VETERINARSKI ARHIV 95 (4), 469-477, 2025 DOI: 10.24099/vet.arhiv.2633

Original scientific paper

The impact of nesfatin-1 on autophagy in brain tissue and dopamine secretion in fasting mice

Siriguleng Yu[1*], Xin Wen[1], Dong Ai[2], Yuchen Luo[1], Ziyi Li[1], Haoyu Bai[1], Wen Yu[1], Xingchuan He[1], Yaru Niu[1], Junjian Jin[1] and Shumin Du[1]

[1]College of Veterinary Medicine, Inner Mongolia Agricultural University, Key Laboratory of Clinical Diagnosis and Treatment Technology for Animal Diseases, Ministry of Agriculture, Hohhot, China
[2]Agriculture and Animal Husbandry Bureau of Alxa League, Alxa league, China

YU, S., X. WEN, D. AI, Y. LUO, Z. LI, H. BAI, W. YU, X. HE, Y. NIU, J. JIN, S. DU: Impact of nesfatin-1 on autophagy in brain tissue and dopamine secretion in fasting mice. Vet arhiv 95, 469-477, 2025.

ABSTRACT

Intermittent fasting is known to affect metabolic and neuroendocrine pathways, and Nesfatin-1, a peptide involved in appetite regulation and metabolism, may play a crucial role in these processes. This study investigates the impact of Nesfatin-1 on brain tissue autophagy and dopamine secretion in fasting mice. Forty C57BL/6 mice were randomly assigned to the control group, 24 h fasting group, 48 h fasting group, Nesfatin-1+24 h fasting group, and Nesfatin-1+48 h fasting group. Following intraperitoneal injections of 1.25 nmol/g Nesfatin-1 after the respective fasting periods, serum levels of Nesfatin-1 and dopamine, as well as mRNA and protein expression of tyrosine hydroxylase and autophagy factors in brain tissues, were assessed using enzyme-linked immunosorbent assay, re- altime quantitative polymerase chain reaction, and Western blot. The results showed significant decreases in serum Nesfatin-1 and dopamine levels (P<0.05) compared to the control group, along with increased mRNA and protein expression of Beclin-1 and microtubule-associated protein 1 light chain 3 in brain tissues (P<0.05), and decreased p62 and tyrosine hydroxylase levels (P<0.01) in fasting groups. However, Nesfatin-1 intervention led to significant increases in serum Nesfatin-1 and dopamine levels (P<0.01), while decreasing Beclin-1 and protein 1 light chain 3 expression (P<0.01), and increasing p62 and tyrosine hydroxylase levels (P<0.01). These findings suggest that acute fasting reduces Nesfatin-1 and dopamine levels, while increasing autophagy in mice, whereas Nesfatin-1 intervention not only effectively diminishes autophagy levels in the brain but also boosts dopamine secretion.

Key words: autophagy; dopamine; fasting; nesfatin-1; tyrosine hydroxylase

Introduction

Nesfatin-1, a recognized satiety peptide, is widely expressed in central and peripheral tissues (OH-I et al., 2006), influencing various physiological functions including the neurological, endocrine, and cardiovascular systems, and lipid metabolism (TEKIN et al., 2019), while also exerting an appetite-suppressing effect (SHIMIZU et al., 2009; SCHALLA and STENGEL, 2018). Research by CHEN et al. (2015) demonstrated that Nesfatin-1 affects dopaminergic neurons, with electrophysiological, electrochemical, and behavioral tests in

* Corresponding author:
Dr Siriguleng Yu, College of Veterinary Medicine, Inner Mongolia Agricultural University, Hohhot, 010018, phone: +86 18247134816, e-mail: srglyu@imau.edu.cn

ISSN 0372-5480
Printed in Croatia

mice following injections in the ventral tegmental area of the midbrain (CHEN et al., 2015). Nota- bly, dopamine neurons in the ventral tegmental area play a key role in the reward system (VAN DEN HEUVEL and PASTERKAMP, 2008), with dopamine serving as a crucial neurotransmitter in regulating feeding reward (JAGER and WITKAMP, 2014). Elevated dopamine concentration is directly linked to reward, influencing behaviors such as addiction and reward. Thus, Nesfatin-1 may modulate appetite by impacting dopamine secretion levels.

Starvation significantly increases appetite in animals; however, short-term fasting has been found to significantly elevate neuronal autophagy in mice, as starvation represents one of the most potent non-genetic inducers of autophagy in the organism (ALIREZAEI et al., 2010). As a result, it remains inconclusive whether Nesfatin-1 affects autophagy and dopamine secretion levels when acting on dopaminergic neurons.

This study aimed to investigate the impact of Nesfatin-1 on autophagy and dopamine secretion in the brain tissue of fasting mice, specifically examining proteins related to fasting, such as tyrosine hydroxylase (TH). Understanding how Nesfatin-1 suppresses appetite will provide a foundation for future research.

Materials and methods

Animals. Forty male C57BL/6 mice, each approximately 6 weeks old and weighing around 20 g, were obtained from Beijing Spearfish Biotechnology in Beijing, China. The mice were housed in a controlled environment with a temperature of (22±2)℃ and a humidity level of (50±10)%. They were kept on a 12 h light/dark cycle and provid- ed with standard laboratory chow and water ad libitum. Each cage housed four mice to minimize stress and allow for adequate social interaction. This study was conducted in accordance with the guidelines approved by the Ethics Committee of Inner Mongolia Agricultural University (Approval no. NND2021034).

Reagents and instruments. The reagents used in this study included Nesfatin-1(Beijing Protein Innovation, Beijing, China), mice enzyme-linked immunosorbent assay (ELISA) kit (Jiangsu Meibiao Biotechnology, Jiangsu, China), Total RNA extraction kit (Axygen, NY, USA), Prime Script™ RT Master Mix (Perfect Real Time) (Takara, Dalian, China), TB Green Premix Ex Taq (Perfect Real Time) (Takara, Dalian, China), antibodies (Abcam, Cambridge, UK) against protein 1 light chain 3 (LC3) (Anti-LC3A/B antibody, ab62721) at 1μg/mL, Beclin-1 (Anti-Beclin I antibody, ab62557) at 1μg/mL, p62 (Anti-SQSTM1/p62 antibody, ab91526) at 1μg/mL and tyrosine hydroxylase (TH) (Anti-Tyrosine Hydroxylase antibody, ab137721) at 1/500 dilution. The main instruments used in this study included a Multi-function Enzyme Labeler (Bio-Rad; Hercules, CA, USA), a 4℃ centrifuge (Thermo Fisher Scientific, Waltham, MA, USA), a Real-Time PCR system (Applied Biosystems, Foster City, CA, USA), and a TU-1901 ultraviolet spectrophotometer (Beijing puxi, Beijing, China).

Grouping and induction of the fasting model. The mice were acclimatized for one week prior to the experiment and had ad libitum access to food and water during this period. The forty mice were randomly assigned to five groups: control group (n=8), 24 h fasting group (n=8), 48 h fasting group (n=8), Nesfatin-1+24 h fasting group (n=8), and Nesfatin-1+48 h fasting group (n=8). The mice in the respective fasting groups were deprived of food for 24 or 48 h with access to water; those in the Nesfatin-1 groups received a single intraperitoneal injection of 1.25 nmol/g body weight of Nesfatin-1, which was diluted in sterile phosphate-buffered saline (PBS) to a final concentration of 1 nmol/μL. The control group received an equal volume (1 mL/kg body weight) of sterile PBS. The injections were administered immediately after the fasting period concluded.

ELISA. Four hours after the completion of the fasting period, the mice were anesthetized using an intraperitoneal injection of sodium pentobarbital for euthanasia. This method involved administering a dose of 60 mg/kg body weight, ensuring deep anesthesia prior to the exsanguination process. Blood samples were collected from the retro-orbital venous plexus. Serum was separated by centrifugation at 3000 rpm for 15 min and stored at -80℃. The concentrations of Nesfatin-1 and dopamine in

the serum were measured using ELISA kits following the manufacturer's instructions. All mice were sacrificed at the same time, and no food was reintroduced to the cages between the end of the fasting period and euthanasia.

Quantitative real-time PCR (qPCR). Total RNA was extracted from the homogenized mouse brain tissue using a protocol based on the Total RNA extraction kit (Axygen Scientific). The brain tissue was minced and homogenized with Buffer R, followed by the addition of Buffer R and centrifugation. After mixing with isopropanol, the RNA was precipitated, washed with Buffers W1A and W2, and finally eluted in Buffer TE. Subsequent cDNA synthesis was performed using the Takara Reverse Transcription Kit. The mRNA levels of TH, Beclin-1, LC3, and p62 were analyzed using RT-qPCR with β-actin as a reference gene. Gene specific primers are shown in Table 1 (Table 1). The RT-qPCR amplification program consisted of pre-denaturation at 95℃ for 30 sec, followed by 45 cycles of denaturation at 95℃ for 10 sec, and annealing at 60℃ for 34 sec. Each sample was run in triplicate, and the expression level was analyzed using the $2^{-\Delta\Delta CT}$ method.

Western Blotting. Total proteins from mouse brain tissues were extracted and quantified using the BCA method. The expression levels of TH, Beclin-1, LC3, and p62 were assessed by Western blotting with β-actin as a loading control. Each sample was run in triplicate to ensure reproducibility. Image J software was used to analyze the protein expression levels on the basis on the intensity of the bands.

Statistical Analysis. Data analysis was performed using GraphPad Prism 8.0 software. The results were expressed as mean ± standard deviation and analyzed by one-way analysis of variance (ANOVA). A significance level of $P<0.01$ was considered highly significant, $P<0.05$ as significant, and $P\geq0.05$ as non-significant.

Results

Nesfatin-1 and dopamine levels. To investigate the effect of fasting on serum levels of Nesfatin-1 and dopamine, we measured the levels in mice subjected to 24 h and 48 h of fasting. The results demonstrated that both Nesfatin-1 and dopamine levels were significantly reduced in the fasting groups compared to the control group ($P<0.05$). However, following the administration of Nesfatin-1, there was a highly significant increase in serum levels of both Nesfatin-1 and dopamine compared to the fasting groups ($P<0.01$) (Fig. 1a,1b).

TH mRNA Levels. The relative mRNA expression of the TH gene in the brain tissues of mice in the 24 h and 48 h fasting groups was significant- ly lower compared to the control group ($P<0.01$) (Fig. 2). This decrease may reflect impaired dopa- mine synthesis due to prolonged fasting. Impor- tantly, after the intervention with Nesfatin-1, TH mRNA levels in the brain tissues of fasting mice

Table 1. RT-qPCR primer sequences

Gene	Gene Accession		Primer sequences (5'→3')
β-actin	NM_007393.5	Forward:	GCTACAGCTTCACCACCACA
		Reverse:	TCTCCAGGGAGGAAGAGGAT
Beclin-1	NM_001359820.1	Forward:	ACAGTGGACAGTTTGGCACA
		Reverse:	CGGCAGCTCCTTAGATTTGT
LC3	NM_025735.3	Forward:	ACCAAGCCTTCTTCCTCC
		Reverse:	GCCTAACAAAATGACAAACCCCACAG
P62	NM_001290769.1	Forward:	GGCCTATCTTCTGGGCAAGG
		Reverse:	AAAAGGCAACCAAGTCCCCA
TH	NM_009377.2	Forward:	GGTATACGCCACGCTGAAGG
		Reverse:	TAGCCACAGTACCGTTCCAGA

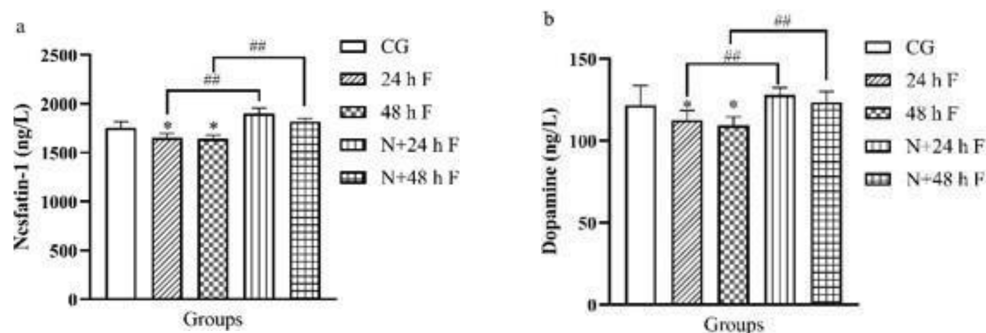

Fig. 1. Nesfatin-1 and dopamine levels

Control group (CG), 24 h fasting group (24 h F), 48 h fasting group (48 h F), Nesfatin-1+24 h fasting group (N+24 h F), and Nesfatin-1+48 h fasting group (N+48 h F). (a) Serum levels of Nesfatin-1 in each group of mice, (b) Serum levels of dopamine in each group of mice. Compared to the negative control, *P<0.05; Comparing between indicated groups, ##P<0.01.

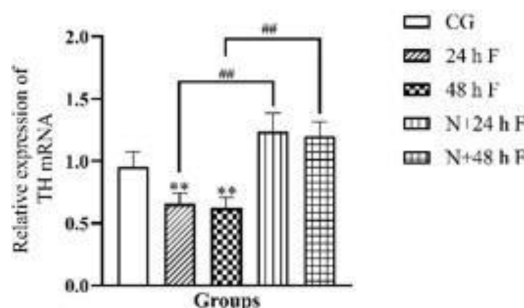

Fig. 2 Tyrosine hydroxylase (TH) mRNA levels

Control group (CG), 24 h fasting group (24 h F), 48 h fasting group (48 h F), Nesfatin-1+24 h fasting group (N+24 h F), and Nesfatin-1+48 h fasting group (N+48 h F). Comparison of the TH mRNA levels. Compared to the negative control, **P<0.01. Comparing between indicated groups, ##P<0.01.

were significantly elevated compared to the corresponding fasting groups (P<0.05 and P<0.01, respectively) (Fig. 2).

Autophagy gene mRNA levels. Compared to the control group, the mRNA expression of autophagy factors Beclin-1 and LC3 was significant- ly increased in mice in the 24 h and 48 h fasting groups, while p62 mRNA expression was significantly decreased (P<0.05 and P<0.01, respectively) (Fig. 3a). Following intervention with Nesfatin-1, Beclin-1 and LC3 mRNA expression in mice was significantly reduced, and p62 mRNA expression was significantly increased compared to the 24 h and 48 h fasting groups (P<0.01 and P<0.05, respectively) (Fig. 3b, 3c).

TH protein expression. The relative expression of TH protein in the brain tissues of mice in the 24 h and 48 h fasting groups was highly significant- ly lower compared to the control group (P<0.01). This supports our earlier findings regarding mRNA expression and further implicates fasting in the reduction of dopamine biosynthesis. However, after intervention with Nesfatin-1, the relative expression of TH protein in the brain tissues of mice was significantly higher than that in the 24 h and 48 h fasting groups (P<0.01) (Fig. 4a, 4b).

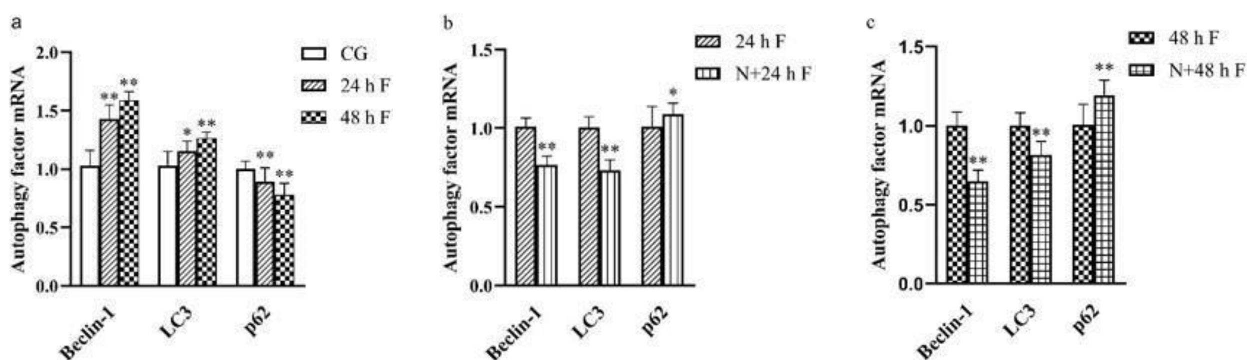

Fig. 3 Autophagy gene mRNA levels

Control group (CG), 24 h fasting group (24 h F), 48 h fasting group (48 h F), Nesfatin-1+24 h fasting group (N+24 h F), and Nesfatin-1+48 h fasting group (N+48 h F). (a) Relative mRNA expression of autophagy genes compared between the control group and fasting group, (b) Relative mRNA expression of autophagy genes in mice after 24 h of fasting with Nesfatin-1 intervention, (c) Relative mRNA expression of autophagy genes in mice after 48 h of fasting with Nesfatin-1 intervention. Compared to the negative control, *P<0.05; **P<0.01.

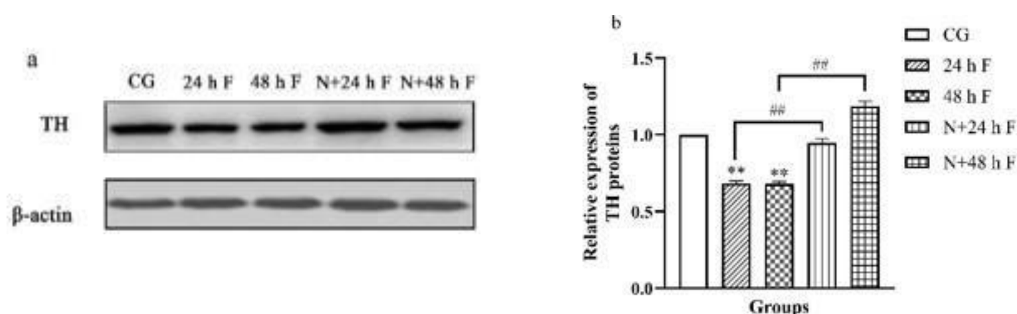

Fig. 4 Tyrosine hydroxylase (TH) expressions

Control group (CG), 24 h fasting group (24 h F), 48 h fasting group (48 h F), Nesfatin-1+24 h fasting group (N+24 h F), and Nesfatin-1+48 h fasting group (N+48 h F). (a) Western Blot results, (b) Comparison of TH protein expression levels. Compared to the negative control, *P<0.01. Comparing between indicated groups, ##P<0.01.

Autophagy Protein Expression. The expression levels of autophagy-related proteins, including Beclin-1 and p62, were measured using Western blot analysis (Fig. 5a). The results indicated that, compared to the control group, the protein levels of Beclin-1 in the brain tissues of mice in the 24 h and 48 h fasting groups showed a significant increase (P<0.01), while the protein levels of p62 showed a significant decrease (P<0.01). However, after the intervention with Nesfatin-1, the protein levels of Beclin-1 and p62 in the brain tissues of mice showed a significant decrease and increase, respectively, compared to the 24 h and 48 h fasting groups (P<0.01) (Fig. 5b, 5c).

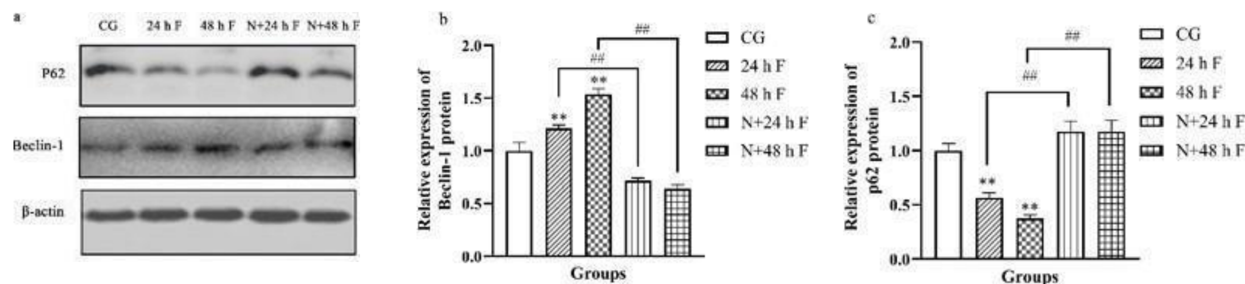

Fig. 5 Autophagy protein expressions

Control group (CG), 24 h fasting group (24 h F), 48 h fasting group (48 h F), Nesfatin-1+24 h fasting group (N+24 h F), and Nesfatin-1+48 h fasting group (N+48 h F). (a) Western Blot results, (b) Comparison of Beclin-1 protein expression levels, (c) Comparison of p62 protein expression levels. Compared to the negative control, **P<0.01. Comparing between indicated groups, ##P<0.01.

Discussion

The identified satiety factor Nesfatin-1, derived from the precursor protein Nucleobindin 2(NUCB2) (OZTURK, 2020), is known to be regulated by fasting and satiety, with circulating levels increasing postprandial and decreasing during fasting (MOGHARNASI et al., 2019). Studies have shown that Nesfatin-1 inhibits feeding behavior and controls weight gain (GONZALEZ et al., 2010), and intraperitoneal injection of Nesfatin-1 results in a feeding inhibitory effect that can last up to 24 h in multiple model organisms.(RIVA et al., 2011). According to the study conducted by Oh-I et al (OH-I et al., 2006), Nesfatin-1 demonstrated a significant reduction in the food intake of mice when administered via intraperitoneal injection at a dose of 1.25 nmol/g. Building upon this finding, our study aimed to investigate the underlying mechanism of Nesfatin-1's appetite-suppressing effect by administering an intraperitoneal injection of Nesfatin-1 at a concentration of 1.25 nmol for every gram of body weight to fasted mice. The results showed a significant reduction in serum Nesfatin-1 levels in mice fasted for 24 h and 48 h compared to the control group, con- sistent with previous studies (RIVA et al., 2011; MOGHARNASI et al., 2019). However, following intraperitoneal injection of Nesfatin-1, we observed a significant increase in serum levels. While this increase reflects the pharmacokinetics of Nesfatin-1, it does not directly indicate regulation of satiety. Further investigation is needed to elucidate how elevated serum Nesfatin-1 interacts with appetite regulation pathways.

Dopamine, an important neurotransmitter in the regulation of ingestive reward, is known to be involved in food intake and energy balance, therefore regulating feeding behavior and appetite control (JAGER and WITKAMP, 2014). TH, the rate-limiting enzyme in dopamine synthesis, is responsive to levels of dopamine secretion (LI et al., 2023). The findings revealed a noteworthy trend: compared to the control group, serum dopamine levels were notably reduced in mice subjected to 24 h and 48 h fasting periods. Strikingly, follow- ing Nesfatin-1 intervention, the serum dopamine levels in the aforementioned fasted mice exhibited a substantial increase. Concurrently, post-Nesfa- tin-1 administration, there was an extremely significant upsurge in the expression of TH mRNA and protein in the brain tissues of the fasting mice. These outcomes indicate that Nesfatin-1 potentially modulates appetite by impacting dopamine secretion levels, but further investigation is need- ed to understand the pathway and mechanism of action.

Acute fasting promotes autophagy in the body (ALIREZAEI et al., 2010; DODSON et al., 2013), making it a major defense mechanism against material and energy metabolism disorders. Short-

term fasting can induce increased appetite in animals, but it also significantly elevates neuronal autophagy in mice (ALIREZAEI et al., 2010). Autophagy, a lysosomal degradation process and a protective mechanism, regulates the homeosta- sis of the intracellular environment (YANG and KLIONSKY, 2010), and is sensitive to chang- es in the body's glucose metabolism. This study aimed to investigate the effect of Nesfatin-1 on autophagy in the brain tissues of fasted mice, spe- cifically examining its impact on the expression of autophagy-related factors, including Beclin-1, LC3, and p62 mRNA and protein, while also assessing alterations in dopamine secretion levels.

Beclin-1 is a critical regulator in the early stages of autophagy, and plays a pivotal role in promoting autophagy (KANG et al., 2011). Meanwhile, LC3 serves as a marker of autophagosome formation and reflects the progression of autophagy (LV et al., 2021; ZHANG et al., 2018). Conversely, P62 is a marker protein indicative of autophagic activity (NAKAHIRA and CHOI, 2013). It undergoes continuous degradation in the cytoplasm when autophagy is active, while impaired autophagy or defective autophagic function leads to the accumulation of p62. Elevated p62 levels are typically regarded as a sign of inhibited autophagic activity.

Our results analysis showed that fasting augments the autophagy activity in mouse brain tissue, and the degree of autophagy exhibited time-dependent characteristics. Previous stud- ies similarly observed upregulated levels of autophagy-related markers in mice following 24 h of fasting, with an even greater enhancement in these markers after 48 h of fasting in mouse hepatocytes and cortical neurons (ALIREZAEI et al., 2010). However, Nesfatin-1 intervention resulted in a notable decline in Beclin-1 and LC3 mRNA, and protein expression and a substantial increase in p62 expression compared to the fasting group. This outcome indicated that Nesfatin-1 interven- tion down-regulated the level of autophagy in the brain tissue of fasted mice, aligning with previous research. Several studies have demonstrated that Nesfatin-1 diminishes intracellular LC3 protein levels and elevates p62 expression, supporting the notion that Nesfatin-1 inhibits organismal autophagy (NAZARNEZHAD et al., 2019; XU et al., 2020). Consequently, Nesfatin-1 might mitigate the impact of fasting on organismal brain tissue by inhibiting autophagy, increasing dopamine secretion, and subsequently modulating appetite. However, further in-depth investigations are required to elucidate the underlying mechanisms, particularly regarding the specific signaling pathways involved in the regulation of autophagy and dopamine levels by Nesfatin-1.

Conclusions

In summary, our study revealed that Nesfa- tin-1 not only reduces the level of autophagy in the brain tissue of fasted mice, but also enhances their dopamine secretion. These findings set the stage for deeper exploration of the mechanism underlying Nesfatin-1's appetite-suppressing ef- fects.

Ethics approval

This study was conducted in accordance with the guidelines approved by the Ethics Committee of Inner Mongolia Agricultural University (Approval no. NND2021034).

Financial support statement

This study received support from the Na- tional Natural Science Foundation of China (No. 32160821, 31860693).

Authorship contribution statement

Siriguleng Yu and Xin Wen contributed equaly to this work.

SY conceived and designed the study; YL, ZL, XW and WY carried out the experimental work; HB, XH and YN analyzed the experimental data; XW and SY contributed to the writing. All authors critically reviewed and approved the final manuscript.

Declaration of competing of interest

There is no conflict of interest.

References

ALIREZAEI, M., C. C. KEMBALL, C. T. FLYNN, M. R. WOOD, J. L. WHITTON, W. B. KIOSSES (2010): Short-term fasting induces profound neuronal autophagy. Autophagy 6, 702-710.
https://doi.org/10.4161/auto.6.6.12376

CHEN, X., X. SHU, Z. K. CONG, Z. Y. JIANG, H. JIANG (2015): Nesfatin-1 acts on the dopaminergic reward pathway to inhibit food intake. Neuropeptides 53, 45-50.
https://doi.org/10.1016/j.npep.2015.07.004

DODSON, M., V. DARLEY-USMAR, J. ZHANG (2013): Cellular metabolic and autophagic pathways: traffic control by redox signaling. Free Radic. Biol. Med. 63, 207-221.
https://doi.org/10.1016/j.freeradbiomed.2013.05.014

GONZALEZ, R., B. KERBEL, A. CHUN, S. UNNIAPPAN (2010): Molecular, cellular and physiological evidences for the anorexigenic actions of nesfatin-1 in goldfish. PLoS One 5, e15201.
https://doi.org/10.1371/journal.pone.0015201

JAGER, G., R. F. WITKAMP (2014): The endocannabinoid system and appetite: relevance for food reward. Nutr. Res. Rev. 27, 172-185.
https://doi.org/10.1017/S0954422414000080

KANG, R., H. J. ZEH, M. T. LOTZE, D. TANG (2011): The Beclin 1 network regulates autophagy and apoptosis. Cell Death Differ. 18, 571-580.
https://doi.org/10.1038/cdd.2010.191

LI, Y., X. WANG, X. XIE, Q. LIU, H. DONG, Y. HOU, Q. XIA, P. ZHAO (2023): Enhanced locomotor behaviour is mediated by activation of tyrosine hydroxylase in the silkworm brain. Insect Mol. Biol. 32, 251-262.
https://doi.org/10.1111/imb.12828

LV, S., H. LIU, H. WANG (2021): Exogenous Hydrogen Sulfide Plays an Important Role by Regulating Autophagy in Diabetic-Related Diseases. Int. J. Mol. Sci. 22, 6715.
https://doi.org/10.3390/ijms22136715

MOGHARNASI, M., H. TAHERICHADORNESHIN, S. A. PAPOLI-BARAVATI, A. TEYMURI (2019): Effects of upper-body resistance exercise training on serum nesfatin-1 level, insulin resistance, and body composition in obese paraplegic men. Disabil. Health J. 12, 29-34.
https://doi.org/10.1016/j.dhjo.2018.07.003

NAKAHIRA, K., A. M. CHOI (2013): Autophagy: a potential therapeutic target in lung diseases. Am. J. Physiol. Lung Cell. Mol. Physiol. 305, L93-L107.
https://doi.org/10.1152/ajplung.00072.2013

NAZARNEZHAD, S., M. RAHMATI, A. SHAYANNIA, Z. ABBASI, M. SALEHI, M. KHAKSARI (2019): Nesfatin-1 protects PC12 cells against high glucose-induced cytotoxicity via inhibiting oxidative stress, autophagy and apoptosis. Neurotoxicology 74, 196-202.
https://doi.org/10.1016/j.neuro.2019.07.001

OH-I, S., H. SHIMIZU, T. SATOH, S. OKADA, S. ADACHI, K. INOUE, H. EGUCHI, M. YAMAMOTO, T. IMAKI, K. HASHIMOTO, T. TSUCHIYA, T. MONDEN, K. HORIGUCHI, M. YAMADA, M. MORI (2006): Identification of nesfatin-1 as a satiety molecule in the hypothalamus. Nature 443, 709-712.
https://doi.org/10.1038/nature05162

OZTURK, O. G. (2020): Effects of Nesfatin-1 on Food Intake and Hyperglycemia. J. Am. Coll. Nutr. 39, 345-351.
https://doi.org/10.1080/07315724.2019.1646678

RIVA, M., M. D. NITERT, U. VOSS, R. SATHANOORI, A. LINDQVIST, C. LING, N. WIERUP (2011): Nesfatin-1 stimulates glucagon and insulin secretion and beta cell NUCB2 is reduced in human type 2 diabetic subjects. Cell Tissue Res. 346, 393-405.
https://doi.org/10.1007/s00441-011-1268-5

SCHALLA, M. A., A. STENGEL (2018): Current Understanding of the Role of Nesfatin-1. J. Endocr. Soc. 2, 1188-1206.
https://doi.org/10.1210/js.2018-00246

SHIMIZU, H., S. OH-I, K. HASHIMOTO, M. NAKATA, S. YAMAMOTO, N. YOSHIDA, H. EGUCHI, I. KATO, K. INOUE, T. SATOH, S. OKADA, M. YAMADA, T. YADA, M. MORI (2009): Peripheral administration of nesfatin-1 reduces food intake in mice: the leptin-independent mechanism. Endocrinology 150, 662-671.
https://doi.org/10.1210/en.2008-0598

TEKIN, T., B. CICEK, N. KONYALIGIL (2019): Regulatory Peptide Nesfatin-1 and its Relationship with Metabolic Syndrome. Eurasian J. Med. 51, 280-284.
https://doi.org/10.5152/eurasianjmed.2019.18420

VAN DEN HEUVEL, D. M., R. J. PASTERKAMP (2008): Getting connected in the dopamine system. Prog. Neurobiol. 85, 75-93.
https://doi.org/10.1016/j.pneurobio.2008.01.003

XU, K., Z. ZHANG, M. CHEN, S. MOQBEL, Y. HE, C. MA, L. JIANG, Y. XIONG, L. WU (2020): Nesfatin-1 Promotes the Osteogenic Differentiation of Tendon-Derived Stem Cells and the Pathogenesis of Heterotopic Ossification in Rat Tendons via the mTOR Pathway. Front. Cell Dev. Biol. 8, 547342.
https://doi.org/10.3389/fcell.2020.547342

YANG, Z., D. J. KLIONSKY (2010): Eaten alive: a history of macroautophagy. Nat. Cell Biol. 12, 814-822.
https://doi.org/10.1038/ncb0910-814

ZHANG, X., T. D. EVANS, S. J. JEONG, B. RAZANI (2018): Classical and alternative roles for autophagy in lipid metabolism. Curr. Opin. Lipidol. 29, 203-211.
https://doi.org/10.1097/MOL.0000000000000509

Received: 11 March 2024
Accepted: 20 September 2024
Online publication: 30 April 2025

YU, S., X. WEN, D. AI, Y. LUO, Z. LI, H. BAI, W. YU, X. HE, Y. NIU, J. JIN, S. DU: Učinak nesfatina 1 na autofagiju moždanog tkiva i lučenje dopamina u miševa nakon gladovanja. Vet. arhiv 95, 469-477, 2025.

SAŽETAK

Poznato je da povremeno gladovanje (engl. *intermittent fasting*) utječe na metaboličke i neuroendokrine puteve a nesfatin 1, peptid koji sudjeluje u regulaciji apetita i metabolizmu, može biti ključan u tim procesima. U radu je istražen utjecaj nesfatina 1 na autofagiju moždanog tkiva i lučenje dopamina u miševa nakon gladovanja. Četrdeset C57BL/6 miševa nasumično je podijeljeno u kontrolnu skupinu, skupinu miševa koji su gladovali 24 sata, skupinu miševa koji su gladovali 48 sati, skupinu miševa nesfatin 1 + 24 h gladovanja i skupinu miševa nesfatin 1+ 48 h gladovanja. Nakon intraperitonealnih injekcija od 1,25 nmol/g nesfatina 1, primijenjenog poslije odgovarajućeg razdoblja gladovanja, određene su serumske razine nesfatina 1 i dopamina, kao i izražaj mRNA i proteina tirozin-hidroksilaze te čimbenika autofagije u moždanim tkivima. Pri navedenom korištene su metode ELISA, PCR u stvarnom vremenu (qPCR) i western blot. U skupinama miševa koji su gladovali rezultati su pokazali znakovito sniženje serumskih razina nesfatina 1 i dopamina ($P<0,05$) u usporedbi s kontrolnom skupinom, zajedno s povećanim izražajem mRNA i proteina Beclin 1 te lakog lanca 3 proteina 1 povezanog s mikrotubulima u moždanim tkivima. Također u skupinama miševa koji su gladovali utvrđena je snižena razina p62 i tirozin - hidroksilaze ($P<0,01$). Dodatak nesfatina 1 doveo je, međutim, do znakovitog porasta serumskih razina nesfatina 1 i dopamina ($P<0,01$), kao i smanjenja izražaja proteina Beclin 1 i lakog lanca 3 proteina 1($P<0,01$) te porasta razine p62 i tirozin-hidroksilaze ($P<0,01$). Ovi rezultati pokazuju da u miševa produljeno gladovanje snižava razine nesfatina 1 i dopamina te povećava autofagiju moždanog tkiva, dok dodatak nesfatina 1 ne samo da učinkovito smanjuje autofagiju u mozgu već i potiče lučenje dopamina.

Ključne riječi: autofagija; dopamine; post; nesfatin 1; tirozin-hidroksilaza