

## MiR-143 and rat embryo implantation



Shi Tian<sup>a,1</sup>, Xing Su<sup>b,c,1</sup>, Lu Qi<sup>b,1</sup>, Xiao-Hua Jin<sup>b,c</sup>, Yi Hu<sup>b</sup>, Chun-Ling Wang<sup>d</sup>, Xu Ma<sup>b,c,\*</sup>, Hong-Fei Xia<sup>b,c,\*</sup>

<sup>a</sup> Haidian Maternal & Child Health Hospital, Beijing 100080, China

<sup>b</sup> Reproductive and Genetic Center of National Research Institute for Family Planning, Beijing 100081, China

<sup>c</sup> Graduate School, Peking Union Medical College, Beijing 100730, China

<sup>d</sup> Cadre Ward, China Mei-Tan General Hospital, Beijing 100028, China

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### ABSTRACT

**Background:** To study the role of miR-143 during embryo implantation in rat.

**Methods:** MiR-143 expression in rat early pregnancy was detected by Northern blot. The relation between miR-143 and *Lifr* predicted and confirmed by bioinformatics method, dual-luciferase activity assay, Western blot and immunohistochemistry. The role of miR-143 was detected by MTS, Edu and *ranswell* chamber assays.

**Results:** The expression level of miR-143 on gestation day 5–8 (g.d. 5–8) was higher than on g.d. 3–4 in uteri of pregnant rat. MiR-143 was mainly localized in the superficial stroma/primary decidual zone, luminal and glandular epithelia. The expression of miR-143 was not significantly influenced by pseudopregnancy, but the activation of delayed implantation and experimentally induced decidualization significantly promoted miR-143 expression. Over-expression of miR-143 in human endometrial stromal cells (ESCs) inhibited cell proliferation, migration and invasion. Knockdown of miR-143 promoted cell proliferation and invasion. The results of recombinant luciferase reporters showed that miR-143 could bind to the 3′-untranslated region (UTR) of *leukemia inhibitory factor receptor (Lifr)* to inhibit *Lifr* translation.

**Conclusions:** Uterine miR-143 may be involved in the successful pregnancy, especially during the process of blastocyst implantation through regulating *Lifr*.

**General significance:** This study may have the potential to provide new insights into the understanding of miR-143 function during embryo implantation.

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### 1. Introduction

Mir-143 is a short non-coding RNAs molecule and located on chromosome 5 position 33 in the human genome [38]. MiR-143 is highly conserved in vertebrates and function to regulate the expression levels of their target genes by binding to their 3′-untranslated regions (3′-UTR) [11,12,38]. MiR-143 also down-regulates the expression levels of their target genes through targeting their coding regions [43].

MiR-143 is thought to play an important role in tumorigenesis. Decreased expression of *mir-143* has been observed in diversified cancer samples, such as bladder cancer, cervical cancer, colorectal cancer, liposarcomas, prostate carcinomas, non-small cell lung cancer, breast cancer, endometrioid carcinomas, renal cell carcinoma and so on [1,9,13,18,19,30,44–47]. Loss of *miR-143/145* cluster enhanced cancer cell migration and invasion in prostate cancer through directly regulating Golgi membrane protein 1 (GOLM1) [22]. The chemically modified miR-143 duplex showed a significant tumor-suppressive effect on

xenografted tumors of human colorectal cancer DLD-1 cells and may be a candidate for an RNA medicine for the treatment of colorectal tumors [21]. There are striking similarities present between the behavior of invasive placental cells during embryo implantation and that of invasive cancer cells [29]. Implantation of the embryo is one of the last great mysteries of reproductive biology. MiR-143 was found to be upregulated at implantation sites in mouse uterus on day 5 of pregnancy compared with inter-implantation sites [15]. Our previous study also showed that miR-143 was differentially expressed in rat uteri between pre-receptive and receptive phase via microRNA (miRNA) microarray analysis [40]. However, the roles of miR-143 during embryo implantation remain unclear.

In the present study, we measure the expression and regulation of miR-143 in uterus during peri-implantation in rat. Additionally, we analyze the effects of miR-143 on cell viability, migration and invasion, and investigate the regulatory target of miR-143.

### 2. Materials and methods

#### 2.1. Experimental animals and protocols

Sexually mature female Sprague Dawley rats (220–260 g body weight) were purchased from the Laboratory Animal Center of the

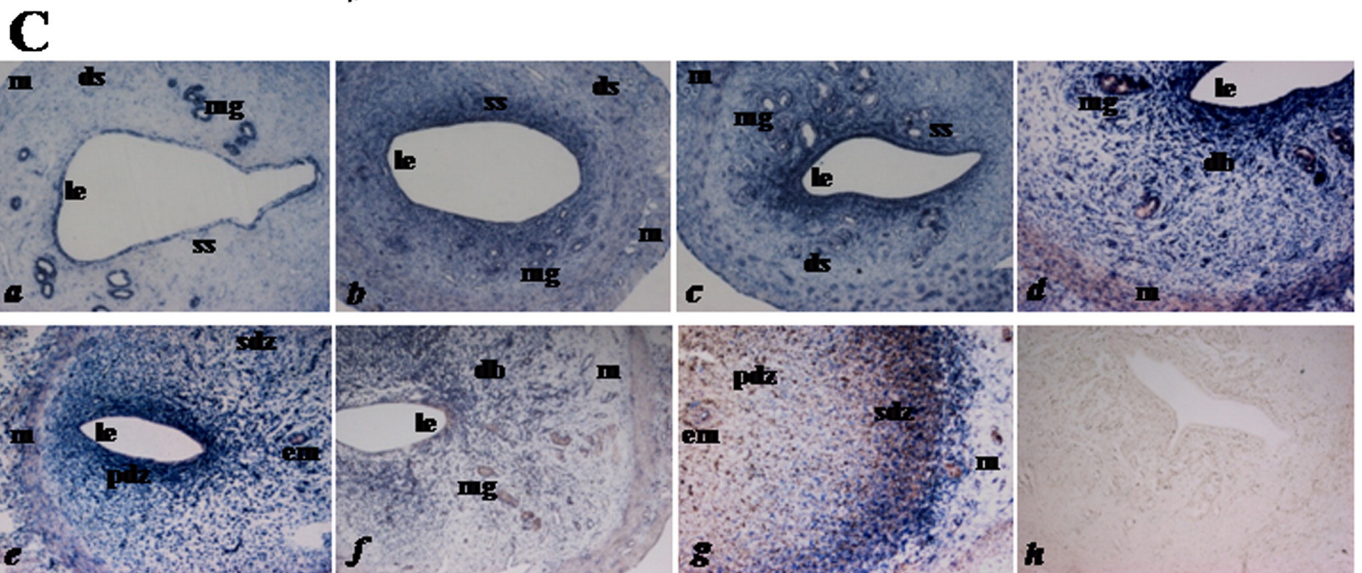
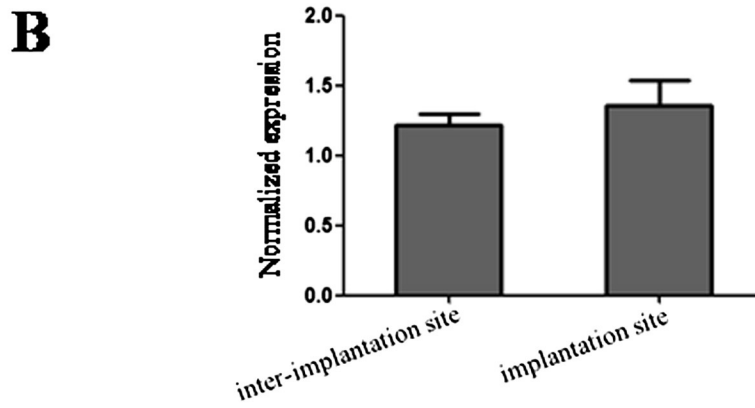
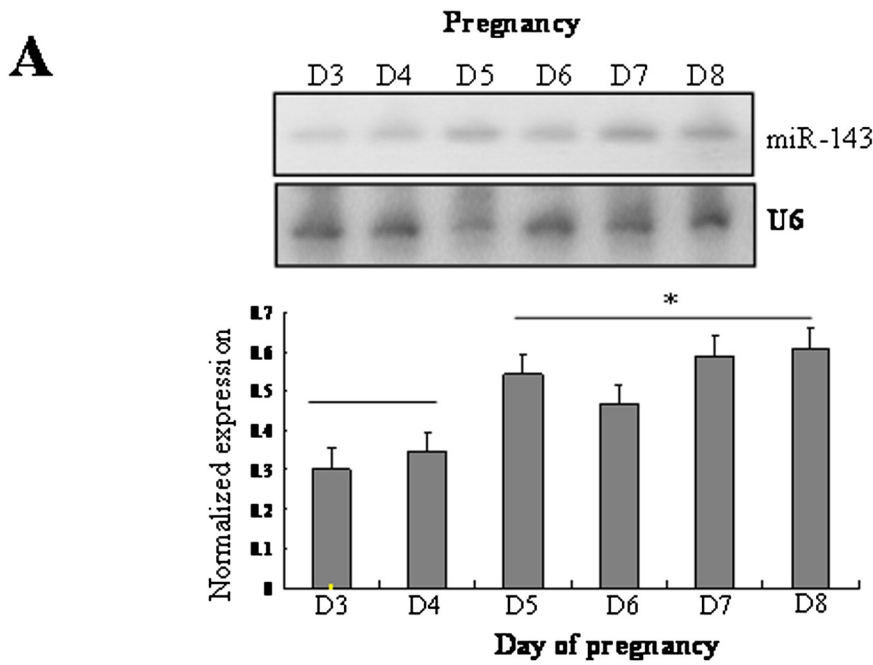
\* Corresponding authors at: Genetic Center of National Research Institute for Family Planning, Beijing 100081, China. Tel.: +86 1 62178932; fax: +86 1 62179059.

E-mail addresses: [chinawang2007@163.com](mailto:chinawang2007@163.com) (C.-L. Wang), [genetic@263.net.cn](mailto:genetic@263.net.cn) (X. Ma), [hongfeixia@126.com](mailto:hongfeixia@126.com) (H.-F. Xia).

<sup>1</sup> These two authors contributed equally to this work.

Academy of Military Medical Sciences (Beijing, China). Rats were housed in a temperature- and humidity-controlled room with a 12/12 h light/dark cycle. All animal procedures were approved by the Institutional

Animals Care and Use Committee of the National Research Institute for Family Planning. Rats were caged overnight with fertile males of the same strain. The presence of a vaginal plug or sperm was considered



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