doi.org/10.36721/PJPS.2026.39.3.REG.13738.1

# Monosodium L-glutamate affects orthodontic outcomes by inhibiting hypothalamic-pituitary-gonadal axis function

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**Abstract**: **Background**: Orthodontic treatment involves complex bone remodeling, with the Osteoprotegerin (OPG)/Receptor Activator of nuclear factor Kappa-B Ligand (RANKL)/Receptor activator of nuclear factor Kappa-B (RANK) (OPG/RANKL/RANK) signaling pathway regulating osteoclast activity. Monosodium L-glutamate (MSG) disrupts the hypothalamic-pituitary-gonadal (HPG) axis and may influence bone metabolism. **Objectives**: This study examined the effects of MSG-induced HPG axis disruption on orthodontic outcomes in neonatal rats. **Methods**: Rats received either intraperitoneal MSG injections or saline (sham). After 56 days, hormone levels were measured. On the 57$^{th}$ day a rat model for orthodontic tooth movement was established. On the 56$^{th}$ and 84$^{th}$ days, hormone levels and inflammatory cytokine expression, including interleukin-6, interleukin-1beta and tumor necrosis factor alpha, was assessed. The OPG/RANKL ratio, osteoclast counts and bone structure were also measured. **Results**: MSG significantly reduced sex hormone levels (GnRH, FSH, LH, estradiol and testosterone), increased osteoclast numbers, elevated cytokine expression decreased the OPG/RANKL ratio, indicating enhanced bone resorption. Micro-CT revealed increased periodontal space, reduced bone volume fraction, greater inter-molar distance in MSG-treated rats, reflecting accelerated orthodontic tooth movement. **Conclusion**: These results suggest that MSG-induced disruption of the HPG axis alters bone metabolism and orthodontic tooth movement, indicating that hormonal imbalance plays an important role in bone metabolism and tooth movement during orthodontic treatment.

**Keywords**: Monosodium L-glutamate; NF-kappaB ligand; Orthodontic tooth movement; Osteoprotegerin; Sex hormone

*Submitted on 19-11-2024 – Revised on 27-01-2025 – Accepted on 29-01-2025*

## INTRODUCTION

Orthodontic treatment relies on mechanical forces to stimulate tooth movement, initially altering the alveolar bone and periodontal ligament and subsequently remodeling the alveolar bone structure (Seddiqi, *et al*., 2023, Montiel, *et al*., 2024). Compressive forces cause tissue shrinkage, vascular constriction, reduced blood flow and collagen fiber degradation, thereby promoting osteoclast-mediated local bone resorption, a process primarily induced by orthodontic forces. In contrast, tensile forces elongate periodontal ligament fibers, expand the periodontal space and stimulate fibroblast and collagen fiber proliferation, thereby triggering osteoblast differentiation and new bone formation. Through continuous bone remodeling, orthodontic forces maintain the dynamic balance of the alveolar bone (Jiao, *et al*., 2023, Liu, *et al*., 2022).

Studies have shown that monosodium glutamate (MSG) interferes with bone metabolism by disrupting the hypothalamic-pituitary-gonadal (HPG) axis. Neonatal exposure to MSG damages gonadotropin-releasing hormone (GnRH) neurons in the hypothalamic arcuate nucleus, leading to reduced GnRH secretion, which subsequently inhibits the release of luteinizing hormone (LH) and follicle-stimulating hormone (FSH), resulting in decreased levels of estrogen (E2) and testosterone (T) (Nemeroff, *et al*., 1981). This dysfunction is characterized by reduced GnRH secretion, subsequently inhibiting the release of luteinizing hormone (LH) and FSH, ultimately resulting in a marked decline in sex hormone levels, including E2 and T. Sex hormones play a central role in regulating bone metabolism. Estrogen reduces bone resorption, particularly in response to systemic hormonal changes, by inhibiting osteoclast differentiation and activity, downregulating Receptor Activator of Nuclear Factor Kappa-B Ligand (RANKL) expression and upregulating Osteoprotegerin (OPG) expression. Additionally, it promotes bone formation by activating osteoblast-related signaling pathways, such as Wnt/β-catenin and TGF-β/Smad pathways (Deng and Guo, 2020). On the other hand, testosterone reduces bone resorption by

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directly suppressing osteoclast activity, an effect mediated systemically and enhances bone formation by stimulating osteoblast differentiation and function (Deng and Guo, 2020). Consequently, the decline in sex hormone levels induced by MSG disrupts the balance between bone formation and resorption, resulting in increased osteoclast activity and reduced osteoblast function, ultimately accelerating bone loss.

MSG further exacerbates its impact on bone metabolism by inducing oxidative stress and inflammatory responses. The metabolism of MSG can lead to excessive production of reactive oxygen species (ROS) (Oluwole, *et al*., 2024), inhibiting osteoblast differentiation and enhancing osteoclast activity. Additionally, MSG activates the nuclear factor kappa-B (NF-κB) signaling pathway (Oluwole, *et al*., 2024), increasing the release of pro-inflammatory cytokines such as TNF-α, IL-6 and IL-1β. These cytokines directly stimulate osteoclastogenesis and accelerate bone resorption, primarily due to systemic hormonal disruption (Udagawa, *et al*., 2021).

Numerous findings have reported the OPG / RANKL / Receptor Activator of Nuclear Factor Kappa-B (RANK) (OPG/RANKL/RANK) signaling pathway in orthodontic root resorption. OPG, a paracrine regulator, plays a potential role in osteoclast generation and bone metabolism (Udagawa, *et al*., 2021). It also exerts inhibitory effects on osteoclast differentiation, absorption and apoptosis. Furthermore, RANKL promotes the formation, fusion, differentiation, activation and survival of osteoclasts, thereby enhancing bone remodeling (Marahleh, *et al*., 2021, Yoshimatsu, *et al*., 2022).

Monosodium L-glutamate (MSG), a neurotoxin, disrupts the hypothalamic-pituitary-gonadal (HPG) axis when administered neonatally, leading to neuroendocrine dysfunction by damaging the hypothalamic arcuate nucleus (Haddad, *et al*., 2021). This damage reduces the HPG axis activity, which may impair bone metabolism and remodeling through inhibition of luteinizing hormone-releasing hormone (LHRH) neurons.

The study hypothesizes that MSG-induced HPG axis inhibition interferes with orthodontic treatment outcomes by altering bone metabolism. The findings of this study provide a theoretical basis for understanding the potential impact of MSG on bone metabolism and orthodontic outcomes and offer valuable insights for developing new strategies to mitigate adverse effects during orthodontic treatment.

## MATERIALS AND METHODS

### *Experimental animals*

Neonatal female Sprague Dawley rats (n = 10, 6.97 ± 2 g, within 24 h of birth, obtained from Bikai Experimental Animals Co., Ltd.) were housed under standard conditions at the Animal Center of Zhongshan Hospital, Fudan University, Shanghai, China. Newborn rats were initially nurtured by their mothers for 28 days before being weaned and housed individually in separate cages. The rats were housed in animal facilities under standard conditions, with a 12-hour light/dark cycle, temperature maintained at 22 ± 1 ℃ and humidity controlled at 50 ± 10%. Water and food were provided ad libitum.

### *Hypothalamic-pituitary-gonadal axis dysfunction model*

The rats were randomly divided into two groups: the model and sham groups, each consisting of 5 rats. Rats in the model group received intraperitoneal injections of 10% MSG (4 mg/g) at specific time points (2, 4, 6, 8 and 10 days after birth) (Nemeroff, *et al*., 1981), while the sham group received intraperitoneal injections of normal saline. Following a 28 day period during which the newborn rats were nurtured by their mothers, they were weaned and housed individually in separate cages. On the 56th day, blood samples were collected via the abdominal aorta to measure sex hormone levels, including E2, LH, FSH, GnRH and T.

### *Experimental orthodontic tooth movement model*

On the 57$^{th}$ day, a rat model for orthodontic tooth movement was established by fitting an orthodontic device (Unitek, Ormco, 3M, USA). Following anesthesia with pentobarbital sodium (2%, 50 mg/kg), the rats' limbs were restrained and their tongues extended to prevent airway obstruction. A nitinol tension spring was placed between the first maxillary molars and incisors to create a gap, thereby inducing medial movement of the first molar. To secure the device, a shallow groove was created on the maxillary incisor using a dental motor, into which a stainless-steel ligature wire (0.025 mm diameter) was inserted and tied around the incisor neck. Additionally, another wire (0.020 mm diameter) was looped through the first and second molars and tied to stabilize the device. The maxillary incisors were subsequently cleaned, etched and reinforced with light-cured resin. Postoperatively, gentamicin was administered for three consecutive days to prevent infection and the orthodontic device was checked daily. A constant tensile force of 30 grams was maintained and monitored weekly using an orthodontic force meter (807-006, 3M, United) (Fu, *et al*., 2022, Liu, *et al*., 2023). Fig. 1 displays the experimental model.

### *Determination of hormone levels*

Blood samples were collected from anesthetized rats on the 56th and 84th days and centrifuged to isolate plasma. The levels of LH, GnRH, T, FSH and E2 in plasma were measured using enzyme-linked immunosorbent assay (ELISA). Commercially available ELISA kits were used for all assays, including the Rat LH ELISA Kit (ml002860), Rat GnRH ELISA Kit (ml003038), Rat T ELISA Kit (ml002868), Rat FSH ELISA Kit (ml002872)

and Rat E2 ELISA Kit (ml002871), all supplied by Mlbio, Shanghai, China.

***Measurement of tooth movement distance***

After anesthesia, the rats were euthanized using $CO_2$ and their maxillary tissues were excised and fixed by perfusion with 4% paraformaldehyde on the 84th day. The entire maxillary tissue was excised from the rats and fixed in 4% paraformaldehyde. The molars, along with their surrounding periodontal tissue, were carefully isolated and the distance between the adjacent contact points of the first and second molars was precisely measured using a vernier caliper. Orthodontic tooth movement distances were calculated, with each measurement repeated five times to ensure accuracy (Zou, *et al*., 2019).

***Micro-CT scanning***

A Micro-Computed Tomography (CT) scanner (eXplore Locus, GE, Boston, MD, USA) was utilized to scan the excised maxillae under the following parameters: bin mode 1 × 1, voltage 80 V, current 450 μA, exposure time 2000 ms, 400 views and a resolution of 9 μm. A 1.5 × 1.5 × 1.5 $mm^3$ region of the alveolar bone was reconstructed in three dimensions (3D) to evaluate parameters such as bone mass, trabecular thickness (Tb.Th), bone volume-to-tissue volume ratio (BV/TV) and trabecular separation (Tb.Sp). Data were extracted and analyzed from five representative images to ensure reliability (Ji, *et al*., 2021).

***H&E and IHC assay***

After anesthesia, the rats were perfused with 4% paraformaldehyde and their maxillae, containing three molars, were fixed in the same solution for 24 hours. Subsequently, decalcification was performed using 10% EDTA for three weeks. Serial 6 μm sections were prepared on polylysine-coated slides for further analysis (Jie Shi, 2022). Structural observations were conducted using Hematoxylin and Eosin (H&E) staining (Rintanalert, *et al*., 2024).

Osteoclasts were identified via tartrate-resistant acid phosphatase (TRAP) staining (Wang, *et al*., 2018), with positive cells counted across 10 high-power fields (×200). The average count per sample was recorded as the final result. Periodontal fiber changes under force were evaluated using Masson staining (Jie Shi, 2022). For immunohistochemical (IHC) analysis (Moreno, *et al*., 2024), the sections were deparaffinized and incubated with primary antibodies, including tumor necrosis factor-α (TNF-α) (1:100, ab307164, Abcam, Shanghai, China), OPG (1:100, ab73400, Abcam, Shanghai, China), RANKL (1:100, ab239607, Abcam, Shanghai, China), interleukin-6 (IL-6),(1:100, ab9324, Abcam, Shanghai, China) and interleukin-1β (IL-1β) (1:100, ab283818, Abcam, Shanghai, China). Secondary antibody incubation followed (1:1000, A0216, Beyotime, Shanghai, China; 1:1000, A0208, Beyotime, Shanghai, China). Visualization was performed using a light microscope (BX51, Olympus, Tokyo, Japan). Image J software (Mac OS X, National Institutes of Health, Bethesda, MD, USA) was used for quantitative analysis of stained sections to assess relative expression levels.

***Statistical analysis***

Statistical analysis was performed using SPSS version 22.0 (SPSS, Chicago, IL, USA). Data were presented as mean ± standard error. To compare two independent datasets, a t-test was employed under the assumptions of normal distribution and equal variance. For paired datasets, a rank-sum test was utilized. Statistical significance was defined as $P < 0.05$.

## RESULTS

***MSG injection inhibited the hypothalamic-pituitary-gonadal axis***

Relative to the sham group, the model group displayed evidently dropped sex hormone levels, including E2, GnRH, LH, FSH and T, on both the 56th ($p<0.01$) and 84th day ($p<0.05$), indicating that MSG significantly reduces the levels of sex hormones (Fig. 2A, B).

***MSG administration promoted osteoclast differentiation and alveolar bone resorption***

H&E staining revealed distinct cellular and morphological changes, including reduced blood flow, collagen fiber and matrix degradation, alveolar bone resorption on the compressed side and increased osteoclast differentiation. Notably, the model group showed significant bone destruction compared to the sham group (indicated by arrows in fig. 3A). TRAP staining further highlighted enlarged osteoclasts with red–stained cytoplasm and unstained nucleus (arrow). The model group exhibited a wider periodontal ligament, more osteoclasts and more pronounced bone resorption depressions, whereas only a few red-stained osteoclasts were observed in the sham group (Fig. 3A, B). These findings suggest that MSG significantly impacts bone metabolism through hormonal imbalance.

***MSG injection enhanced periodontal space expansion and osteoblast differentiation and promoted new bone deposition on the tensile side***

H&E staining revealed expanded tensile periodontal spaces, accompanied by increased collagen fibers and matrix proliferation. In the model group, no morphological changes were observed in the periodontal fibers and blood vessels on the tensile side. However, mechanical tension led to fractures in the cementum of the model group.

Masson staining further showed gradual new bone formation along the alveolar bone surface, with newly formed bone extending along the periodontal ligament fibers in the model group (Fig. 4).

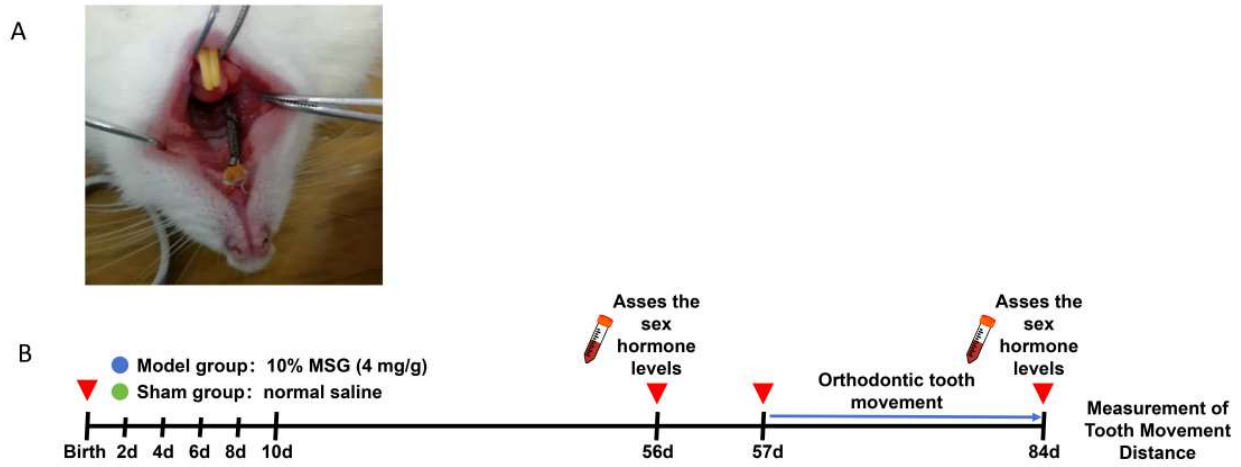


**Fig. 1**: Experimental orthodontic tooth movement model.

(A) Experimental rat model for orthodontic tooth movement with a nickel-titanium spring applying 30-gram force to move the first molar mesially.

(B) Timeline of the experiment. The model group received MSG (4 mg/g) on days 2–10, while the sham group received saline. Hormone levels were measured on day 56, orthodontic devices were installed on day 57, and tooth movement distance was assessed on day 84.

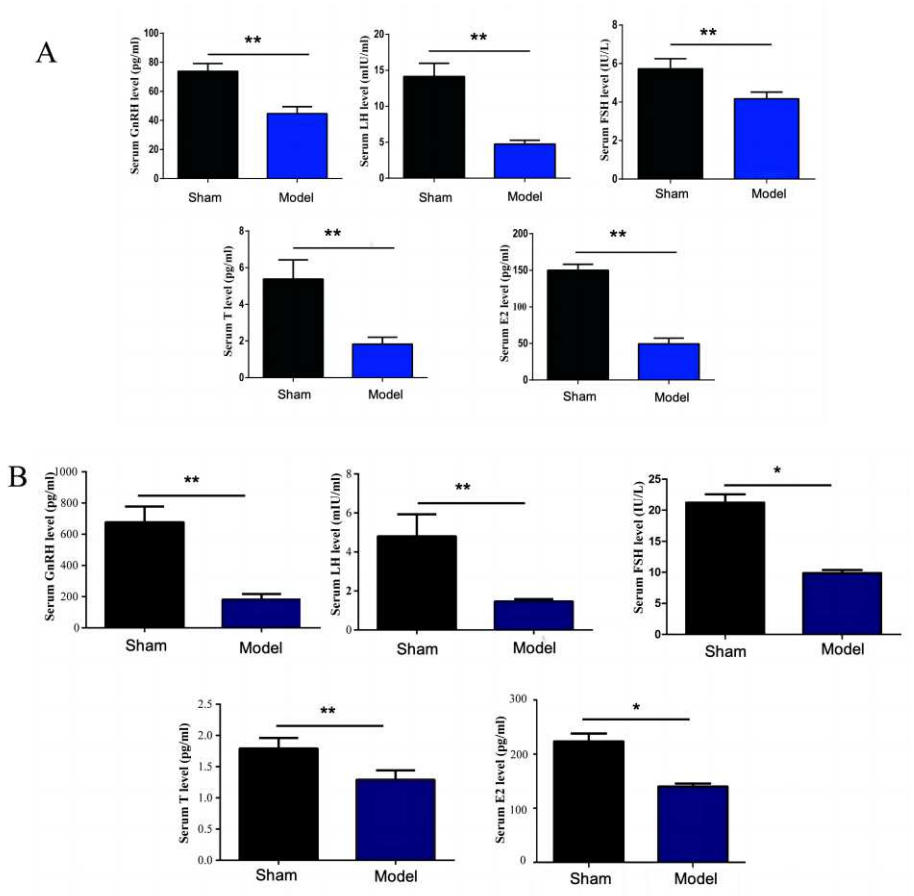


**Fig. 2**: The comparison of GnRH, FSH, LH, E2 and T levels in the serum between model and sham groups (n = 5 per group. *p < 0.05, **p < 0.01). (A) Comparison of hormones on day 56. (B) Comparison of hormones on day 84. GnRH, Gonadotropin Releasing Hormone; FSH, Follicle Stimulating Hormone; LH, luteinizing hormone; E2, estradiol; and T, testosterone.

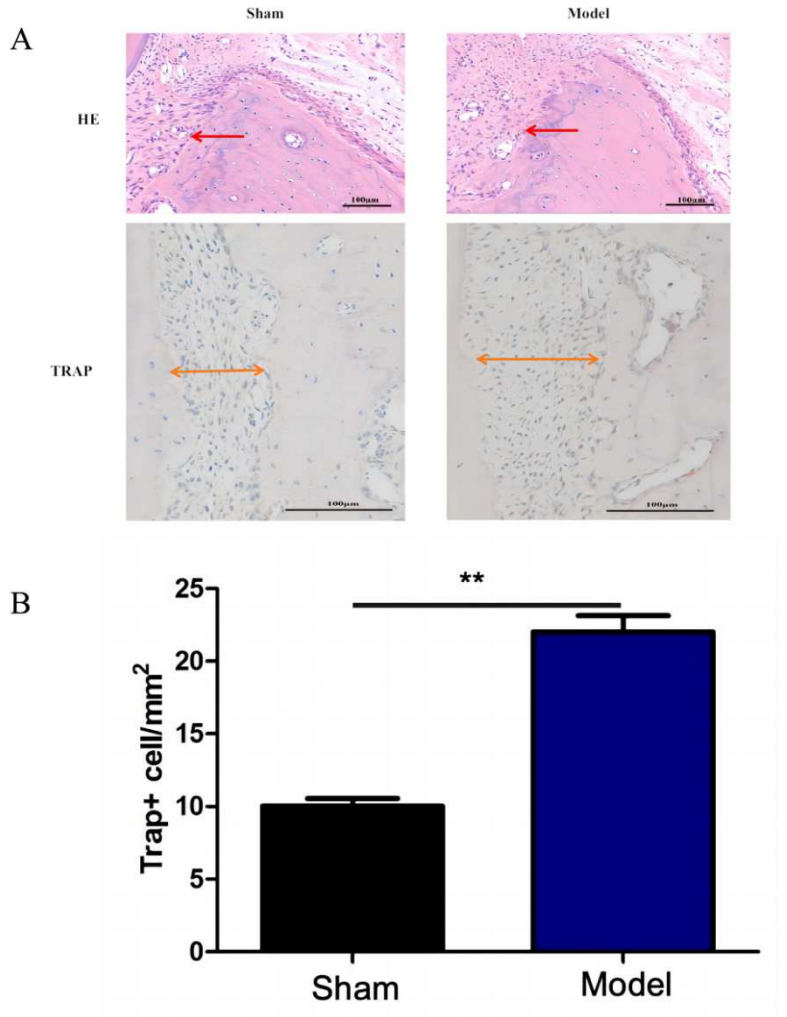


**Fig. 3**: H&E and TRAP staining of periodontal membrane and osteoclasts count on the compressed side.
(A) H&E and TRAP staining of periodontal membrane on the compressed side. (B) Osteoclasts count of TRAP staining. n = 5 per group. $^{**}p < 0.01$. H&E, Hematoxylin and Eosin; TRAP, Tartrate-resistant acid phosphatase. Red arrows: HE staining revealed specific cellular and morphological alterations of the alveolar bone on the compressed side.
Orange arrows: TRAP staining identified enlargement in the osteoclasts.

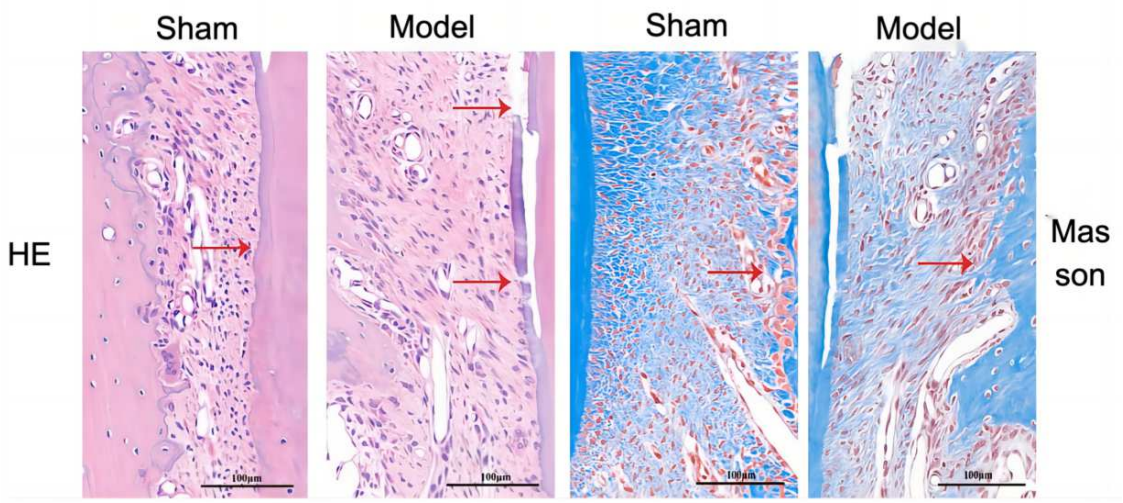


**Fig. 4**: H&E and Masson staining of periodontal membrane on the tensile side.
Arrow: H&E staining revealed expanded tensile periodontal spaces along with elevated collagen fibers and matrix proliferation. Masson staining revealed new bone formation gradually occurring along the surface of the alveolar bone on the tensile side.

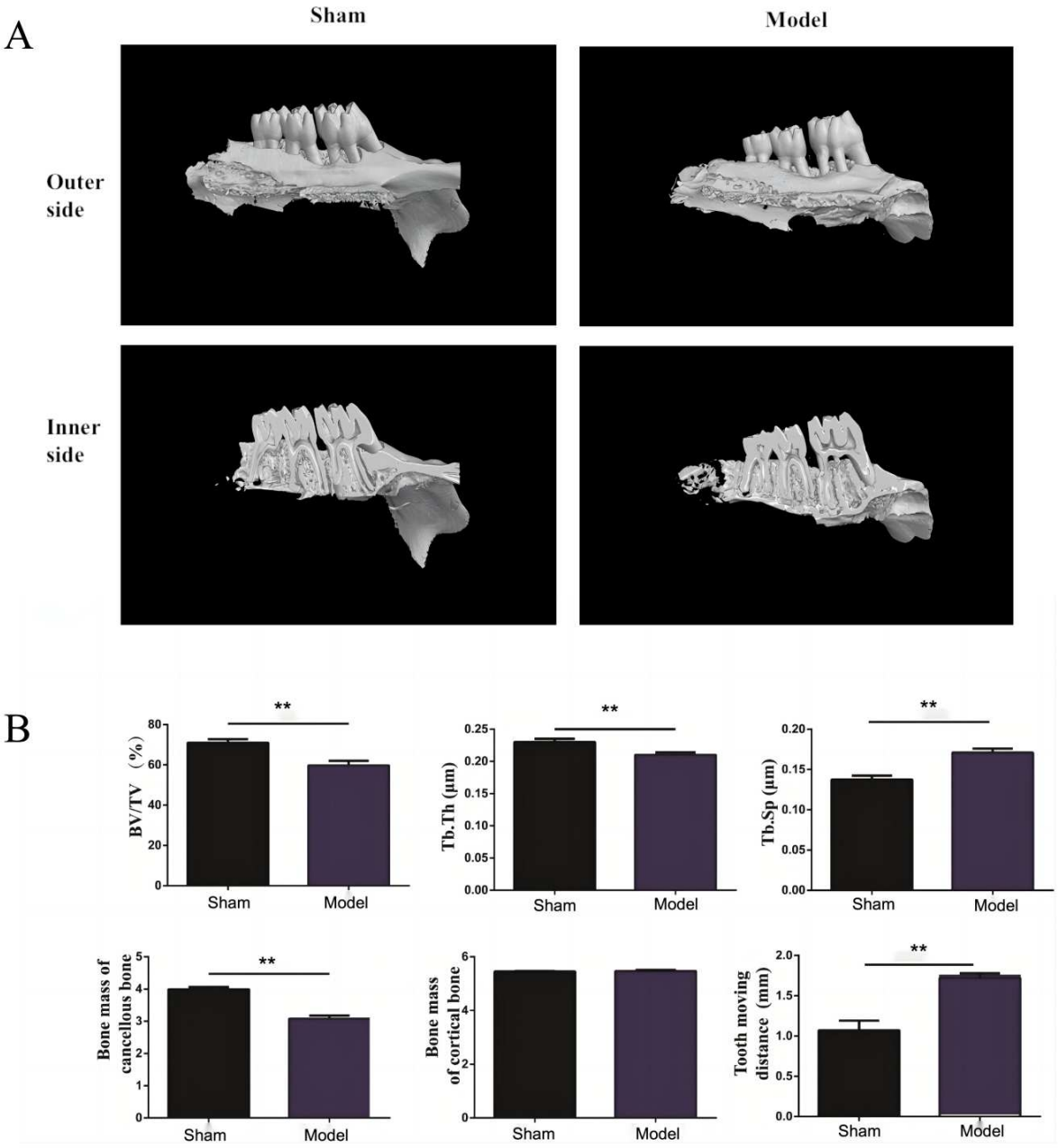


**Fig. 5**: The results of Micro-CT scanning.
(A) Micro-CT images of the maxilla (scale bar = 1 mm). (B) The distance of orthodontic tooth movement, Bone mass, BV/TV, Tb. Th and Tb. Sp (n = 5 per group), $^{**}p < 0.01$. CT, Computed Tomography; BV/TV, bone volume to tissue volume; Tb. Th, trabecular thickness; Tb. Sp, trabecular separation.

### *Micro-CT scanning*

The model group exhibited a significantly larger interdental space compared to the sham group under identical orthodontic forces, as shown by micro-CT scanning. In the model group, alveolar ridge resorption extended to the root bifurcation, exposing the root at the alveolar crest, while the periodontal membrane on the compressed side demonstrated marked changes (Fig. 5A). Analysis of alveolar bone indicated a significant decline in Tb. Th, BV/TV and bone mass ($p < 0.01$), along with an increase in Tb. Sp due to MSG treatment prior to orthodontic force stimulation. The model group also exhibited a significantly larger distance between the first and second molars ($p < 0.01$) (Fig. 5B). Micro-CT results further confirmed a greater periodontal space in the model group.

### *MSG promoted cytokine expression*

Cytokine levels (IL-1β, TNF-α and IL-6) were remarkably raised in the model group on the tensile side, as demonstrated by IHC analysis ($p < 0.05$, Fig. 6).

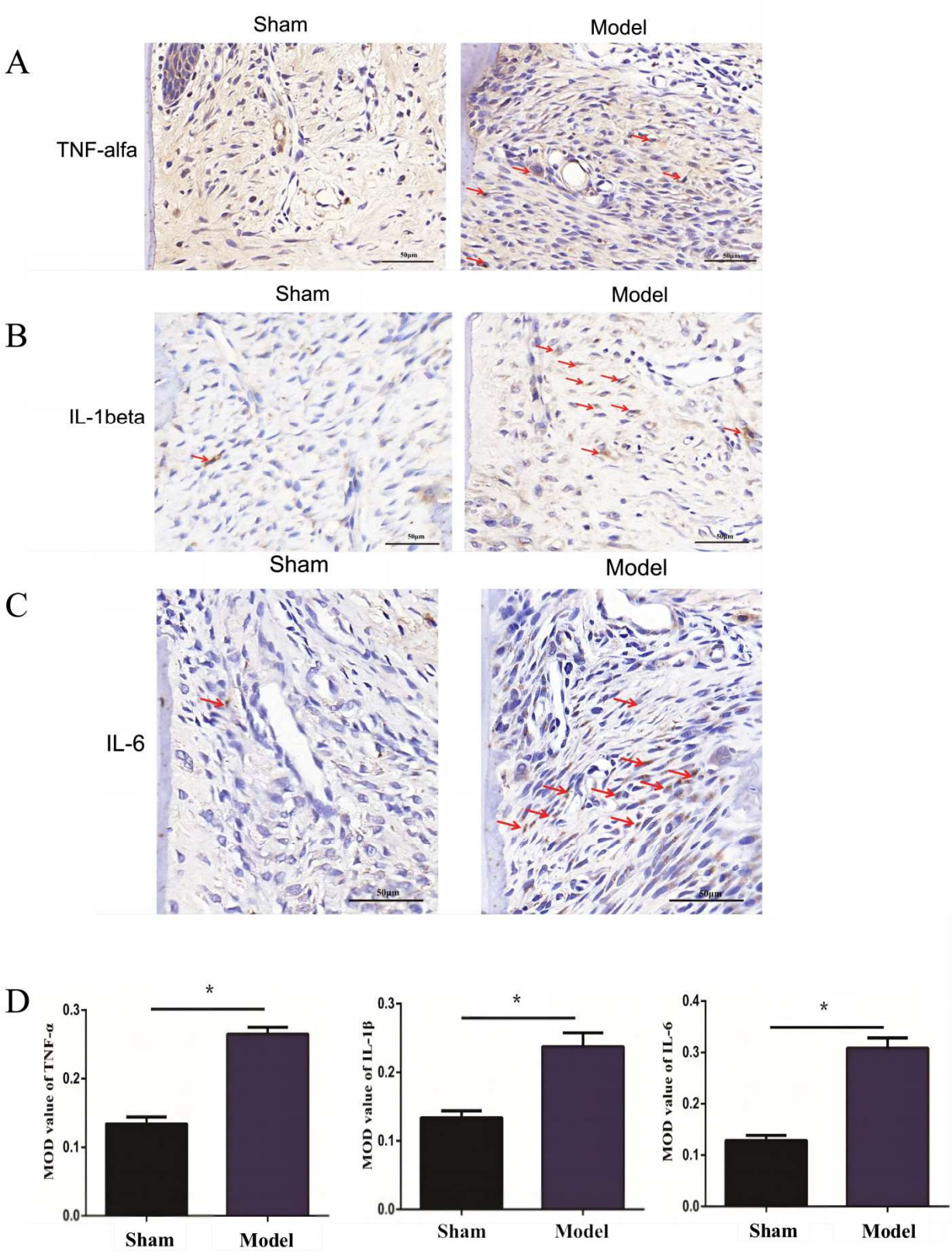


**Fig. 6**: IHC staining and quantitative analysis of TNF-α, IL-1β, and IL-6 on the tensile side.
(A) TNF-α expression on the tensile side. (B) IL-1β expression on the tensile side. (C) IL-6 expression on the tensile side. (D) Quantitative analysis for TNF-α, IL-1β, and IL-6 levels on the tensile side (n = 5 per group, $^{*}p < 0.05$). The arrows in the figure represent positive cells. IHC, immunohistochemical; TNF-α, tumor necrosis factor-α; IL, interleukin-1β.

***MSG increased rankl expression and inhibited opg expression***

RANKL was primarily localized in the membrane and cytoplasm as brown-yellow granules, with high expression in periodontal ligament fibroblasts, osteoblasts and osteoclasts. Both OPG and RANKL exhibited similar positive expression patterns across various cell types, predominantly in the cytoplasm of periodontal ligament cells, osteoblasts and osteoclasts, with osteoblasts showing the highest expression of OPG/RANKL. Compared to the sham group, the model group displayed a significant decrease in OPG expression on both the tensile and compressed sides ($p < 0.05$, fig. 7A, C), along with a marked increase in RANKL expression ($p < 0.05$, fig. 7B, D). Furthermore, the OPG/RANKL ratio was significantly reduced in the model group ($p < 0.05$, fig. 7E, F). These findings indicate that MSG treatment significantly reduced OPG expression and increased RANKL expression, leading to a decrease in the OPG/RANKL ratio, which may promote bone resorption and affect tooth movement.

## DISCUSSION

This study provides evidence that neonatal MSG administration in rats significantly impacts orthodontic tooth movement (OTM) by inhibiting the hypothalamic-pituitary-gonadal (HPG) axis. The findings reveal that MSG disrupts bone remodeling processes, primarily through systemic hormonal effects rather than the direct mechanical force, by increasing osteoclast activity and promoting alveolar bone resorption, as demonstrated by micro-CT and histological analysis. These results align with prior studies suggesting that the HPG axis plays an essential role in bone metabolism and orthodontic response (Salonia, *et al*., 2019).

Our data indicate that MSG exposure significantly reduces sex hormone levels, including GnRH, FSH, LH, E2, and T. The HPG axis is regulated by GnRH release from the hypothalamus, which stimulates the anterior pituitary to secrete LH and FSH. These gonadotropins then promote the production of sex hormones such as T and E2 from the gonads. MSG-induced damage to the hypothalamus, particularly the arcuate nucleus, likely impairs GnRH secretion. This leads to reduced levels of LH and FSH, impaired gonadal function, and suppressed sex hormone production (Nemeroff, *et al*., 1981). Since E2 and T are critical for bone health—where E2 regulates bone resorption by inhibiting osteoclasts and T promotes osteoblast activity—this hormonal imbalance disrupts bone turnover, potentially leading to reduced bone density (Salonia, *et al*., 2019).

This phenomenon may be associated with MSG-induced oxidative stress impairing the function of Kisspeptin neurons in the arcuate nucleus, which are key regulators of GnRH secretion (Xie, *et al*., 2022). In terms of signaling pathways, MSG may inhibit Kisspeptin neuron function by activating the AMPK signaling pathway and disrupting the mechanistic target of rapamycin (mTOR) signaling pathway (Jais and Brüning, 2022). The reduction in GnRH secretion directly impairs the pituitary's ability to produce FSH and LH, ultimately leading to lower E2 and T levels. This finding is consistent with Deng and Guo (2020), who emphasized the critical role of estradiol and testosterone in maintaining bone density by inhibiting osteoclast differentiation. Consequently, the observed upregulation of osteoclast activity and bone resorption in the model group can be attributed to reduced hormonal regulation, which further affects the OPG/RANKL/RANK pathway.

In this study, the model group exhibited a significant increase in RANKL expression alongside a decrease in OPG expression. This imbalance resulted in an elevated RANKL/OPG ratio, a crucial regulator of osteoclastogenesis and bone resorption. (Udagawa, *et al*., 2021). Consistent with Marahleh *et al*. (Marahleh, *et al*., 2019), who reported that elevated RANKL promotes osteoclast differentiation and activation, our findings confirm that MSG exacerbates osteoclast-mediated bone resorption during OTM. This is further supported by Brodetska *et al*. (Brodetska, *et al*., 2020), who highlighted the role of an imbalanced OPG/RANKL pathway in facilitating bone resorption under compressive orthodontic forces.

The model group also exhibited notably higher levels of inflammatory cytokines, such as IL-6, TNF-α and IL-1β. This aligns with studies indicating that pro-inflammatory cytokines contribute to osteoclastogenesis and bone resorption in orthodontic treatments (Zhou, *et al*., 2023, Santamaria-Jr, *et al*., 2020). Cytokine upregulation increases vascular permeability and promotes osteoclast formation, creating conditions conducive to bone resorption (Sims, 2021). The elevated cytokine expression observed here likely results from MSG-induced dysfunction of the HPG axis, leading to increased inflammation and accelerated alveolar bone resorption.

In the MSG-treated group, micro-CT analysis revealed new bone deposition on the tensile side and significant periodontal ligament expansion, consistent with findings that orthodontic forces promote osteoblast differentiation on the tensile side (Jiao, *et al*., 2023, Liu, *et al*., 2022). However, fractures in the cementum of the model group suggest that MSG disrupts the structural integrity of periodontal tissues, potentially increasing susceptibility to mechanical stress and affecting the stability of orthodontic outcomes.

This study is the first to use MSG to disrupt the HPG axis and assess its impact on OTM. Despite its innovation, this study has limitations. The use of a neonatal rat model and the wire ligation method may not fully replicate human orthodontic conditions or endocrine physiology, limiting clinical extrapolation. The small sample size (five rats per

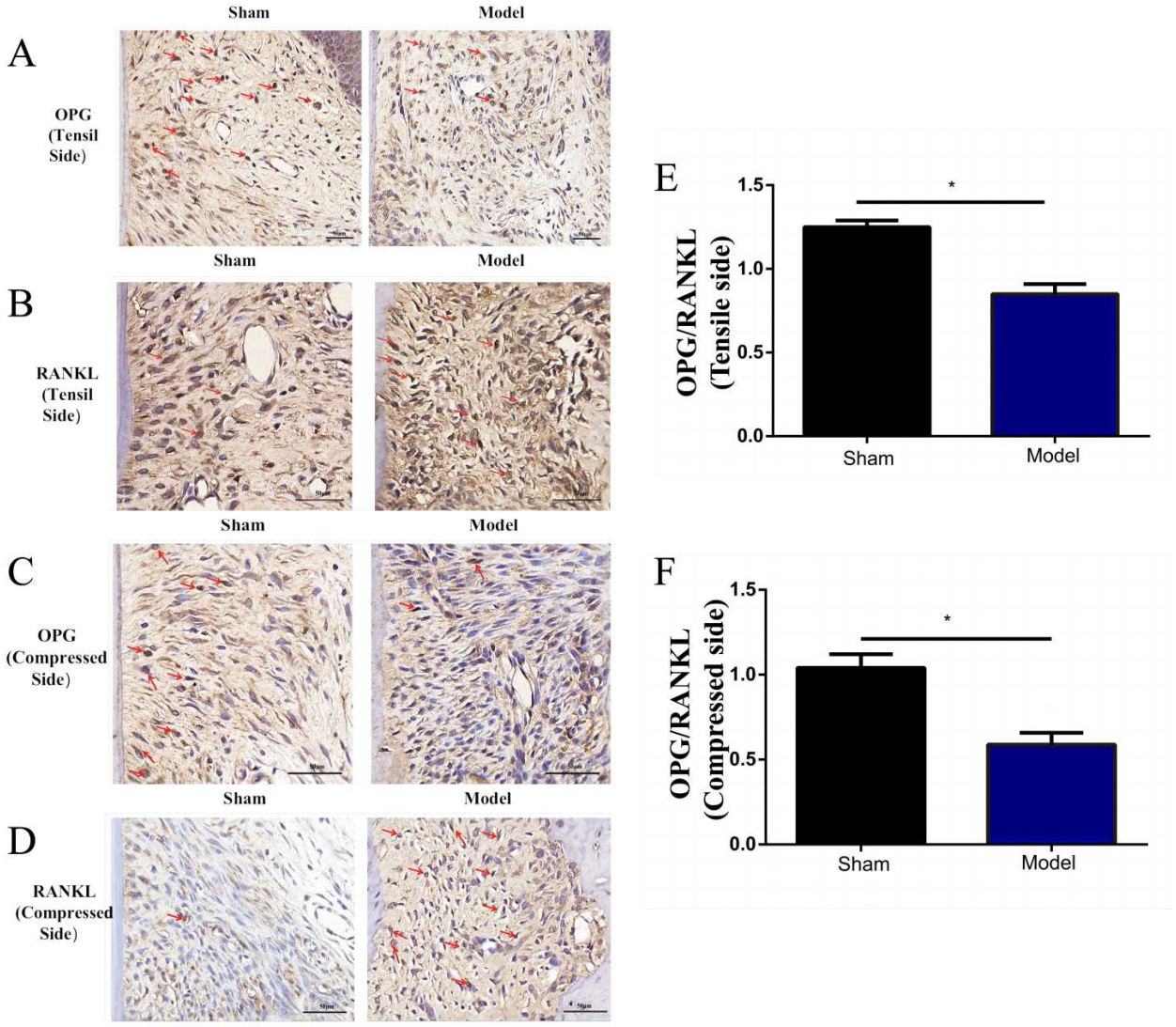


**Fig. 7**: Quantitative analysis of OPG and RANKL using IHC staining.
(A) OPG expression on the tensile side. (B) RANKL expression on the tensile side. (C) OPG expression on the compressed side. (D) RANKL expression on the compressed side. (E) OPG/RANKL on the tensile side, $^{*}p < 0.05$. (F) OPG/RANKL on the compressed side (n = 5 per group, $^{*}p < 0.05$). The arrows in the figure represent positive cells. OPG, osteoprotegerin; RANKL, NF-kappaB ligand.

group) suggests that increasing the sample size would improve statistical power. Finally, the study only evaluated OTM after 56 days of MSG administration, lacking long-term follow-up. The long-term effects of MSG on bone metabolism and orthodontic outcomes remain unclear and merit further investigation. Although speculative, the findings suggest that MSG exposure may have implications for human orthodontic treatment, particularly regarding its potential effects on endocrine regulation and bone metabolism.

## CONCLUSION

MSG-induced HPG axis dysfunction can significantly influence orthodontic outcomes by altering bone metabolism through increased osteoclast activity, decreased OPG expression and elevated inflammatory cytokine levels. These outcomes lay the groundwork for understanding the hormonal regulation's impact on orthodontic tooth movement and suggest that monitoring or modifying endocrine function may enhance orthodontic treatment efficacy and stability. While this study highlights the potential impact of MSG on orthodontic treatment, further validation in human models is needed to confirm these findings and explore their clinical relevance.

***Acknowledgement***
The authors thank the Laboratory of Neurology, Institute of Integrative Medicine, Fudan University, for technical support.

***Authors' contributions***

Xiaofang Cai, Conceptualization, Formal Analysis, Writing - Original Draft, Writing - Review and Editing, Funding acquisition; Min Cai, Methodology, Formal Analysis, Writing - Original Draft, Writing - Review and Editing, Funding acquisition; Yuxiao Zhao, Formal Analysis; Jun Xiang, Formal Analysis; Feng Yang, Data curation; Wen Zhu, Data curation; Wei Bi, Conceptualization, Writing - Review and Editing and Project administration; Xiaojun Ding, Methodology, Writing - Review and Editing, Supervision; All authors given final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

***Funding***

This work was supported by the National Natural Science Foundation of China (Grant no. 81703849), the constructi on of Tertiary Laboratory of Neurophysiology and Pathology & Institute of Internal Medicine, National Adm inistration of Traditional Chinese Medicine (2024XKPT0 5), Shanghai Municipal Key Clinical Specialty TCM Eme rgency (Grant no. shslczdzk04403), and Excellent Youth Foundation of Zhongshan Hospital, Fudan University (Gr ant no. 2019ZSYQ33).

***Data availability statement***

Data is available from the corresponding author upon reasonable request.

***Ethical approval***

All animal handling and care procedures were carried out in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals and the guidelines established by the Animal Care and Use Committee of Fudan University (Approval No. A2020-098).

***Conflict of interest***

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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