irrigation channels were made around each block.

#### Collection of data

Data on various parameters such as total weight, root weight, shoot weight, total plant length, root length, shoot length, dry shoot weight/100 gm, dry root weight/100 gm, number of leaves per plant, root diameter, and yield were

recorded from five specific plants. The MSTAT-C package program was used to assemble and statistically analyze the collected data. Mean values of all treatments were calculated, and the analysis of variance for most of the characters was performed using the F variance test. The significance of the difference among the treatment means was evaluated using Duncan's Multiple Range Test (DMRT) at 1% and 5% levels of probability, following the method described by Gomez and Gomez (1984).

# Results

# **Total Plant Weight**

Nitrogen application significantly influenced the total plant weight of carrots at a 1% level of probability. The highest total plant weight (144.7 g) was observed in T<sub>3</sub> (140 kg N/ha), followed by T<sub>4</sub> (141.3 g), whereas the lowest (133.7 g) was recorded in T<sub>1</sub> (0 kg N/ha) (Table 1). Variety-wise, *Pusa Kesar* exhibited higher total biomass (145.58 g) compared to *New Kuroda* (133.48g), as shown in Fig.1A. In the combined effect, V<sub>2</sub>T<sub>3</sub> (Pusa Kesar with 140 kg N/ha) exhibited the highest total plant weight (149.0 g), while the lowest (125.3 g) was recorded in V<sub>1</sub>T<sub>1</sub> (New Kuroda with 0 kg N/ha) (Table 2).

#### **Total Root Weight**

The application of nitrogen significantly affected the total root weight at a 1% level of probability. The highest total root weight (104.2 g) was observed in  $T_3$ , whereas  $T_1$  recorded the lowest (100.0 g) (Table 1). In Fig.1A, Pusa Kesar demonstrated a heavier root system (103.25 g) than New Kuroda (101.08g). In the combined effect,  $V_2T_3$  showed the highest total root weight (104.7 g), while  $V_1T_1$  had the lowest (98.0 g) (Table 2).

# **Total Shoot Weight**

Nitrogen levels significantly influenced total shoot weight. The highest total shoot weight (40.50 g) was found in T<sub>3</sub>, whereas T<sub>1</sub> recorded the lowest (33.67 g) (Table 1). Pusa Kesar had a greater shoot weight (42.33 g) than New Kuroda 32.08g) as shown in Fig.1A. In the combined effect, the highest shoot weight (44.33 g) was observed in V<sub>2</sub>T<sub>3</sub>, while the lowest (27.33 g) was recorded in V<sub>1</sub>T<sub>1</sub> (Table 2).



**Fig. 1.** Single effect of variety on growth and yield of carrot (A). Single effect of variety on leaf number of carrot (B). Main effect of nitrogen level on leaf number of carrot ( $T_{1=} 0 \text{ kg N/ha}$ ;  $T_2=70 \text{ kg N/ha}$ ;  $T_3=140 \text{ kg N/ha}$ ;  $T_4=210 \text{ kg N/h}$ ) (C). Combination effect of nitrogen level and variety on leaf number of carrot ( $V_1$ = New Kuroda,  $V_2$ = Pusa Kesar, T1=0 kg N/ha;  $T_2=70 \text{ kg N/ha}$ ;  $T_3=140 \text{ kg N/ha}$ ;  $T_3=140 \text{ kg N/ha}$ ;  $T_4=210 \text{ kg N/ha}$ ) (D).

# **Dry Shoot Weight**

The dry shoot weight per 100 g of fresh shoot weight was not significantly affected by nitrogen application (Table 1). In the combined effect, the highest dry shoot weight (29.36 g) was observed in  $V_2T_3$ , whereas  $V_2T_1$  had the lowest (24.02 g) (Table 2) which was significantly different from the other treatments

#### **Dry Root Weight**

Nitrogen did not significantly affect the dry root weight per 100 g of fresh root weight (Table 1). New Kuroda exhibited a higher dry root weight (22.22 g) than Pusa Kesar (21.47 g) (Fig. 1). However, the combined effect was significant, with the highest dry root weight (23.54 g) observed in V<sub>1</sub>T<sub>3</sub>, and the lowest (20.00 g) in V<sub>2</sub>T<sub>4</sub> (Table 2), which was significantly different from the other treatments.

## **Root Diameter**

A significant difference at the 5% probability level was observed in root diameter among

nitrogen treatments. The highest root diameter (35.04 mm) was found in T<sub>3</sub>, whereas the lowest (31.06 mm) was in T<sub>1</sub> (Table 1). Pusa Kesar had a larger root diameter than New Kuroda (Fig.1A). In the combined effect,  $V_2T_3$  had the highest root diameter (36.04 mm), while  $V_1T_1$  had the lowest (30.39 mm) (Table 2).

## **Total Plant Length**

Total plant length was not significantly affected by nitrogen levels. The highest plant length (78.47 cm) was recorded in  $T_3$ , while the lowest (74.20 cm) was observed in  $T_1$  (Table 1). However, the combination effect was significant, with  $V_2T_3$  having the highest plant length (80.00 cm) and  $V_1T_1$  the lowest (72.00 cm) (Table 2).

#### Leaf Number

Leaf number was significantly influenced by nitrogen levels and variety. Among the varieties, Pusa Kesar recorded the highest leaf number (11.88), while New Kuroda had the lowest (10.49) (Fig. 1B). Regarding treatments, the highest leaf number (11.39) was observed in  $T_4$ , whereas  $T_1$ 

Treatments	Total plant weight (gm)	Total root weight (gm)	Total shoot weight (gm)	Dry shoot weight/100 g of fresh shoot weight (gm)	Dry root weight/100 g of fresh root weight (gm)	Diameter (mm)	Total length (cm)	Total root length (cm)	Total shoot length (cm)	Yield (t/ha)
T <sub>1</sub>	133.7d	100.0c	33.67c	24.99	21.21	31.06c	74.20	15.85	58.35	19.67d
T <sub>2</sub>	138.3c	102.5b	35.83b	27.08	22.24	32.73c	76.37	17.18	59.18	21.88c
T₃	144.7a	104.2a	40.50a	29.22	23.46	35.04a	78.47	20.07	58.40	25.52a
<b>T</b> 4	141.3b	102.5b	38.83b	27.83	20.47	34.03b	77.27	19.22	58.05	24.15b
Level of significance	*	*	*	NS	NS	*	NS	NS	NS	*

Table 1. Main effect of different levels of nitrogen on growth and yield of carrot

Means followed by the same letter(s) within a column are not significantly different at the 1% or 5% level of significance, as tested by DMRT.  $T_{1=} 0$  kg N/ha;  $T_{2=} 70$  kg N/ha;  $T_{3=} 140$  kg N/ha;  $T_{4=} 210$  kg N/ha. \*= Significant at 5% level of probability, NS= Not Significant.

had the lowest (2.66) at 90 DAS (Fig. 1C). In the combined effect,  $V_2T_3$  exhibited the highest leaf number (12.44), whereas  $V_1T_1$  recorded the lowest (10) at 90 DAS (Fig. 1D).

#### Total Root Length

Nitrogen had no significant effect on total root length. The longest root length (20.07 cm) was observed in  $T_3$ , while the shortest (15.85 cm) was recorded in  $T_1$  (Table 1). However, the combined treatment effect was significant, with

# **Carrot Yield**

Nitrogen application significantly affected carrot yield at a 5% probability level. The highest yield (25.52 t/ha) was recorded in T<sub>3</sub>, while the lowest (19.67 t/ha) was found in T<sub>1</sub> (Table 1). Pusa Kesar exhibited a higher yield potential (24.94 t/ha) than New Kuroda (20.66 t/ha). In the combined effect,  $V_2T_3$  produced the highest yield (27.03 t/ha), whereas  $V_1T_1$  had the lowest (16.50 t/ha)(Table2).

#### Discussion

The results demonstrate that both nitrogen application and varietal differences significantly influenced carrot growth and yield. The combination of nitrogen levels and variety suggests that nitrogen plays a crucial role in optimizing carrot performance, but the extent of  $V_2T_3$  exhibiting the highest root length (23.03 cm) and  $V_1T_1$  the lowest (13.10 cm) (Table 2).

# **Total Shoot Length**

Total shoot length was not significantly affected by nitrogen application. The highest shoot length (59.18 cm) was found in T<sub>2</sub>, while the lowest (58.05 cm) was recorded in T<sub>4</sub> (Table 1). On the other hand, the combined treatment effect was significant, V<sub>1</sub>T<sub>2</sub> had the highest (60.53 cm), whereas V<sub>2</sub>T<sub>4</sub> had the lowest (56.83 cm) (Table 2).

its benefits depends on genetic factors. An increase in nitrogen levels was found to positively impact plant height, which is possibly related to nitrogen functions in protein synthesis, cell division, and enlargement (Kumar et al., 2021). Conversely, the absence of nitrogen application resulted in the lowest recorded plant height of 23.33 cm, as observed by MK Ali et al. (2006).

Total plant weight increased with nitrogen application up to 140 kg N/ha, after which a decline was observed at 210 kg N/ha. Among varieties, Pusa Kesar had the highest plant weight, indicating a stronger response to nitrogen fertilization than New Kuroda. The increase in plant weight aligns with findings by Shoaib Rahman (2021) and Moniruzzaman et al. (2013), Stefanelli et al. (2010), who reported that nitrogen promotes vegetative growth and development.

Combination	Total	Total root	Total	Dry shoot	Dry root	Diameter	Total	Total	Total	Yield
of	nlant	weight (g)	shoot	weight/	weight/	(mm)	length	root	shoot	(t/ha)
treatments	weight	Weight (g)	weight	100g of	100g of	(1111)	(cm)	length	length	(0/110)
treatments	(a)		(a)	freeb	freeh		(CIII)	(om)	(om)	
	(g)		(g)	riesn	nesn			(CIII)	(CIII)	
				snoot	root					
				weight	weight					
				(gm)	(gm)					
	105.01		07.00	05.06	01.00	00.00.0	70.001	10.10	50.001	16 506
V1I1	125.3 h	98.00e	27.33g	25.96 d	21.60 c	30.39 f	/2.00 h	13.10h	58.90d	16.501
	140.0.1	100.0	40.00	04.00	00.00.1	01 70	76.40	10 (0)	57.00	00.00.1
V2I1	142.0 d	102.0 Ca	40.00C	24.02 e	20.82 a	31.72 e	76.40 e	18.600	57.80e	22.830
V T	100.0 a	100 7bod	20.675	27 12 0	22.01 h	21 72 6	74 50 ~	12.07a	60 520	10.076
<b>V</b> <sub>1</sub> <b>I</b> <sub>2</sub>	132.3 g	102.7000	29.071	27.13 C	22.810	31.72 e	74.50 g	13.97g	60.538	19.27e
V T	144.2 0	102.2 had	42.00h	27.02.0	21.60 a	22.74 od	70.00 0	20,40 a	E7 02o	24 50 0
V212	144.3 C	102.3 bcu	42.000	27.02 C	21.08 C	33.74 Cu	78.23 C	20.400	57.836	24.500
V T	140.2 0	102 7 abo	26.67d	20.09 0	22.54.0	24.04.0	76.02 d	17 100	50 02h	24.000
V1I3	140.5 e	105.7 dbC	30.07U	29.00 d	23.34 d	34.04 0	70.95 u	17.100	J9.03D	24.000
V <sub>2</sub> T <sub>2</sub>	140.0 0	10470	44.220	20.26.0	22.20 0	26.04.0	<u> 00 00 a</u>	22.020	56 07f	27.020
V 2 I 3	149.0 d	104.7 d	44.33d	29.30 d	23.30 d	30.04 a	00.00 a	23.03d	50.971	27.03d
V.T.	135 7 f	101 0 d	34.670	27.54 bc	20.03 d	33 36 d	75 57 f	16 30f	50.27c	22 00d
<b>v</b> 114	155.71	101.0 u	54.07e	27.54 bC	20.95 u	55.50 u	75.571	10.501	39.270	22.900
V <sub>2</sub> T <sub>4</sub>	1470b	104.0 ab	43.00	28 11 h	20.00 ค	34 70 h	78 97 h	22 13h	56 83f	25.40h
¥214	147.00	104.0 00	-10.00 ah	20.110	20.00 0	04.705	/0.5/ 5	22.100	00.001	20.400
			ab							
Level of	**	**	**	*	*	*	*	*	*	*
significance										
Significance										
CV%	1.13	2.01	5.09	3.19	2.47	1.88	0.80	1.88	0.68	2.69
										,

Table 2. Combined effect of variety and nitrogen level on growth and yield of carrot

Means followed by the same letter(s) within a column are not significantly different at the 1% or 5% level of significance, as tested by DMRT. Whereas; V<sub>1</sub>= New Kuroda, V<sub>2</sub>= Pusa Kesar, T<sub>1=</sub> 0 kg N/ha; T<sub>2=</sub> 70 kg N/ha; T<sub>3=</sub> 140 kg N/ha; T<sub>4=</sub> 210 kg N/ha, CV= Coefficient of Variation. \*\*= Significant at 1% level of probability, \*= Significant at 5% level of probability.

Root length and total plant length exhibited a positive response to nitrogen, with the longest root recorded at 140 kg N/ha. The combination effect showed that Pusa Kesar at 140 kg N/ha had the longest root, emphasizing the role of genetic variation in root elongation (Choudhary et However, excessive al.. 2023). nitrogen application did not lead to a further increase in root length, indicating that beyond a certain threshold, nitrogen may promote vegetative growth at the expense of root expansion-a trend also noted by Gastal et al. (2015). Shoot length did not show significant variation with nitrogen levels, though the longest shoot was recorded at 70 kg N/ha. Among the variety-nitrogen combinations, New Kuroda at 70 kg N/ha exhibited the longest shoot. These findings align with Patil and Gill (1981), who reported that nitrogen application increases shoot height but may lead to resource diversion at excessive levels. Skrbic (1987) noticed that nitrogen fertilization had a significant effect on the dynamics of the increase in the number of leaves in carrots. Total shoot weight increased significantly with nitrogen application, peaking at 140 kg N/ha, followed by a decline at 210 kg N/ha. The combination effect confirmed that Pusa Kesar at 140 kg N/ha produced the highest shoot weight, suggesting that this variety efficiently utilizes nitrogen for vegetative growth.

Similarly, total root weight followed a comparable trend, with 140 kg N/ha having the highest value. In the combined effect, Pusa Kesar at 140 kg N/ha had the highest root weight. This response can be attributed to increased photosynthesis and carbohydrate accumulation in roots, as suggested by Dawuda et al. (2019). Dry shoot weight and dry root weight were not significantly affected by nitrogen alone. However, in the variety-nitrogen combination, the highest

dry shoot weight was observed in Pusa Kesar at 140 kg N/ha, whereas New Kuroda at 140 kg N/ha had the highest dry root weight. These results suggest that Pusa Kesar is more efficient in biomass accumulation, while New Kuroda allocates more resources to root dry matter, a trend also reported by Sarker (1989) and Kushwah et al. (2019).

Root diameter significantly increased with nitrogen application, reaching its maximum at 140 kg N/ha. Among the variety-nitrogen combinations, Pusa Kesar at 140 kg N/ha exhibited the highest root diameter. These results are in agreement with previous studies by Shoma et al. (2014) and Nikmatullah et al. (2021), which reported that nitrogen enhances root expansion but may lead to excessive foliage at higher rates.

Carrot yield was significantly influenced by nitrogen, with the highest yield recorded at 140 kg N/ha. The combination effect showed that Pusa Kesar at 140 kg N/ha had the highest yield, confirming that this variety performed better under optimal nitrogen application. The yield decline at 210 kg N/ha suggests that excessive nitrogen may lead to nutrient imbalances, reducing the efficiency of carbohydrate partitioning toward root development (Ladumor et al., 2020). These findings align with previous research by Sarker (1989), Sagiv et al. (1994), and Abdel Razik and El-Haris (1997), emphasizing that both nitrogen optimization and variety selection are key factors for achieving high carrot productivity.

# Conclusion

This study shows that nitrogen levels and carrot variety play a big role in growth and yield. Applying up to 140 kg N/ha helped plants grow

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bigger, with more root and shoot weight, but adding more (210 kg N/ha) led to lower results. Among the two varieties, Pusa Kesar performed better than New Kuroda, especially in total plant weight, root diameter, and yield. The best results (27.03 t/ha) were seen in Pusa Kesar with 140 kg N/ha, showing that nitrogen needs to be managed based on the variety. Too much nitrogen led to imbalanced growth, reducing the benefits. These findings confirm that the right nitrogen amount helps plants grow well by supporting root development and carbohydrate storage. To improve carrot production, farmers should carefully manage nitrogen use and choose the right variety. Future research should explore other farming techniques to further boost carrot yield while keeping nitrogen use efficient and sustainable.

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# **Conflicts of Interest**

The authors declare no conflicts of interests.

# Authors' contribution

Conceptualization, MKA; Methodology, MKA, and FM; Investigation, FM; Data curation, AM, MAM, IN, MJA, and MKF; Formal analysis, AM, IN, MJA, and MMH; Visualization, FM; Writingoriginal draft preparation, FM; Writing-review and editing, all authors; Supervision, MKA. All authors have read and agreed to the published version of the manuscript.

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