Long non-coding RNA GAS5 aggravate renal epithelial cell apoptosis in cisplatin-induced AKI by regulating miR-205-5p

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Research

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Abstract

Introduction: The present study focuses on the interaction between long non-coding RNA GAS5 and microRNA-205-5p and their roles in cisplatin-induced acute kidney injury.

Methods: Human kidney tubular cells (HK-2) were used to simulate acute renal injury induced by cisplatin with the consequent fluctuating expression levels of GAS5 and MIR-205-5p being determined respectively. Furthermore, the modulating effects of miR-205-5p and GAS5 in cisplatin-induced apoptosis of renal tubular epithelial cells and the possible binding sites between them were evaluated.

Results: The results depicted that the expression of GAS5 was significantly up-regulated after AKI induced by cisplatin, while inhibiting the increase of expression would alleviate the apoptotic-promoting effect of cisplatin on renal tubular epithelial cells. MIR-205-5p is negatively regulated by GAS5, thus down-regulation of GAS5 will consequently elevate the expression of miR-205-5p and further alleviate the damage of HK-2 cells induced by cisplatin.

Conclusions: In conclusion, in cisplatin-induced AKI, the expression of GAS5 was increased and consequently inhibited that of miR-205-5p by direct binding, which eventually aggravate the renal tubular epithelial injury, indicating their potential of being important diagnostic markers and therapeutic targets in the treatment of cisplatin-induced AKI.

Background

Acute kidney injury (AKI), being a common clinical disease with an up to 20% incidence in hospitalized patients, has a significant relevance with the deterioration of renal diseases and mortality(1). Several statistical clinical studies with fairly large sample size had delineated that the severity of AKI is evidently correlated with mortality, demonstrated by the four-week survival rate of severe AKI patients being only 50%(2). In recent years, the increasing incidence and mortality of AKI has been extensively confirmed, which has consequently aroused the vigilance and widespread concern of this none-negligible, pernicious and thorny disease (2).

Cisplatin is a metal coordination compound of platinum which has a molecular structure of planar quadrilateral (3). Although firstly synthesized in 1844, it was not until the discovery of its inhibiting cell division characteristic in 1960 that cisplatin became an indispensable constituent of antineoplastic therapies and was extensively exerted in the treatment of diverse cancers of head and neck, lung, ovary, testicle and bladder until today (3). Although cisplatin has exceptional anti-tumor properties, various parlous side effects, including nephrotoxicity, electrolyte imbalance, bone marrow toxicity and ototoxicity, were gradually confirmed by clinical practice, and has consequently become a critical constraint restricting the further development of cisplatin-based chemotherapy in the field of cancer treatment (4). In fact, adequate studies have depicted that cisplatin-induced AKI is a elusive pathophysiological process with complex mechanisms and interaction of multiple factors(5). Being one of the most common and
fatal complications among hospitalized patients treated with cisplatin, the discovery of the potent drug to alleviate this disease with elucidation of underlying mechanism have become a top priority.

In recent years, various members of non-coding RNAs were identified and depicted to correlate with diverse metabolic reactions through elaborated interaction with each other, the most common of which are long-non coding RNAs (lncRNAs) and micro RNAs (miRs) (6, 7). IncRNAs has been delineated to be key link of multiple path physiological procedures, covering various fields such as apoptosis, cell proliferation and migration (8). It was revealed that growth arrest-specific 5 (GAS5), as a member of lncRNAs, plays a role of apoptotic gene and thus have the potential of being promising non-invasive biomarker for various diseases including AKI (9). Being a member of non-coding endogenous short RNA, miRs can interact with the untranslated 3'-terminal of messenger RNA (mRNAs), hence, inhibit the expression of relevant genes (10, 11). Despite the potential regulatory mechanisms of lncRNAs and miRs on apoptosis may be intrinsically associated with cisplatin-induced AKI, the related studies are scarce. MiR-205-5p was abnormally expressed in various cancers, by directly affected cell proliferation, invasion, migration and cisplatin-resistant (12–14). Bioinformatics analysis showed a targeting relationship between miR-205-5p and GAS5. In view of the present situation, this paper, focusing on lncRNA GAS5 and miR-205-5p, through various experimental methods, intends to investigate their possible therapeutic effects in cisplatin-induced AKI, and briefly elucidate their underlying mechanisms.

**Methods**

having received bone marrow or organ transplantation, or having acquired immunodeficiency syndrome were the exclusive criteria for this study. All the research protocol of the present study was approved by the Human Ethics Committee with all the patients who participated in this study having signed the informed consent before the beginning of the experiment.

**Evaluation of GAS5 in cisplatin-induced AKI**

Serum samples were obtained by centrifuging the blood samples from the subjects at 5000g, 4°C for 3 min. After centrifuging, the serum were administrated with the miRNeasy Mini kit (QIAGEN, Germany) to get the total RNA of individual patients respectively with the correlated cDNA further synthetized exerting RT2 PreAMP cDNA Synthesis Kit (QIAGEN, Germany). Furthermore, RT2 SYBR® Green qPCR Master Mix (QIAGEN, Germany), StepOnePlus thermo-cycler (Applied Biosystems) and $2^{-\Delta\Delta C_{t}}$ in method were exerted to perform the qRT-PCR aiming to evaluation the fluctuation of GAS5’s expression levels. The association of GAS5 and Creatinine clearance rate (CCr) were also revealed by Pearson’s correlation coefficient by SPSS 19.0 (SPSS, Chicago, IL), aiming to investigate the potential pernicious effects in this pathological disease.

**Lentiviral vector construction and infection**

The LV10-CMV-RFP-Puro vector (GenePharma, Shanghai, China) was administrated with si-GAS, a short-hairpin RNA directed against GAS5, while the negative sequence was exerted for negative controlling.
After receiving the lentiviral expression vectors, the 293T cells were further subjected to virus particles incubated by Polybrene (Sigma, St. Louis, Missouri, USA) and an exclusion procedure of puromycin. Besides, the GAS5 cDNA was multiplied with PfuUltra II Fusion H DNA Polymerase (Stratagene, Agilent Technologies, Santa Clara, CA, USA), followed by the subcloning to EcoRI and HindIII of pcDNA3.1 vector (Invitrogen, USA) to construct pcDNA-GAS5. The mimics and inhibitors of miR-205-5p and NC mentioned were provided by GenePharma (Shanghai, China), while Lipofectamine 2000 (Life Technologies Corporation, Carlsbad, CA) were exerted to transfect the cells. The sequences for GAS5 siRNA were 5′-GCGAGGCACATGTAAGCAA-3′ Non targeting sequence (5′-ACGUGACACGUUCGGAGAATT-3′) was used as a negative control.

miR-205-5p mimic, 5′-UCCUCAUCCACCGAGUCUG-3′; miR-NC mimic, 5′-UCGUUGUGCAGGGCGGGAA-3′; miR-205-5p inhibitor, 5′-CAGACUCCGGGAAUGAAGGA-3′; miR-NC inhibitor, 5′-CAGUACUUUGUGUAGUAACAA-3′

**qRT-PCR**

After the total RNA was isolated from the cells, the cDNA was obtained exerting High-Capacity cDNA Reverse Transcription Kit (Applied Biosystems, CA, USA) with that of miRNAs being fabricated by TaqMan MicroRNA Reverse Transcription kit (Applied Biosystems, Foster City, CA, USA). TaqMan Universal Master Mix II, TaqMan miRNA assays for miR-205-5p and U6, and TaqMan gene expression assays for GAS5 and GAPDH were further exerted for qRT-PCR. Expressions were calculated utilizing $2^{\triangle\triangle Ct}$ method. The primers utilized are:

GAS5 F: 5′-GGCAAATGAGCACTAAAG -3′ R: 5′-CACCCACTCCTCTATCTACA -3′

GAPDH F: TCAACGACCCCTTCATTGACC R: CTTCGCCGTTGATGACAAGCTTC

miR-205-5p forward, 5′-TCTCTTATCCACCGAGGTCT-3′ and reverse, 5′-GCGAGCAGAGATTAATACGAC-3′; U6 forward, 5′-ATTGGAACGATACAGAGATT-3′ and reverse, 5′-GGAACGTTCACGAATTTG-3′

**Western blot**

Before experiment, the HK-2 cells was plated in a 6-well plate at a rate of 2 × 106 cells/well and incubated for 24 h. Cells were lysed and 50 μg protein was isolated utilizing SDS-PAGE. The polyvinylidene fluoride membrane were exerted to receive the protein sample underwent electrophoresis and then administrated with the blockage of non-fat milk. The antibodies of GAPDH (ab9485, 1:5000, Abcam) and cleaved Caspase-3 (ab49822, 1:1500, Abcam) were further exerted to administrate the foregoing membranes, followed by incubation of Goat Anti-Rabbit IgG H&L (HRP, ab205718, 1:3000, Abcam) or Goat Anti-Mouse IgG H&L (HRP, ab6789, 1:3000, Abcam), with the relevant protein levels eventually revealed by enhanced chemiluminescence reagents (Pierce, Rockford, IL, USA).

**Luciferase reporter analysis**
The GAS5-wt (embodying the specific sequence able to bind with miR-205-5p) and GAS5-mut (embodying the mutation of foregoing sequence) were obtained from Genescript (Nanjing, China). The luciferase analysis was performed by the dual-luciferase reporter analysis system (Promega, USA) after the HEK 293T cells’ being co-transfected with miR-205-5p or miR-NC and GAS5-wt or GAS5-mut. According to the manufacturer’s protocol, a dual-luciferase reporter assay system (Promega, USA) was used to detect the luciferase activity after co-transfection in HEK 293T cells for 48 h.

**Pull-down analysis**

The HK2 cells underwent a transfection of biotinylated probe (Sangon, Shanghai, China) targeting GAS5 and miR-205-5p, and 48h later, the HK2 cells were lysed, the products of which received the incubation of Dynabeads M-280 Streptavidin (Invitrogen, Carlsbad, CA, USA). The qRT-PCR was exerted for analyzing the bound RNAs.

**RNA Immunoprecipitation (RIP) analysis**

The Magna RIP RNA-Binding Protein IP Kit (Millipore, Bedford, MA, USA) and Ago2 antibody (2897, Cell Signaling, Danvers, MA, USA) were exerted for RIP analysis, by which the expression levels of GAS5 and miR-205-5p were consequently revealed.

**HK-2 model culture**

Antibiotics (100 U/ml penicillin G + 100 μg/ml streptomycin + 0.25 μg/ml amphotericin B, Invitrogen) and Dulbecco's Modified Eagle Media/F12 with 10% fetal bovine serum (Invitrogen, Carlsbad, CA) at 37°C in a 100% humidified atmosphere of 5% CO₂-95% air were exerted for culturing the HK-2 cells (American Type Culture Collection, Manassas, VA). The foregoing cells’ conservation of original phenotypic and metabolic characteristics of the proximal tubule during the culturing procedure were confirmed.

**Apoptosis analysis**

According to the manufacturer's, the cells were transfected for 48h and further stained for 15min in the dark at room temperature with Annexin V and PI (Annexin V-FITC Apoptosis Detection Kit, eBioscience), the HK-2 cells’ apoptosis status were evaluated by BD FACSCalibur flow cytometer (BD Biosciences, California, USA) and Cell Quest software (BD Biosciences). After all the cells being classified as alive, early-apoptotic, late-apoptotic and dead, the apoptosis status was consequently revealed by the early-apoptotic ratio. All the above analysis was repeated triplicately.

**Statistical Analysis**

The data were administrated with one-way ANOVA and Dunnett's multiple comparison tests or paired *t* test (version 5.00; Graph Pad Prism Software Inc., San Diego, CA). *P*<0.05 was selected for representing statistical significance.
Results

The expression of GAS5 and miR-205-5p in the cisplatin-induced AKI patients and in HK-2 cells

Comparing to their corresponding, GAS5 was upregulated while miR-205-5p was reduced in AKI group (Fig. 1A, 1B, P < 0.01), indicating that GAS5 and miR-205-5p may be of importance in cisplatin-induced AKI. Furthermore, the expression levels increase of GAS5 was to revealed to correlating with the dose of Cisplatin. And the expression of miR-205-5p was decreased as well as in dose dependent manner. (Fig. 1C and 1D)

The apoptosis-promoting effects were significantly inhibited by suppressing GAS5 in Cisplatin-stimulated HK-2 cells.

In order to explore the role of GAS5 in cisplatin-induced AKI, we induced in the cell model using Cisplatin. The HK-2 cells with consistently suppressed expression levels of GAS5 were obtained by being transfected with si-GAS5 or pc-GAS5 (Fig. 2A), after which the inhibitory effects the two mentioned above and the apoptosis-promoting effects of GAS5 were further evaluated. As shown in Fig. 2, the rate of cleaved-casepase5/Gapdh(Fig. 2B), the activity of caspase-3(Fig. 2C), and the apoptosis rate (Fig. 2D and 2E) show that cisplatin induced apoptosis (control group). However, the regulation of GAS5 reduced this apoptosis induced by Cisplatin, on the contrary, the downregulation of GAS5 aggravates this effect. It was revealed that the Cisplatin-stimulated apoptosis was significantly suppressed after the decrease of GAS5 (P < 0.05).

The apoptosis-promoting effects were significantly amplified by suppressing miR-205-5p in Cisplatin-stimulated HK-2 cells.

The HK-2 with diverse expression levels of miR-205-5p were obtained by being transfected with miR-205-5p mimic and miR-205-5p inhibitor, after which the inhibitory effects the two mentioned above and the apoptosis-inhibiting effects of miR-205-5p were further evaluated (Fig. 3A). In Cisplatin-stimulated HK-2 cells model group, It shows that the up-regulation of miR-205-5p inhibits cell apoptosis, while the down-regulation of miR-205-5p promotes cell apoptosis in western blot(Fig. 3A and Fig. 3B) and flow cytometry(Fig. 3D and 3E).

Reciprocal inhibition existed between miR-205-5p and GAS5 in HK-2 cells.

Upregulation of GAS5 reduced miR-205-5p expression and downregulation of GAS5 induced miR-205-5p expression in both control group(−) and cisplatin-stimulated group(+) (Fig. 4A). Targetscan, Starbase, and microRNA.org were exerted to reveal that GAS5 can directly bind with miR-205-5p and consequently serves as the predictive target of it (Fig. 4B). Besides, the luciferase activity was suppressed by miR-205-5p with the foregoing trend being significantly reversed by Mut(Fig. 4C). Furthermore, it was confirmed that miR-205-5p can significantly pull down while the mut of that can not (Fig. 4D). Similar trends was
also revealed by GAS5 probe (Fig. 4E), collectively suggesting the direct interacting relationship between miR-205-5p and GAS5.

**Up-Regulation of miR-205-5p mostly reversed GAS5-induced apoptosis in Cisplatin-stimulated HK-2 cells**

**Cisplatin-stimulated** HK-2 cells with consistent high expression levels of GAS5 were transfected by miR-205-5p mimic, revealing that the apoptosis-promoting effects of GAS5 can be significantly inhibited by miR-205-5p (Fig. 5A, 5B).

**Discussion**

Being members of none-coding RNAs, GAS5 and miR-205-5p were repeatedly delineated to be evidently involved in the regulation of apoptotic mechanisms in various pathophysiological procedures (15–18). Functional experiments have further revealed that GAS5 is able to perform two-pronged functions of inhibiting the proliferation and promoting apoptosis of various cell types (19). These cellular mechanisms of GAS5 are not only significantly involved in drug resistance mechanism of specific cell type like tumor, but also play an critical role in normal cells’ damage tolerance under many pathophysiological procedures. miR-205-5p was proved to be one of the downstream factors of GAS5’s cell proliferation modulation (20). It was revealed to be of importance in kidney development, homeostasis and pathology, and participate in various histopathological changes such as the occurrence and deterioration of tubulointerstitial sclerosis and terminal stage glomerulopathy in diverse types of kidney disease (21). Interestingly, recent studies have depicted the crossroad-like role of miR-205-5p as an apoptosis-suppressing gene or as an anti-survival gene under different physiological conditions, leaving the potential correlation and underlying mechanism between GAS and cisplatin-induced AKI rather complicated and thus need to be delineated urgently (22). So far, however, to the best of our knowledge, the related research is still rare. In the present study, it is confirmed that GAS5 was significantly increased in AKI group compared with their corresponding, with miR-205-5p being evidently reduced in the mean time. Besides, these changes in expression were further proved to be dose-dependent with cisplatin, jointly making the involvement of GAS5 and miR-205-5p in this cisplatin-induced AKI preliminarily confirmed.

The interaction between lncRNAs and microRNAs was delineated to elaborately modulate the elusive balance of survival and programmed death (23). In the present study, through the evaluation of cell apoptosis in the Cisplatin-stimulated HK-2 cells, it was revealed that the down-regulation of GAS5 significantly correlate with the alleviation of cell apoptosis, while that of miR-205-5p conversely delivered a apoptosis-promoting effect, which conforms to the revealed role of miR-205-5p to elevate the resistance of oxidative and endoplasmic reticulum stresses and consequently be in favor of cell survival recent years (24, 25). Furthermore, the administration with miR-205-5p mimic also suppressed the severe apoptotic procedures in the present study. The above-mentioned conforms to the general regulation pattern of GAS5 and miR-205-5p on apoptosis in the pathophysiological procedures of various cell types, and also indicates the intrinsic causal relationship between GAS5, miR-205-5p and cisplatin-induced AKI
A antagonistic relationship between GAS5 and miR-205-5p in this pathological procedure was also implied.

To further elucidate the contradictory modulating effects between GAS5 and miR-205-5p in cisplatin-induced AKI, the fluctuation of their expression levels after interaction were evaluated. As presented in Fig. 4A, a reciprocal inhibitory relationship of miR-205-5p and GAS5 in HK-2 cells was suggested by the result that upregulation of GAS5 can significantly down-regulate the expression levels of miR-205-5p, and vice versa, further confirming the antagonistic relationship between them.

It was suggested that miR-205-5p can directly interact to GAS5 via specific binding site (27). In the present study, this potential interacting site structure of GAS5 and miR-205-5p was further evaluated by bioinformatics analysis. As shown in Fig. 4B, it was revealed to be a functional binding site in transcription products GAS5, elucidating the interaction between them at the molecular structure level, which conforms to the reported characteristics of these two members of none-coding RNAs to directly bind with each other (28, 29). The relative luciferase activity was decreased by up-regulated miR-205-5p expression, with this inhibition further reversed with the administration of GAS5-mut, conforming to the demonstration of binding site and antagonistic expression results mentioned above. Moreover, in this study, bi-directional pull-down assay was further exerted to confirm their putative binding structure. It was revealed that GAS5 was significantly pulled down by biotinylated miR-205-5p, while the foregoing trend was further reversed by miR-205-5p-mut, collectively suggesting their reciprocal inhibitory interaction synthetically. Finally, a significant reversed of GAS5’s cell apoptosis-promoting effects by administration of miR-205-5p directly confirmed that miR-205-5p participates in GAS5’s modulation mechanism with an antagonistic role. In conclusion, the present study revealed that IncRNA GAS5 can aggravate renal epithelial cell apoptosis in cisplatin-induced AKI by antagonistically interacting with miR-205-5p, and thus have the potential of being promising therapeutic target of hindering the development of this parlous disease.

The present study have some limitations: Initially, it was proved that CMTM4, ZEB2 and PTEN can perform as downstream regulators of miR-205-5p, and thus have the potential to be of importance in the GAS5’s apoptosis modulation mechanism of cisplatin-induced AKI (10, 21, 26, 30–33). Suggesting that they can be exerted as complementary indicators in follow-up studies to render the mechanism revealed more comprehensive. Secondly, the presented study was mainly performed in vitro, leaving the complex interaction between specific factors in vivo environment being not properly simulated, making the comparative experiment with in vivo evaluation of the mechanism underlying cisplatin-induced AKI further warranted. Finally, as a member of microRNAs like miR-205-5p, miR-34 was also revealed to interact with GAS5 in regulation of many pathological procedures, thus may potentially participate in the elusive mechanism underlying this disease (6). Its role remains to be explored in our subsequent research to illuminate the holistic mechanism of the apoptosis modulating effects of miR-205-5p and GAS5.

Conclusion
The expression of GAS5 in cisplatin-induced AKI was increased and consequently inhibited that of miR-205-5p by direct binding, which eventually aggravate the renal tubular epithelial injury. We indicated the potential of GAS5 being important diagnostic markers and therapeutic targets in the treatment of cisplatin-induced AKI.

**Abbreviations**

qRT-PCR: quantitative real-time PCR, cDNA: complementary DNA,

**Declarations**

**Acknowledgement**

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**Competing Interests**

The authors declare that there are no competing interests associated with the manuscript.

**Ethical approval and Consent to participate**

All procedures performed in studies involving human participants were in accordance with the Ethical Standards of the Institutional Research Committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Written consent was obtained from study participants. All the research protocol of the present study was approved by the Human Ethics Committee of The First Affiliated Hospital of Xiamen University with all the patients who participated in this study having signed the informed consent before the beginning of the experiment.

**Consent for publication**

Not applicable

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**Availability of data and materials**

The analyzed data sets generated during the study are available from the corresponding author on reasonable request.

**Authors contribution**

Yinan L designed the study, analyzed the data, performed the experiments, prepared the manuscript...
Min Zhang, Hui Bi, Shaoyan Wang, Xuejuan Sun, analyzed the data, performed the experiments, prepared the manuscript

All authors have read and approved the manuscript.

References

1. Bagshaw SM, George C, Bellomo R. A comparison of the RIFLE and AKIN criteria for acute kidney injury in critically ill patients. Nephrology, dialysis, transplantation: official publication of the European Dialysis and Transplant Association - European Renal Association. 2008;23(5):1569-74.

2. Patel SS, Palant CE, Mahajan V, Chawla LS. Sequelae of AKI. Best practice & research Clinical anaesthesiology. 2017;31(3):415-25.

3. Dasari S, Tchounwou PB. Cisplatin in cancer therapy: molecular mechanisms of action. European journal of pharmacology. 2014;740:364-78.

4. Li RY, Zhang WZ, Yan XT, Hou JG, Wang Z, Ding CB, et al. Arginyl-fructosyl-glucose, a Major Maillard Reaction Product of Red Ginseng, Attenuates Cisplatin-Induced Acute Kidney Injury by Regulating Nuclear Factor kappaB and Phosphatidylinositol 3-Kinase/Protein Kinase B Signaling Pathways. Journal of agricultural and food chemistry. 2019.

5. Lee SA, Cozzi M, Bush EL, Rabb H. Distant Organ Dysfunction in Acute Kidney Injury: A Review. American journal of kidney diseases: the official journal of the National Kidney Foundation. 2018;72(6):846-56.

6. Toraih EA, Alghamdi SA, El-Wazir A, Hosny MM, Hussein MH, Khashana MS, et al. Dual biomarkers long non-coding RNA GAS5 and microRNA-34a co-expression signature in common solid tumors. PloS one. 2018;13(10):e0198231.

7. Wang Y, Xie Y, Li L, He Y, Zheng D, Yu P, et al. EZH2 RIP-seq Identifies Tissue-specific Long Non-coding RNAs. Current gene therapy. 2018;18(5):275-85.

8. Geng X, Xu X, Fang Y, Zhao S, Hu J, Xu J, et al. Effect of long non-coding RNA growth arrest-specific 5 on apoptosis in renal ischaemia/reperfusion injury. Nephrology (Carlton, Vic). 2019;24(4):405-13.

9. Wu GC, Li J, Leng RX, Li XP, Li XM, Wang DG, et al. Identification of long non-coding RNAs GAS5, linc0597 and Inc-DC in plasma as novel biomarkers for systemic lupus erythematosus. Oncotarget. 2017;8(14):23650-63.

10. Zhang F, Liu J, Xie BB. Downregulation of microRNA-205 inhibits cell invasion and angiogenesis of cervical cancer through TSLC1-mediated Akt signaling pathway. Journal of cellular physiology. 2019.

11. Lee SH, Ju HM, Choi JS, Ahn Y, Lee S, Seo YJ. Circulating Serum miRNA-205 as a Diagnostic Biomarker for Ototoxicity in Mice Treated with Aminoglycoside Antibiotics. International journal of molecular sciences. 2018;19(9).

12. Li X, Li Y, Han Y, Dong B, Liu D, Che L, et al. miR-205 Promotes Apoptosis of Cervical Cancer Cells and Enhances Drug Sensitivity of Cisplatin by Inhibiting YAP1. Cancer biotherapy & radiopharmaceuticals. 2020;35(5):338-44.
13. Zhang P, Lu X, Shi Z, Li X, Zhang Y, Zhao S, et al. miR-205-5p regulates epithelial-mesenchymal transition by targeting PTEN via PI3K/AKT signaling pathway in cisplatin-resistant nasopharyngeal carcinoma cells. Gene. 2019;710:103-13.

14. Shi X, Xiao L, Mao X, He J, Ding Y, Huang J, et al. miR-205-5p Mediated Downregulation of PTEN Contributes to Cisplatin Resistance in C13K Human Ovarian Cancer Cells. Frontiers in genetics. 2018;9:555.

15. Ke K, Sun Z, Wang Z. Downregulation of long non-coding RNA GAS5 promotes cell proliferation, migration and invasion in esophageal squamous cell carcinoma. Oncology letters. 2018;16(2):1801-8.

16. Qiao HP, Gao WS, Huo JX, Yang ZS. Long non-coding RNA GAS5 functions as a tumor suppressor in renal cell carcinoma. Asian Pacific journal of cancer prevention : APJCP. 2013;14(2):1077-82.

17. Mourtada-Maarabouni M, Pickard MR, Hedge VL, Farzaneh F, Williams GT. GAS5, a non-protein-coding RNA, controls apoptosis and is downregulated in breast cancer. Oncogene. 2009;28(2):195-208.

18. Liu L, Pang X, Shang W, Xie H, Feng Y, Feng G. Long non-coding RNA GAS5 sensitizes renal cell carcinoma to sorafenib via miR-21/SOX5 pathway. Cell cycle (Georgetown, Tex). 2019;18(3):257-63.

19. Pickard MR, Williams GT. Molecular and Cellular Mechanisms of Action of Tumour Suppressor GAS5 LncRNA. Genes. 2015;6(3):484-99.

20. Dong L, Li G, Li Y, Zhu Z. Upregulation of Long Noncoding RNA GAS5 Inhibits Lung Cancer Cell Proliferation and Metastasis via miR-205/PTEN Axis. Medical science monitor : international medical journal of experimental and clinical research. 2019;25:2311-9.

21. Zhang H, Zhang X, Yuan X, Wang L, Xiao Y. MicroRNA-205 inhibits renal cells apoptosis via targeting CMTM4. Iranian journal of basic medical sciences. 2015;18(10):1020-6.

22. Qin AY, Zhang XW, Liu L, Yu JP, Li H, Wang SZ, et al. MiR-205 in cancer: an angel or a devil? European journal of cell biology. 2013;92(2):54-60.

23. Chen Y, Qiu J, Chen B, Lin Y, Chen Y, Xie G, et al. Long non-coding RNA NEAT1 plays an important role in sepsis-induced acute kidney injury by targeting miR-204 and modulating the NF-kappaB pathway. International immunopharmacology. 2018;59:252-60.

24. Lin Y, Ding Y, Song S, Li M, Wang T, Guo F. Expression patterns and prognostic value of miR-210, miR-494, and miR-205 in middle-aged and old patients with sepsis-induced acute kidney injury. Bosnian journal of basic medical sciences. 2019.

25. Muratsu-Ikeda S, Nangaku M, Ikeda Y, Tanaka T, Wada T, Inagi R. Downregulation of miR-205 modulates cell susceptibility to oxidative and endoplasmic reticulum stresses in renal tubular cells. PloS one. 2012;7(7):e41462.

26. Chen Z, Tang ZY, He Y, Liu LF, Li DJ, Chen X. miRNA-205 is a candidate tumor suppressor that targets ZEB2 in renal cell carcinoma. Oncology research and treatment. 2014;37(11):658-64.

27. Yang W, Hong L, Xu X, Wang Q, Huang J, Jiang L. LncRNA GAS5 suppresses the tumorigenesis of cervical cancer by downregulating miR-196a and miR-205. Tumour biology : the journal of the
28. Ballantyne MD, McDonald RA, Baker AH. IncRNA/MicroRNA interactions in the vasculature. Clinical pharmacology and therapeutics. 2016;99(5):494-501.

29. Furio-Tari P, Tarazona S, Gabaldon T, Enright AJ, Conesa A. spongeScan: A web for detecting microRNA binding elements in IncRNA sequences. Nucleic acids research. 2016;44(W1):W176-80.

30. Wilhide ME, Feller JD, Li B, Mohamed AZ, Becknell B, Jackson AR, et al. Renal epithelial miR-205 expression correlates with disease severity in a mouse model of congenital obstructive nephropathy. Pediatric research. 2016;80(4):602-9.

31. Saal S, Harvey SJ. MicroRNAs and the kidney: coming of age. Current opinion in nephrology and hypertension. 2009;18(4):317-23.

32. Li J, Hu K, Gong G, Zhu D, Wang Y, Liu H, et al. Upregulation of MiR-205 transcriptionally suppresses SMAD4 and PTEN and contributes to human ovarian cancer progression. Scientific reports. 2017;7:41330.

33. Tao H, Zhang JG, Qin RH, Dai C, Shi P, Yang JJ, et al. LncRNA GAS5 controls cardiac fibroblast activation and fibrosis by targeting miR-21 via PTEN/MMP-2 signaling pathway. Toxicology. 2017;386:11-8.