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Original Article

Effects of lncRNA uc.48+ siRNA on the release of CGRP in the spinal cords of rats with diabetic neuropathic pain

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Abstract: Long noncoding RNA (lncRNA) and factors influencing lncRNA expression are related to the nervous system diseases. The aims of the project are to study the effect of lncRNA uc.48+ siRNA on calcitonin gene related peptide (CGRP) release in the spinal cords (SCs) of diabetic neuropathic pain (DNP) rats to identify its possible mechanism and to provide new experimental evidence for the prevention and treatment of DNP. Male Sprague-Dawley rats were used to create a DNP rat model by feeding the rats a high-fat and fructose diet in addition to an intraperitoneal injection of streptozocin. Fasting blood glucose (FBG), mechanical withdrawal threshold (MWT) and thermal withdrawal latency (TWL) were measured to select the DNP rats. The DNP rats were randomly divided into the following 4 groups: (1) a normal control group (Control), (2) a DNP rats treated with saline group (DNP), (3) a DNP rats treated with uc.48+ siRNA group (DNP + uc.48+ siRNA) and (4) a DNP rats treated with scrambled siRNA group (DNP + scramble siRNA). After intrathecal injection of uc.48+ small interfering RNA, the MWT and TWL of the DNP group significantly decreased compared to the Control group, but after the injection of uc.48+ small interfering RNA, the MWT and TWL of the DNP rats significantly increased ($P<0.01$, ANOVA test). The application of the methods of qPCR and WB produced results that revealed that the expressions of lncRNA uc.48+, CGRP, IL-1β and TNF-α in the SCs of the DNP group were much higher than those in the Control group ($P<0.01$, ANOVA test), but the expressions of these molecules in the DNP + uc.48+ siRNA group significantly decreased compared with the DNP group ($P<0.01$, ANOVA test). The phosphorylations of p38 and ERK1/2 in the DNP group were significantly enhanced compared with the Control group, whereas uc.48+ siRNA significantly reduced the increased phosphorylations of p38 and ERK1/2 pathway in the SCs of the DNP rats ($P<0.01$, ANOVA test). ELISA results revealed that uc.48+ siRNA significantly decreased the high levels of IL-1β and TNF-α in the sera of the DNP rats ($P<0.01$, ANOVA test). Therefore, lncRNA uc.48+ may play an important role in the transmission of DNP by promoting the release of CGRP in the SC. Small interfering lncRNA uc.48+ might alleviate the hyperalgesia and allodynia of DNP rats by suppressing the release of CGRP in the SCs of DNP rats, which might inhibit the phosphorylations of p38 and ERK1/2 and suppress the release of IL-1β and TNF-α in the SCs of DNP rats.

Keywords: Diabetic neuropathic pain, lncRNA uc.48+, calcitonin gene related peptide, spinal cord

Introduction

Diabetes is a metabolic disease that is characterized by chronic hyperglycemia. Diabetes affected more than 382 million people worldwide in 2013, and the number is expected to reach 592 million by the end of 2035 [1]. Diabetes patients often suffer from complications, for example cardiovascular disease, retinopathy, neuropathy, etc. [2]. Diabetic neuropathic pain (DNP) is one of the major clinical symptoms of diabetic neuropathy and often manifests in the distal limbs, especially in the lower limbs, as a skin burning pain accompanied by spontaneous pain, a feeling of allergies and plant nerve dysfunction [3]. DNP is poorly controlled by analgesics and requires high doses of opioids, which triggers side effects and exerts substantial influences on the patient's quality of life [4].

Although human protein-coding genes represent only approximately 1.5% of the DNA, up to 90% of eukaryotic genomes are transcribed, which generates a large population of non-coding RNAs (ncRNAs) [5, 6]. Long noncoding RNAs

(lncRNAs) are ≥ 200 nt long and are an abundant class of non-protein coding RNAs that are transcribed in complex, sense-and antisense patterns from the intergenic and intronic regions of the mammalian genome. At present, it has been confirmed that some lncRNAs participate in physiological processes that maintain cellular and tissue homeostasis [5, 7, 8]. Moreover, changes in the expression of lncRNAs and factors influencing lncRNA expression are related to nervous system diseases [9, 10].

It has been reported that sensory neurons that express calcitonin gene related peptide (CGRP) in the trigeminal ganglion (TN) or dorsal root ganglion (DRG) might play a role in nociceptive afferent input transmission [11-13]. When sensory axons exhibit reflexes following the activation of primary afferent neurons, the release of CGRP in spinal cord (SC) may contribute to inflammatory or pain responses [14-17]. Therefore, CGRP plays an important role in chronic neuropathic pain.

The uc.48+ (http://genome.ucsc.edu/cgi-bin/hgc?hgsid=42796671_glIKDUiygua FXCbRBiWSM7gFOgqn&c=chr2&o=20462844&t=20463142&g=ct_Ultra_7128&i=%28null%29+uc.48) is a lncRNA and is involved in the expression of the $P2X_3$ receptor in the DRG in DNP rats [18, 19]. The aim of this research is to observe the effects of lncRNA uc.48+ siRNA on CGRP release in the spinal cords in DNP rats.

Materials and methods

Animals and animal groups

Male Sprague Dawley rats (180-230 g) were provided by the Center of Laboratory Animal Science of Nanchang University. The procedures were approved by the Animal Care and Use Committee of Nanchang University Medical School. The animals were housed in plastic boxes in groups of three at 21-25°C. The ethical guidelines of the International Association for the Study of Pain (IASP) for pain research in animals were followed. Diabetic rat models were induced by an intraperitoneal (i.p.) injection of streptozotocin (STZ) (i.e., diabetes was induced by high-calorie food and a single STZ (65-mg/kg) injection in the rats) [10, 19]. STZ was dissolved in citrate buffer (pH 4.5). The control animals received only the buffer. After 7 days, blood samples were obtained from the tail vein, and glycemia was determined using a glucometer. A non-fasting blood glucose>200 mg/dl (11.1 mM) was considered indicative of DM. The mechanical withdrawal threshold (MWT) and the thermal withdrawal latency (TWL) of the DM rats were measured after STZ injection on the weekends for 3 weeks. Those rats for which the MWT and TWL were less than 60% of the baseline were selected as the DNP rats.

The DNP rats were randomly divided into four groups: (1) a normal control group (Control); (2) a DNP rats treated with saline group (DNP); (3) a DNP rats treated with uc.48+ siRNA group (DNP + uc.48+ siRNA); and (4) a DNP rats treated with scrambled siRNA group (DNP + scramble siRNA).

The siRNA specific for uc.48+ was purchased from Invitrogen (Carlsbad, CA), and the target sequence was 5'-GGCACTACTACTTGCAGAA-3'. The siRNA oligonucleotides that specifically targeted uc.48+ were used in this experiment. The uc.48+ was knocked down by RNA interference (RNAi) using an Entranster™-in vivo Transfection Reagent [19].

Each rat was anesthetized with 10% chloral hydrate anesthesia (0.3 mL/100 g) during the intrathecal injection, which was positioned at the L4-6 lumbar segments and performed with a 16 G coarse needle to pierce into the subarachnoid space. As Mestre et al. reported, we took the typical rat tail lashing and jittering to indicate lateral puncture success [20]. Then, uc.48+ siRNA and scramble siRNA were dissolved in 20 mL saline as vehicle and applied once a day at a dosage of 1.0 nmol/kg for intrathecal injection for 3 days, while 20 mL saline was used in group DNP in the same manner.

Measurement of the mechanical withdrawal threshold (MWT)

Noxious-pressure stimulation was used to evaluate mechanical hyperalgesia. Unrestrained rats were placed inside a clear plastic chamber (22×12×22 cm) on a stainless steel mesh floor and allowed to acclimate. Withdrawal responses to mechanical stimulation were determined using calibrated von Frey filaments (BME-403, Tianjin) that were applied through an opening in the stainless steel mesh floor of the cage (grid 1×1 cm^2) to an area adjacent to the paw. Each von Frey filament was applied once starting

with a 0.13-g filament and continuing until a withdrawal response occurred or the force reached 20.1 g (i.e., the cut-off value). The hind paws were tested alternately at 2 min intervals. Three measurements were taken using the up-and-down method on each side, and the lowest value was taken as the threshold value. The filaments were applied in order of increasing bending force (0.13, 0.20, 0.33, 0.60, 1.30, 3.60, 5.00, 7.30, 9.90 and 20.1 g) with each filament applied ten times at intervals of 15 s to different parts of the midplantar glabrous skin. The strength of the filaments in the series that evoked at least five positive responses among the ten trials was determined as the pain threshold. When the value extended beyond 20.1 g, the value was recorded as 20.1 g.

Measurement of the thermal withdrawal latency (TWL)

Noxious heat stimulation was applied with a Thermal Paw Stimulation System (BME-410C, Tianjin) for the assessment of thermal hyperalgesia. The rats were placed in a transparent, square, bottomless acrylic box (22×12×22 cm), on a glass plate under which a light was located. Radiant heat stimuli were applied by directing a beam of light at the foot pad of each hind paw through the glass plate. The light beam was turned off automatically when the rat lifted the paw, which allowed for the measurement of the time between the beginning of the application of the light beam and the elevation of the foot. This time was designated the paw withdrawal latency. The hind paws were tested alternately at 5 min intervals. The cut-off time for the heat stimulation was 25 s.

Quantitative real-time PCR

After the intrathecal injection of uc.48+ siRNA or scrambled siRNA for 3 days, the rats of each group were anesthetized and killed. The rats were decapitated after anesthetization with urethane [1.2 g/kg, intraperitoneally (i.p.)] on day 5 post-injection. The L4-L5 lumbar SC was removed from the inner side of each half of the dissected vertebra with fine dissecting forceps and transferred immediately into phosphate-buffered saline (PBS). Total RNA was isolated with an RNA simple Total RNA Kit (TIANGEN, Beijing, China) according to the manufacturer's protocol. The cDNA synthesis was performed with a TransScript First Strand cDNA Synthesis Super Mix (TransGen, Beijing, China). The cDNA was stored at 4°C after reaction for 30 min at 42°C. The primers were designed with the Primer Express 3.0 software (Applied Biosystems), and the sequences were as follows: uc.48+, sense: 5'-GTGGCGTAAGTGAATGTCCT-3', and antisense: 5'-GTTGGCAGTTCTGCAAGTAG-3'; CGRP, sense: 5'-GTCATCGCTCACCAGGGAG-G-3', and antisense: 5'-CACACCGCTTAGATCTG-GGG-3'; IL-1β, sense: 5'-AGGCTGACAGACCCCA-AAAG-3', and antisense: 5'-CTCCACGGGCAAG ACATAGG3';TNF-α:sense:5'-CACGTCGTAGCAAA-CCACCAA-3', and antisense: 5'-GTTGGTTGTCTT-TGAGATCCAT-3'; and β-actin, sense: 5'-TAAAGA-CCTCTATGCCAACACAGT-3', and antisense: 5'-C-ACGAT GGAGGGGCCGGACTCATC-3'. Quantitative PCR was performed with the SYBR® Green MasterMix in an ABI PRISM® 7500 Sequence Detection System (Applied Biosystems, Inc., Foster City, CA, USA). The thermal cycling parameters were as follows: 95°C for 30 s; followed by 40 cycles of amplification at 95°C for 5 s and 65°C for 34 s; 95°C for 15 s; 60°C for 1 min; and 95°C for 15 s. The dissolution curve was used to determine the specificity of the amplification, and the results were processed with the software within the ABI7500 PCR instrument. The quantification of gene expression was performed using the ΔΔCT calculation with CT as the threshold cycle. The relative levels of target genes, normalized to the sample with the lowest CT, are given as $2^{-\Delta\Delta CT}$.

Western blotting analysis

The four groups of rats were anesthetized and killed. The L4-L5 SC segments were isolated and flushed with ice-cold PBS. The SC segments were homogenized by mechanical disruption in lysis buffer (for the detection of phosphorylated substrates containing phosphatase inhibitors) and incubated on ice for 40 min. The homogenate was then pelleted at 6000 g for 10 min, and the supernatant was collected. The quantity of total protein was determined in the supernatant with the Lowry method. After diluton with sample buffer (100 mmol/L TrisCl, 200 mmol/L dithiothreitol, 4% sodium dodecylsulfate (SDS), 0.2% bromophenol blue, 20% glycerol) and heating to 95°C for 10 min, the samples containing equal amounts of protein (20 μg) were separated through SDS-polyacrylamide gel electrophoresis using a Bio-Rad system and 12% gel. In the wake of electrophoretic transfer onto PVDF membrane using the same system, the membrane was blocked with 5% non-fat dry milk in 25 mmol/L Tris buffered

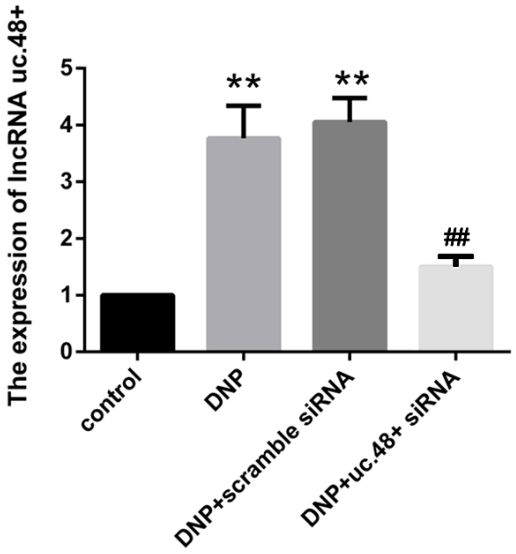

Figure 1. Effects of lncRNA uc.48+ siRNA on the expressions of lncRNA uc.48+ in the SCs tested by qPCR. The expressions of lncRNA uc.48+ in the SCs were evaluated by qPCR. N=6 per group, **P<0.01 *vs*. Control group; ##P<0.01 *vs*. DNP group.

saline, pH 7.2, plus 0.1% Tween 20 (TBST) for 3 h at room temperature followed by incubation with the primary antibody (rabbit anti-CGRP, anti-p38, anti-p-p38, anti-ERK1/2, or anti-p-ERK1/2; 1:500; Chemicon International Company of America) in the same buffer overnight at 4°C. The membrane was then washed in TBST and incubated with the secondary antibody, i.e., horseradish peroxidase (HRP)-conjugated goat anti-rabbit IgG (1:1000, Beijing Zhongshan Biotech. Co.) in the same buffer for 1 h at room temperature. After a final wash in TBST, chemiluminescent signals were collected on autoradiography film using an enhanced chemiluminescence (ECL) kit (Shanghai Pufei Biotech. Co.). The quantificaton of the band intensities was performed with the AlphaImager 2200 software. The band densities were normalized to each β-actin internal control.

Measurement of serum IL-1β and TNF-α

The serum IL-1β and TNF-α levels were quantified using enzyme-linked immunosorbent assay (ELISA) with commercially available antibodies according to the protocol provided by the supplier (Senxiong Company, Shanghai, China). The reactions were recorded using an ELISA reader (Rayto, RT-6000, USA) at 450 nm. The concentrations of IL-1β and TNF-α were determined based on a standard curve [21].

Statistical analysis

All results are expressed by mean ± S.E.M. Differences between treatment groups were analyzed with Student's t-tests or, where appropriate, ANOVAs followed by Dunnett's posthoc tests for multiple comparisons. A *P-value* <0.05 was considered statistically significant.

Results

Changes in uc.48+ expression in the SCs of the rats

The expression of lncRNA uc.48+ in the SC was tested by qPCR. The expression of lncRNA uc.48+ in the SC of the DNP group increased significantly compared with the Control group, whereas the expression of lncRNA uc.48+ in the SC of the rats treated with uc.48+ siRNA decreased by 60% compared with the DNP group ($F(3, 20)=1370.496$, $P=0.000$, ANOVA test). See **Figure 1**.

Effects of uc.48+ siRNA on mechanical and thermal hyperalgesia in DNP rats

The mechanical withdrawal threshold (MWT) in the DNP rats at d 5 after intrathecal injection was lower than that in the Control rats, whereas the MWT in the DNP + uc.48+ siRNA group was significantly increased compared with that in the DNP group ($F(3, 20)=551.233$, $P=0.000$, ANOVA test), which indicated a relief of the mechanical hyperalgesia due to uc.48+ siRNA. There were no differences in the MWTs between the DNP group and the DNP + scramble siRNA group ($P=0.851$, Student's t-test). See **Figure 2**.

The thermal withdrawal latency (TWL) in the DNP rats at d 5 after intrathecal injection was lower than that in the Control rats. The TWL in the DNP + scramble siRNA group exhibited no difference relative to the DNP group ($F(3, 20)=281.265$, $P=0.000$, ANOVA test). The TWL in the DNP + uc.48+ siRNA group was significantly increased compared with that in the DNP group ($P=0.335$, Student's t-test). See **Figure 2**.

Effects of uc.48+ siRNA on the expression levels of CGRP mRNA and protein in the SC of DNP rats

The expression of CGRP mRNA within the SC was observed via qPCR. There were no differences in the intensities of CGRP mRNA expression between the DNP + scramble siRNA group

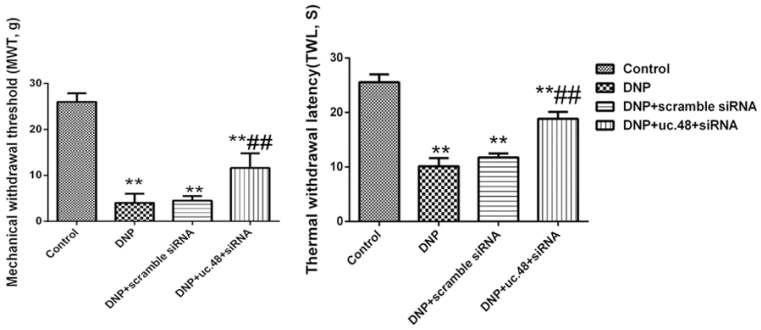

Figure 2. Effects of lncRNA uc.48+ siRNA on the mechanical or thermal hyperalgesia of rats. The mechanical withdrawal threshold (MWT) and the thermal withdrawal latency (TWL) of rats in the four groups were measured. N=6 per group, **P<0.01 vs. Control group; ##P<0.01 vs. DNP group.

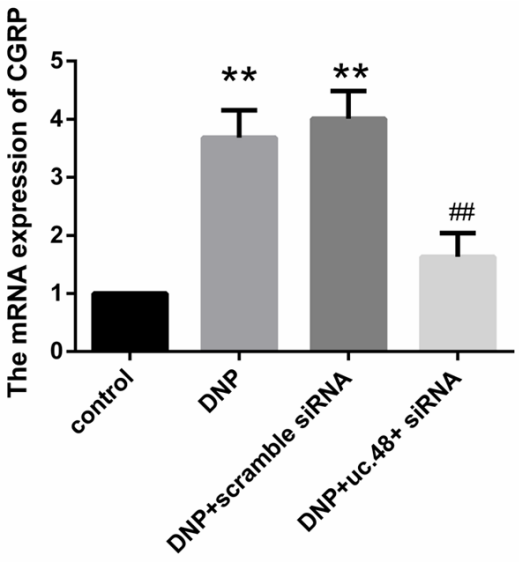

Figure 3. Effects of lncRNA uc.48+ siRNA on the expression of CGRP mRNA in the SCs tested by qPCR. The expressions of CGRP mRNAs in the SCs of each group were evaluated by qPCR. N=6 per group, **P<0.01 *vs.* Control group; ##P<0.01 *vs.* DNP group.

and the DNP group. The expressions of CGRP mRNA in the DNP + uc.48+ siRNA group were significantly lower than those in the DNP group (F(3, 20)=908.402, P=0.000, ANOVA test). See **Figure 3**.

We additionally observed the expression of CGRP within the SC via Western blotting. There was no difference in the intensity of CGRP expression between the DNP + scramble siRNA group and the DNP group. The expression of CGRP in the DNP group was obviously higher than that in the Control group, and CGRP expression in the DNP + uc.48+ siRNA group was significantly lower than that in the DNP group (F(3, 20)=1321.799, P=0.000, ANOVA test). See **Figure 4**.

Effects of uc.48+ siRNA on the concentrations of IL-1β and TNF-α in the DNP rats' SCs and sera

The expressions of IL-1β and TNF-α mRNAs within the SC were observed via qPCR. There were no differences in the intensities of IL-1β or TNF-α mRNA expression between the DNP + scramble siRNA group and the DNP group. The expressions of the IL-1β and TNF-α mRNAs in the DNP + uc.48+ siRNA group were significantly lower than those in the DNP group (for IL-1β: F(3, 20)=125.501, P=0.000; for TNF-α: F(3, 20)=355.965, P=0.000, ANOVA test). See **Figure 5**.

The serum levels of IL-1β and TNF-α were measured using ELISA kits. The serum levels of IL-1β and TNF-α in the DNP rats were significantly higher than those in the Control group (P<0.01). The serum levels of IL-1β and TNF-α in the DNP + uc.48+ siRNA group were significantly decreased compared with rats in the DNP group (for IL-1β: F(3, 20)=372.253, P=0.000; for TNF-α: F(3, 20)=725.005, P=0.000, ANOVA test). No differences in the serum levels of IL-1β or TNF-α were observed between the DNP + scramble siRNA group and the DNP group. See **Figure 6**.

Effects of uc.48+ siRNA on the phosphorylations of P38 and ERK1/2 in the SCs of the DNP rats

Western blotting of the SCs revealed that the phosphorylation of P38 in the DNP group was stronger than that in the Control group; however, the phosphorylation of P38 in the DNP + uc.48+ siRNA group was much lower than that in the DNP group (F(3, 20)=433.490, P=0.000, ANOVA test). There was no significant change of the total P38 MAPK protein among the four groups (F(3, 20)=3.107, P=0.94, ANOVA test). See **Figure 7**.

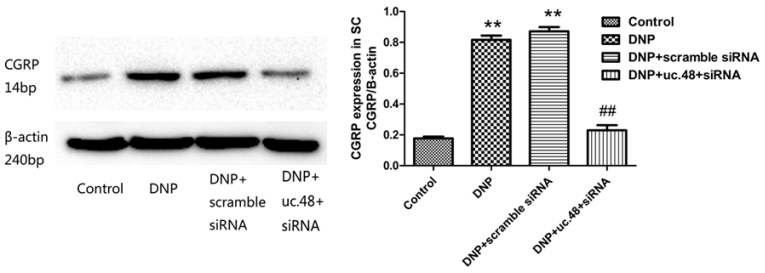

Figure 4. Effects of lncRNA uc.48+ siRNA on the expression of CGRP protein in the SCs tested by WB. The expressions of CGRP protein in the SCs of each group were evaluated by WB. N=6 per group, **P<0.01 *vs*. Control group; ##P<0.01 *vs*. DNP group.

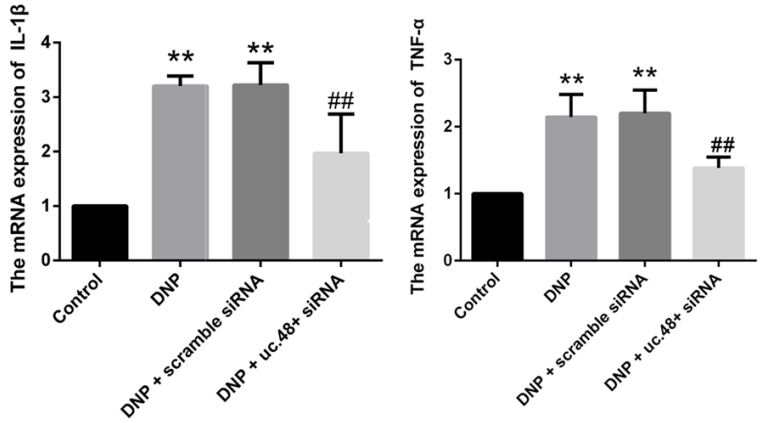

Figure 5. Effects of lncRNA uc.48+ siRNA on the levels of IL-1β and TNF-α mRNAs in the SCs tested by qPCR. The expressions of IL-1β and TNF-α mRNAs in the SCs of each group were evaluated by qPCR. N=6 per group, **P<0.01 *vs*. Control group; ##P<0.01 *vs*. DNP group.

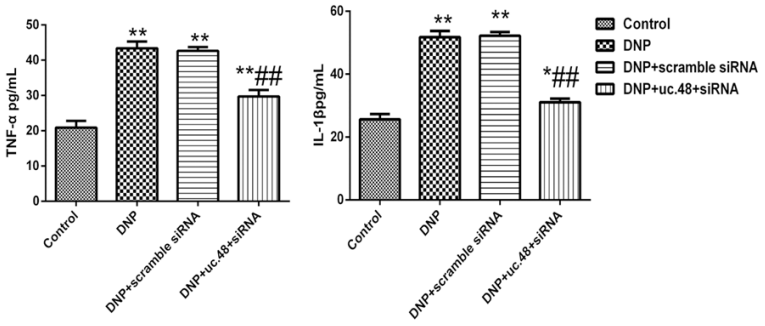

Figure 6. Effects of lncRNA uc.48+ siRNA on the levels of IL-1β and TNF-α mRNAs of sera tested by ELISA. The levels of IL-1β and TNF-α in sera of each group were evaluated by qPCR. N=6 per group, *P<0.05 vs. Control group; **P<0.01 *vs*. Control group; ##P<0.01 *vs*. DNP group.

Western blotting also revealed that the phosphorylation of ERK1/2 in the DNP group was stronger than that in the Control group, and the phosphorylation of ERK in the DNP + uc.48+ siRNA was much lower than that in the DNP group (F(3, 20)=1206.349, P=0.000, ANOVA test). There was no significant change of the total ERK1/2 MAPK protein among the four groups (F(3, 20)=0.734, P=0.56, ANOVA test). See **Figure 7**.

Discussion

Diabetic neuropathic pain is one of the most common complications of type 2 diabetes. Studies have demonstrated that CGRP plays a key role in peripheral inflammatory and neuropathic models of pain [22]. Ellington et al. investigated the effects of cannabinoids on the capsaicin-evoked release of CGRP from the rat paw skin in vitro by comparing non-diabetic and streptozotocin-induced diabetic animals, and the results revealed that diabetes caused a greater than two-fold increase in the basal and capsaicin-evoked CGRP release [23]. The releases of substance P and CGRP in the superficial dorsal horn of the spinal cord would facilitate pain transmission in states of neuropathic pain and cancer pain [17, 24]. Our research also found that CGRP mRNA and protein in the L4/5 segment of the spinal cord were significantly elevated in the DNP rats compared with the control rats.

LncRNA is a type of transcription that results in a functional RNA molecule with an unit length of more than 200 nt that is located in the nucleus or cytoplasm, lacks the ability to encode proteins, and is widely involved in the pathological process of DNP [10, 21]. Our laboratory has found that the expression level of lncRNA uc.48+ in the sera of diabetes mellitus (DM) patients is increased, and further experiments confirmed that uc.48+ siRNA treatment may

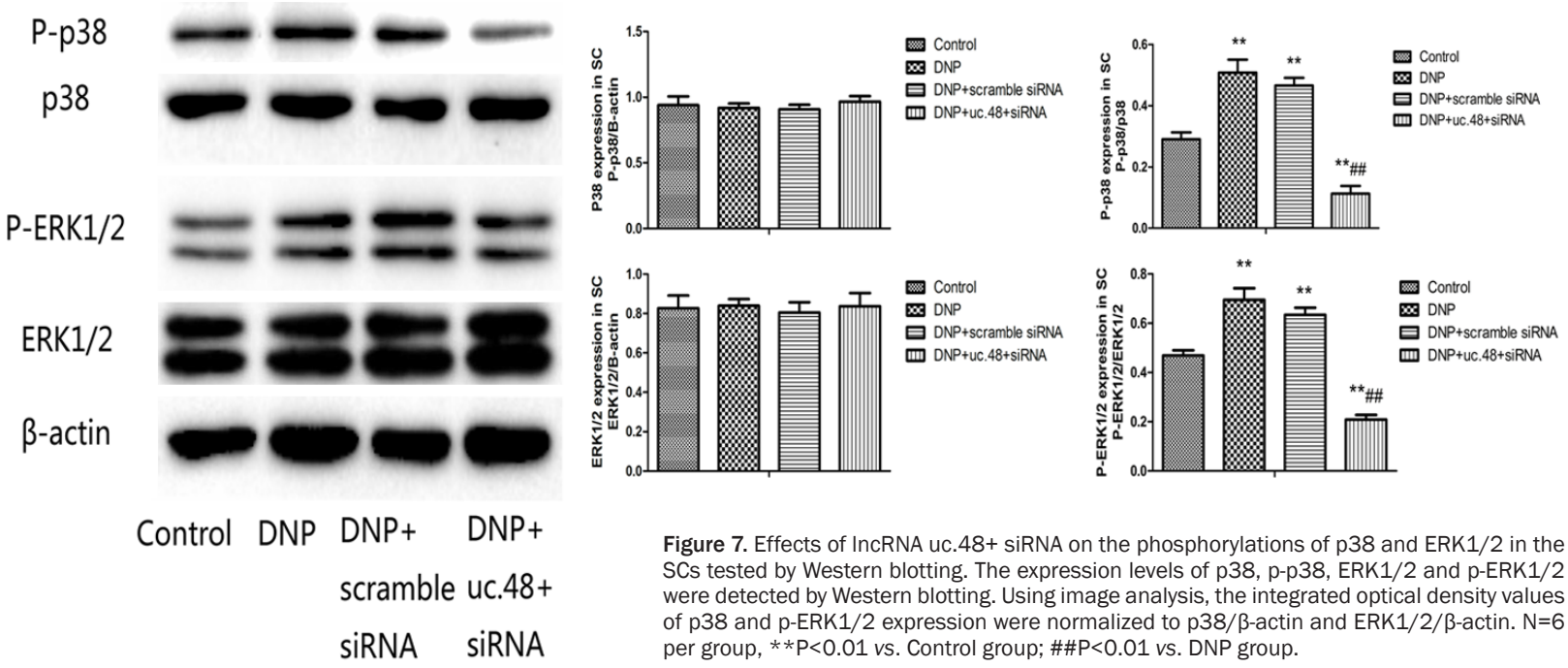

Figure 7. Effects of lncRNA uc.48+ siRNA on the phosphorylations of p38 and ERK1/2 in the SCs tested by Western blotting. The expression levels of p38, p-p38, ERK1/2 and p-ERK1/2 were detected by Western blotting. Using image analysis, the integrated optical density values of p38 and p-ERK1/2 expression were normalized to p38/β-actin and ERK1/2/β-actin. N=6 per group, **P<0.01 *vs*. Control group; ##P<0.01 *vs*. DNP group.

alleviate DNP by inhibiting the excitatory transmission mediated by the $P2X_3$ receptor in the DRG [19]. Eccentric muscle contraction and stretching can evoke mechanical hyperalgesia and modulate CGRP and P2X3 receptor expression, CGRP might cooperate with the $P2X_3$ receptor in pain transmission [25]. Whether uc.48+ siRNA treatment might affect CGRP in the SCs of DNP rats was still not clear and required further study. Thus, we observed the effects of uc.48+ siRNA treatment on the SC, and the results revealed that uc.48+ siRNA treatment might also alleviate DNP by inhibiting the expression of CGRP in the SC.

In the migraine state, CGRP release by the trigeminal nerves and the release of proinflammatory factors caused by CGRP play a key role; thus, migraine pharmacotherapies can both reduce CGRP release and inhibit endogenous inflammatory mediator (such as IL-1β and TNF-α) release [26]. Our experiments found that small interfering uc.48+ could significantly decrease the high levels of IL-1β and TNF-α in the SCs and sera of DNP rats. We guessed that uc.48+ siRNA treatment might down-regulate the expression of CGRP and then inhibit the release of proinflammatory IL-1β and TNF-α.

The increased CGRP synthesis and release might be mediated by the activation of mitogen-activated protein kinase pathways, which in turn, can be modulated by endogenous inflammatory substances [27, 28]. It has been reported that p38 inhibitors significantly attenuate the pain response in the Hargreaves hyperalgesia assay and inhibit the release of IL-1β and TNF-α [29]. Many investigations have revealed that the suppression of ERK phosphorylation can attenuate the pain transmission [30, 31] and reduce the release of IL-1β and TNF-α in the spinal dorsal horn of CCI rats [32]. In our experiments, the levels of IL-1β and TNF-α in the sera of the DNP rats increased significantly compared with the normal rats, and the phosphorylations of p38 and ERK1/2 were significantly stronger than those in the normal rats. t is possible that the high level of CGRP release and the phosphorylations of p38 and ERK1/2 in the spinal cord promoted each other and increased the levels of IL-1β and TNF-α in the serum. Uc.48+ siRNA treatment reversed the enhanced phosphorylations of p38 and ERK1/2 in the spinal cord, which might be a key mechanistic pathway for suppressing pain transmission, although the exact molecular mechanism still requires more study. Therefore, the attenuation of DNP by uc.48+ siRNA is related to the down-regulation of the phosphorylations of p38 and ERK1/2 in the spinal cord.

Conclusions

LncRNA uc.48+ may play an important role in the transmission of DNP by promoting the release of CGRP in the SC. Small interfering lncRNA uc.48+ might alleviate the hyperalgesia and allodynia of DNP rats by suppressing the release of CGRP in the SCs of DNP rats, which could inhibit the phosphorylation of p38 and ERK1/2, and suppress the release of IL-1β and TNF-α. Therefore, our results indicate that lncRNA uc.48+ siRNA may be a new and effective target for alleviating diabetic neuropathic pain.

Acknowledgements

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Disclosure of conflict of interest

None.

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