

INFLUENCE OF DIETARY SUPPLEMENTED GLUTAMATE ON GROWTH PERFORMANCE, HAEMATOLOGICAL AND SERUM INDICES OF BROILER CHICKENS IN HOT TROPICAL ENVIRONMENT

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 Supporting Information



ABSTRACT: This study investigated the impact of glutamate on growth, haematological and serum indices of broiler chickens in hot tropical environments (> 30°C). One-hundred-and-sixty-day old Arbor acre male broiler chicks were randomly allotted into four dietary treatment groups of four replicates having 10 chicks per replicate in a completely randomized design. Dietary treatments groups contain different inclusion levels of glutamate (basal diet + 0%, 0.3%, 0.6% and 0.9% glutamate (G), in an open sided pen). The experiment lasted for seven weeks during which data were collected on growth, haematological and serum indices. Result showed birds fed diet containing 0.9% G at the starter phase had ($P < 0.05$) increased feed intake compared to other treatment groups. Inclusion of 0.6% G at the finisher phase ($P < 0.05$) improved final weight and weight gain in birds compared to 0.3% G group. Best feed conversion ratio ($P < 0.05$) was obtained from birds fed diet containing 0.6% G compared to birds on 0.3% and 0.9% inclusion. Inclusion of 0.6% G at the starter phase ($P < 0.05$) improved total protein. Increased mean corpuscular haemoglobin ($P < 0.05$) was obtained from birds on 0.6% G compared to lower mean values obtained from birds on 0.3% G. The study concluded that inclusion of glutamate in the diet of broiler chickens at 0.6% improved final weight, weight gain and feed conversion ratio without causing any adverse effect on blood indices of broiler chickens as glutamate increased serum protein and mean corpuscular haemoglobin of broiler chickens.

Keywords: Blood profile, Broiler chickens, Essential nutrients, Glutamic acid, Growth response.

INTRODUCTION

Feed additives used in poultry industry that provide essential nutrients, increase palatability of the feed, improve growth performance and ensure efficient feed utilization. Broiler chickens with high genetic potentials need to maintain good immune functions in order to achieve their genetic potentials. The use of suitable and quality feed additives is important in the poultry industry in order to enhance bird genetic potentials, maintain immune functions, mitigate effect of stress and ensure balance in gut environment (Adedokun and Olojede 2019).

With increasing industry standards and consumer awareness as well as demand for healthy food products of animal origin, there is an increased awareness for alternatives than the conventional feed additives used in animal feed products (Pandey et al., 2019). Higher feed efficiency and profitability are essential in broiler production. Heat-stressed broilers have higher glutamate requirement, hence supplementing with glutamate or its decarboxylated derivatives (γ -GABA) can enhance the intestinal morphology and survival rate in heat-stress conditions (Porto et al., 2015; Zhong et al., 2020). This has necessitated the search for additional feed additives that can enhance growth for livestock, reduce impact of different stressors and protect cells against oxidative damage in tropical environment. Monosodium glutamate, according to (Young and Ajami, 2000), is a feed additive that can improve growth rate and enhance immune system in broiler chickens. Glutamate functions as a constituent of proteins, a substrate in the synthesis of amino acids and a precursor to several nonessential amino acids which helps the metabolism. Monosodium glutamate is the sodium salt of glutamic acid and the main component of many proteins (Tawfik and AlBadr, 2012).

Inclusion of glutamate in the feed as feed additive was reported to improve performance by improving the quality of small intestine for better nutrient utilization and enhancing immune system function (Maslami et al., 2019). Glutamate in temperate environment has proven to mitigate heat stress and enhance oxidative defenses by protecting cells from oxidative damage which in turn enhance growth performance (Olubodun et al., 2015). Broiler production in tropics has been characterized by different stressors which restrict growth, result in economic losses and comprise immune status of the birds.

RESEARCH ARTICLE
 PII: S222877012500042-15
 Received: July 28, 2025
 Revised: November 27, 2025
 Accepted: November 28, 2025

Therefore, there is need to investigate the use of glutamate in tropical environment where different stressors prevent broilers from achieving their genetic potentials. Thus, this study aims to evaluate the influence of glutamate on growth performance, haematological and serum indices of broiler chickens in tropical environment.

MATERIALS AND METHODS

The experiment was carried out at the poultry unit of the teaching and research Farms, Federal University of Agriculture, Abeokuta, Ogun State. It lies on Latitude 7 10' N and Longitude 3 2'E, it is 76 metres above sea level and located in the tropical rainforest vegetation zone with an average temperature of 34.7C and relative humidity of 82% (Google-Earth, 2023).

Source of test Ingredients

Glutamate was sourced from a market in Abeokuta metropolis Ogun State Nigeria. The brand name of the Glutamate was VEDAN. The monosodium glutamate was kept in an air-tight container before usage.

Experimental animals and management

A total number of 160 day-old Arbor acre broiler chicks were purchased from a reputable hatchery in Ibadan, Oyo State. Prior to the arrival of the chicks, the pens were cleaned and disinfected thoroughly with a disinfectant. The chicks were assigned to four dietary treatment groups of four replicates each having ten chicks per replicate in a completely randomized design. Dietary inclusion of the test ingredient (glutamate) commenced at the beginning of the experiment. Routine vaccination and medication were administered to the birds according to routine procedures while, feed and water were provided *ad libitum*

Experimental diet and design

Birds were fed with Commercial starter mash between day one to 28 days. The starter diet contains (2900 kcal/kg Metabolizable Energy, 23% crude protein while, the finisher diet was given from 29 days to the end of the experiment (2600 kcal/kg Metabolizable Energy, 20% crude protein. Both diets contain different inclusion levels of monosodium glutamate at (0%, 0.3%, 0.6% and 0.9%). The treatment groups were arranged thus, Treatment 1: Control diet without glutamate, treatment 2: Control diet + 0.3% Glutamate, treatment 3: Control diet + 0.6% Glutamate, treatment 4: Control diet + 0.9% Glutamate. The experiment lasted for a period of seven weeks.

Feed Intake

Feed intake was recorded weekly, the leftover of the feed was subtracted from total feed given to the birds and expressed as Feed intake (kg) = Total feed given (kg) – Feed leftover (kg)

$$\text{Average feed intake} = \frac{\text{Total feed intake}}{\text{Number of birds in the replicate}}$$

Weight gain

Birds were weighed on weekly basis throughout the experimental period. The body weight gain was calculated by subtracting initial body weight from the final body weight. Weight gain = Final body weight – Initial body weight.

Feed conversion ratio

This was calculated by the feed intake divided by weight gain.

$$\text{FCR} = \frac{\text{Feed intake} \left(\frac{\text{g}}{\text{day}} \right)}{\text{Weight gain} \left(\frac{\text{g}}{\text{day}} \right)}$$

Water intake

Water intake was recorded daily as Water intake = Total volume of water given (ml) – Volume of water left (ml).

Mortality

Mortality was recorded as number of dead birds divided by the number of birds stocked and expressed in percentage.

$$\text{Mortality (\%)} = \frac{\text{Total number of dead birds}}{\text{Total number of birds stocked}} \times 100$$

Blood collection

Blood samples were collected randomly from two birds across treatments groups making a total of 32 samples. Blood samples were collected on the 28th and 49th day of the experiment for haematological and serum biochemical

indices. Blood was collected through the brachial vein with a disposable syringe. 5 ml of blood was collected from each bird, 3ml was dispensed into well labeled sterile anticoagulant bottles for hematological indices while, 2ml of the blood was dispensed into well labeled plain bottle for serum biochemical indices.

Determination of haematological indices

The Packed cell Volume and Haemoglobin were determined using microhematocrit and cyanmethemoglobin methods respectively as outlined by Jain (1986). Red blood cells count was determined using haemocytometry method as outlined by Jain (1986). White blood cell was determined using Neubauer counting chamber according to Jain (1986). The Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and the mean corpuscular haemoglobin concentration were calculated using standard formulae (Samour 2015).

Statistical analysis

All data collected were subjected to one-way Analysis of Variance (ANOVA) in a Completely Randomized Design (CRD) using (SAS, 2012). Significant ($P < 0.05$) different means were separated using Duncan multiple range test of the same statistical package. The statistical model was: $Y_{ij} = \mu + T_i + \sum ij$

Where; Y_{ij} = Observed mean value; μ = Population mean; T_i = Effect of glutamate levels; $\sum ij$ = Random residual error.

RESULTS

Performance of broiler chickens fed diet containing glutamate at starter phase is shown in table 1. Inclusion of glutamate in the diet of broiler birds had no significant ($P > 0.05$) effect on all performance parameters measured except total feed intake and daily feed intake that was significantly ($P < 0.05$) influenced by the inclusion of glutamate in the diet. Significantly ($P < 0.05$) higher feed intake and daily feed intake was observed in birds fed diet containing 0.9% glutamate compared to lowest mean value recorded for the control groups. Numerically higher final weight and weight gain were observed in birds fed diet containing 0.3% glutamate than mean values obtained from birds across the dietary treatment groups. Performance of broiler chickens fed diet containing monosodium glutamate at finisher phase is shown in table 2. Inclusion of glutamate in diet of broiler chickens significantly ($P < 0.05$) influenced final weight, weight gain and feed conversion ratio. While, all other parameters measured are not significantly ($P > 0.05$) influenced. Highest final weight and weight gain was observed from groups 0.6% glutamate compared to lowest final weight and weight gain observed from groups fed 0.3% glutamate. Numerically higher feed intake was observed in groups fed 0.90% glutamate than lowest feed intake obtained in groups fed 0.3% glutamate. Best feed conversion ratio was observed in birds fed diet containing 0.6% glutamate than poorer and higher feed conversion obtained from birds fed diet containing 0.3% and 0.9%. The effect of glutamate on the hematological parameters of broiler chickens at the starter phase is shown in table 3. There were no significant ($P > 0.05$) differences obtained on all the parameters measured except eosinophil and lymphocyte. Significantly higher ($P < 0.05$) eosinophils and lymphocyte count was observed from groups fed diet containing 0.6% monosodium glutamate compared to significantly ($P < 0.05$) lowered eosinophils and lymphocytes count observed for the control groups. The effect of glutamate on serum biochemistry indices of broiler chickens at the starter phase is shown in table 4. There were no significant ($P > 0.05$) differences obtained on all the parameters measured except for total protein. Significantly ($P < 0.05$) higher total protein was observed in groups fed 0.6% monosodium glutamate than lowered mean values observed in groups fed 0.3% and 0.9% monosodium glutamate. The effect of glutamate on haematological parameters of broiler chickens at finisher phase is shown in Table 5. Significant differences ($P > 0.05$) were observed in all the parameters measured except for mean corpuscular haemoglobin that was significantly influenced by the inclusion of glutamate. Significantly ($P < 0.05$) higher mean corpuscular haemoglobin was observed in groups fed 0.6% compared to lowered mean values obtained from birds fed 0.3% glutamate. The effect of glutamate on serum biochemistry indices of broiler chickens at finisher phase is shown in table 6. There were no significant differences ($P > 0.05$) observed in all parameters measured except for Aspartate aminotransferase. Significantly ($P < 0.05$) higher aspartate aminotransferase was observed in the control groups compared to significantly lowered mean values observed from groups fed 0.3% and 0.6%, respectively.

Table 1 - Effect of glutamate on performance of broiler chickens at starter phase.

Parameters	Inclusion level	0%	0.3%	0.6%	0.9%	SEM	P-value
Initial weight (g)		45.55	44.45	45.60	48.27	0.79	0.40
Final weight (g)		1096.11	1172.50	1145.56	1155.83	20.63	0.64
Weight gain (g)		1050.56	1128.05	1099.56	1107.56	20.22	0.62
Total feed intake/bird (g)		1945.28 ^b	2025.50 ^{ab}	2078.63 ^{ab}	2195.63 ^a	36.54	0.04
Daily feed intake/bird/day (g)		69.47 ^b	73.30 ^{ab}	74.24 ^{ab}	78.42 ^a	1.31	0.01
Feed conversion ratio		1.87	1.82	1.89	1.98	0.04	0.21
Total water intake (l)		42.71	43.54	4.61	42.53	42.82	0.08
Water intake/bird (l)		4.38	4.35	4.61	4.63	63.96	0.16
Mortality rate		0.25	0	0.25	0.75	0.15	0.38

^{ab} Means values in the same row with different superscript differ significantly ($P < 0.05$); SEM: Standard Error of Mean

Table 2 - Effect of glutamate on performance of broiler chickens at finisher phase.

Parameters	Inclusion level	0%	0.3%	0.6%	0.9%	SEM	P-value
Initial weight (g)		1096.11	1172.50	1145.56	1155.83	20.63	0.64
Final weight (g)		2126.94 ^{ab}	2105.00 ^b	2286.11 ^a	2162.78 ^{ab}	41.46	0.04
Weight gain (g)		1030.84 ^{ab}	932.5 ^b	1140.55 ^a	1006.95 ^{ab}	30.44	0.01
Total feed intake/bird (g)		2995.50	2862.50	2998.33	3085.31	44.57	0.39
Daily feed intake/bird/day (g)		149.77	143.12	149.92	154.27	2.23	0.02
Feed conversion ratio		2.91 ^{ab}	3.06 ^a	2.65 ^b	3.06 ^a	0.07	0.02
Total water intake (l)		60.18	59.72	60.96	60.96	444.81	0.27
Water intake/bird (l)		6.18	5.97	6.26	6.36	83.47	0.44
Mortality rate		0	0	0	0	0	0

^{ab} Means values in the same row with different superscript differ significantly (P<0.05); SEM: Standard Error of Mean**Table 3 - Effect of Glutamate on the hematological parameters of broiler chickens at starter phase.**

Parameters	Treatment	Control	0.3% G	0.6% G	0.9% G	SEM	P-value
Packed cell volume (%)		27.50	27.00	27.50	23.00	1.01	0.23
Haemoglobin (g/dl)		9.20	9.00	9.20	7.65	0.37	0.21
Red blood cells (x10 ⁹ /L)		3.41	4.28	4.11	2.42	0.36	0.11
White blood cells (x10 ⁹ /L)		14.76	24.00	16.25	0.02	54.98	0.22
Lymphocytes (%)		84.50	65.00	82.50	100.00	6.45	0.11
Neutrophil		29.50	30.50	30.50	32.00	0.46	0.11
Monocytes (%)		1.00	2.50	1.50	1.50	0.26	0.08
Eosinophil (%)		1.00 ^b	2.00 ^{ab}	2.50 ^a	2.00 ^{ab}	0.23	0.04
Lymphocytes (%)		2.83 ^b	3.56 ^{ab}	4.07 ^a	3.80 ^{ab}	0.20	0.03
Mean corpuscular volume (%)		94.10	93.90	92.90	92.15	0.44	0.19
Mean corpuscular hemoglobin (g/l)		29.07	21.01	27.74	32.95	0.24	0.14

^{abc}: Means values in the same row with different superscript differ significantly different at P<0.05.**Table 4 - Effect of glutamate on the serum biochemistry of broiler chickens at starter phase.**

Parameters	Treatment	Control	0.3% G	0.6% G	0.9% G	SEM	P-value
Albumin (g/dl)		1.51	1.36	1.64	1.33	0.31	0.45
Cholesterol (mg/dl)		60.30	76.65	57.16	35.01	10.79	0.24
AST (U/L)		32.52	29.95	30.57	30.24	3.75	0.08
ALT (U/L)		7.54	11.07	12.85	12.05	0.94	0.07
Total protein (g/dl)		2.89 ^{ab}	2.41 ^b	3.88 ^a	2.73 ^b	0.23	0.04
Globulin (g/dl)		1.39	1.06	1.09	1.40	0.13	0.49
Glucose (mg/dl)		58.57	85.57	86.48	88.13	7.59	0.06
Creatinine		0.29	0.50	0.40	0.66	0.87	0.24

^{abc}: Means with different superscript are significantly different at P<0.05. AST:Aspartate aminotransferase; ALT: Alanine aminotransferase**Table 5 - Effect of Glutamate on the hematological parameters of broiler birds at finisher phase.**

Parameters	Treatment	Control	0.3% G	0.6% G	0.9% G	SEM	P-value
Packed cell volume (%)		31.50	31.00	30.00	31.00	0.72	0.59
Haemoglobin (g/dl)		10.30	10.00	10.10	10.10	0.20	0.71
Red blood cells (x10 ⁹ /L)		3.13	3.11	3.02	3.12	0.81	0.73
White blood cells (x10 ⁹ /L)		15.77	15.50	15.50	14.02	0.85	0.29
Lymphocytes		65.50	68.00	69.00	66.00	0.74	0.15
Neutrophil		27.00	25.00	25.00	28.00	0.75	0.24
Monocytes (%)		3.50	3.00	1.50	3.00	0.13	0.09
Eosinophil (%)		2.50	3.50	4.00	3.00	0.56	0.48
Basophil		0.50	0	0	0	0.13	0.24
Mean corpuscular haemoglobin (g/l)		33.15 ^{ab}	30.30 ^b	33.78 ^a	32.41 ^{ab}	0.58	0.04

^{abc}: Means with different superscript are significantly different at P<0.05.

Table 6 - Effect of glutamate on the serum biochemistry of broiler chickens at finisher phase

Parameters	Control	0.3% G	0.6% G	0.9% G	SEM	P-value
Albumin (g/dl)	1.42	1.48	1.44	1.38	0.08	0.74
Cholesterol (mg/dl)	38.03	34.42	38.11	30.32	3.69	0.58
AST(U/L)	50.75 ^a	32.10 ^b	33.03 ^b	43.56 ^{ab}	3.36	0.03
ALT (U/L)	2.08	2.24	3.92	7.04	0.96	0.10
Total Protein(g/dl)	2.71	2.54	2.60	2.45	0.06	0.22
Globulin (g/dl)	1.18	1.01	1.17	1.07	0.09	0.65
Glucose (mg/dl)	152.38	132.61	134.44	193.48	0.2	0.42
Creatinine	0.75	0.90	0.71	0.85	0.52	0.31

^{abc}: Means with different superscript are significantly different at $p < 0.05$. AST:Aspartate aminotransferase; ALT: Alanine aminotransferase

DISCUSSION

Higher feed intake observed at the starter phase in groups fed 0.9% glutamate could be attributed to flavor enhancing attributes of glutamate as it has been reported that glutamate have flavoring enhancing attributes which influences feed intake, this result affirms the attributes of glutamate as a food enhancer. This result agrees with the findings of [Khadiga et al. \(2009\)](#) that reported increased feed intake in chicks fed 1% monosodium glutamate. This result is also in tandem with the findings of [Maslami et al. \(2019\)](#) who reported significant difference in feed intake of broiler chickens fed dietary monosodium glutamate inclusion. Research findings by [Gbore et al. \(2016\)](#) also aligns with this studies that increased feed intake in response to an increased level of monosodium glutamate among female rabbits showed that monosodium glutamate is capable of enhancing feed palatability which in turn influences appetite positively and induces weight gain, which has been linked with stimulation of the sensory receptors ([Moore, 2003](#)).

Highest final weight observed at the finisher phase in groups fed 0.6% glutamate might be attributed to better nutrient absorption and utilization at this inclusion level of the additive as a result of improved feed intake, since glutamate has capability of improving intestinal conditions by increasing the villi length and increasing the intestinal integrity ([Reeds et al., 2000](#); [Newsholme et al., 2003](#)). This result corroborates the findings of [Olarotimi and Adu \(2022\)](#) that reported heavier final weight in broiler chickens fed diet containing 0.5% monosodium glutamate. Highest weight gain observed in groups fed 0.6% glutamate might be attributed to better nutrient absorption and improved digestion that improved weight gain as a result of glutamate inclusion as glutamate is a precursor to other non- essential amino acids such as arginine, glutamine and proline ([Blachier et al., 2009](#)). This result corroborates the findings of [Maslami et al. \(2019\)](#) that reported significant difference in the body weight of broiler chickens supplemented monosodium glutamate at 0.6 - 0.8% inclusion. Best feed conversion ratio was observed in groups fed 0.6% glutamate, this might be attributed to better nutrient absorption and utilization as a result of glutamate inclusion because as body weight increases as a result of increased feed nutrient absorption improved feed conversion ratio is achieved. This result agrees with the findings of ([Maslami et al., 2019](#); [Olarotimi and Adu 2022](#)) that reported significant and best feed conversion in broiler chickens fed dietary monosodium glutamate at 0.5 g/kg and between 0.4-0.8% inclusions respectively.

Result observed on packed cell volume, hemoglobin, red blood cell, Neutrophils and mean corpuscular hemoglobin at the starter phase corroborates the findings of [Olarotimi and Adu \(2022\)](#) who reported no significant difference when monosodium glutamate was fed to broiler chickens. Higher eosinophils count at starter phase was observed in group fed diet 0.6% glutamate. This result might be attributed to ability of glutamate to stimulate body eosinophils to protect body cells from parasites, allergies and foreign bacteria within and outside the birds thereby acting as immune booster. This result is contrary to the findings of [Olarotimi and Adu \(2022\)](#) who reported no significant difference in eosinophils of broiler chickens fed monosodium glutamate. Higher lymphocytes count was observed in groups fed 0.6 % glutamate; this result could be attributed to ability of glutamate to stimulate the immune system to prevent against foreign bacteria and organism within the system of the birds. This result is contrary to the findings of [Zanfirescu et al. \(2019\)](#) who reported monosodium glutamate to decrease the level of lymphocytes in the blood without interfering with the basal phagocytes of Neutrophils. Similarly, this result also disagree with the findings of [Olarotimi and Adu \(2022\)](#) that reported no significant difference in the lymphocytes of broiler chickens fed monosodium glutamate. Result observed on serum indices at the starter phase showed significantly higher total protein was observed in groups fed 0.6% glutamate. This might be attributed to inclusion of glutamate acting as precursor for other amino acids and increasing the serum protein content. Similarly, glutamate has been reported to act as a transcriptional promoter and enhancer used for control of gene expression that promoted the ability of ribonucleic acid polymerase to identify the nucleotide at the initiation stage thereby improving protein synthesis. This result is in agreement with the findings of [Azine et al. \(2018\)](#) and [Obochi et al.](#)

(2009) that reported significant difference and increase in the serum protein of broilers and rats respectively when monosodium glutamate was fed to the animals but contrary to the findings of Okediran et al. (2014) that reported decrease in the serum protein of rats supplemented with diet containing monosodium glutamate.

Mean corpuscular hemoglobin was significantly influenced by dietary inclusion of glutamate. Groups fed 0.6% glutamate had higher 33.78g/l corpuscular hemoglobin than groups on 0.3% glutamate. The significant effect observed on mean corpuscular haemoglobin signified enough presence of oxygenated blood within the system of the birds thereby preventing anaemic condition. This result agrees with the findings of Olorotimi and Adu (2022) that reported significant differences in the mean corpuscular volume of broiler chickens fed dietary monosodium glutamate.

Result observed on serum indices at the finisher phase showed AST was significantly influenced by dietary inclusion of glutamate. Higher AST was observed in the control group compared to lower mean values observed from the treatment groups. The significantly lower AST observed from the treatment groups showed that glutamate does not have any negative impact on the liver cells of the birds as the values obtained were within the range for healthy chicken. This result is contrary to the findings of Azine et al. (2018) and Prabakar et al. (2023) that reported no significant difference in the AST of broiler chickens fed diets containing jumbo cube as source of monosodium glutamate and L-glutamate in the diets of broilers chickens respectively.

CONCLUSION

In conclusion, dietary inclusion of glutamate improved growth performance of broiler chickens at the finisher phase with no detrimental effect on haematological, liver and kidney functions in hot tropical environment. Also, glutamate improved total protein, while maintaining higher mean corpuscular haemoglobin in broiler chickens. As a practical suggestion, glutamate could be added up to 0.9% in the diet of broiler chickens.

DECLARATIONS

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Ethical approval

The authors complied with the ARRIVE guidelines and Interdisciplinary Principles and Guidelines for the Use of Animals in Research, Testing, and Education by the New York Academy of Sciences, Ad Hoc Animal Research Committee. Guidelines of the Animal Ethical Ethics committee (FUNAAB, 2020) of the Federal University of Agriculture, Abeokuta, was also strictly adhered to ensure the welfare of the birds was not compromised.

Authors' contribution

A.A ADEYEMO: Conception, design and final approval, U. O ABDULAH: Drafting, Field Study and Monitoring, A.J ADEYEMO: Design and Proof reading, A.I IYANDA: Coordination and revising, T.O ADEKOLA: Acquisition of data, E.O ADEKUNLE: statistical analysis. All authors read and approved the final manuscript

Availability of data and materials

The data sheet generated during and /or analysed during this study are not publicly available but are available from the corresponding author based on reasonable request.

Funding

The authors declare that no funds, grants, or other support were received during the project and preparation or publication of this manuscript.

Competing interests

The authors declared they have no competing interest with respect to research, authorship and publication of this article.

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