

Supplementary Information For

HypoxamiR-210 accelerates wound healing in diabetic mice by improving cellular metabolism

Sampath Narayanan^{1,2}, Sofie Eliasson Angelstig^{1,2}, Cheng Xu^{1,2}, Jacob Grünler^{1,2}, Allan Zhao¹, Wan Zhu³, Ning Xu Landén⁴, Mona Ståhle⁴, Jingping Zhang⁵, Mircea Ivan⁶, Raluca Georgiana Maltesen⁷, Ileana Ruxandra Botusan^{1,2}, Neda Rajamand Ekberg^{1,2}, Xiaowei Zheng^{1,2,*†}, Sergiu-Bogdan Catrina^{1,2,8,*†}

¹Department of Molecular Medicine and Surgery, Karolinska Institutet, Stockholm, Sweden.

²Department of Endocrinology and Diabetes, Karolinska University Hospital, Stockholm, Sweden.

³Department of Nosocomial Infection Control, China Medical University, Shenyang, China.

⁴Unit of Dermatology and Venereology, Department of Medicine, Karolinska Institutet, Stockholm, Sweden.

⁵Department of Infectious Disease, China Medical University, Shenyang, China.

⁶Departments of Medicine, Microbiology and Immunology, Indiana University, Indianapolis, USA

⁷Department of Anesthesia and Intensive Care Medicine, Aalborg University Hospital, Denmark

⁸Center for Diabetes, Academic Specialist Centrum, Stockholm, Sweden

*To whom correspondence should be addressed:

E-mail: Xiaowei.zheng@ki.se; Sergiu-Bogdan.Catrina@ki.se.

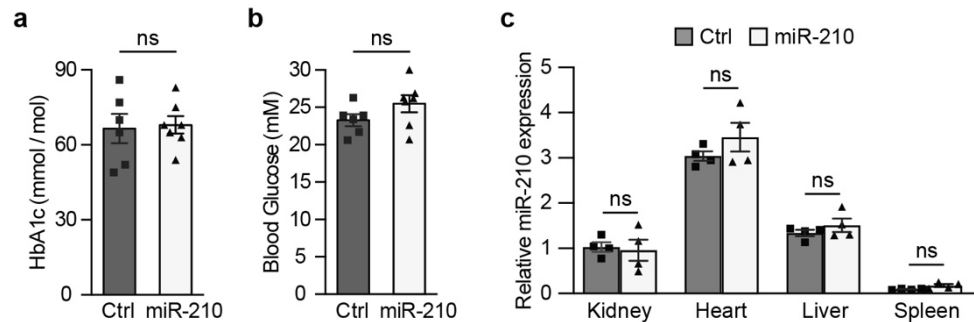
† These authors contributed equally to this work.

This PDF file includes:

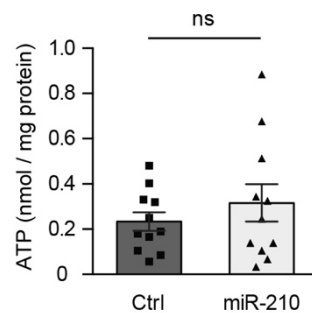
Supplementary figure and figure legends: Supplementary Figure 1 – 3.

Supplementary methods

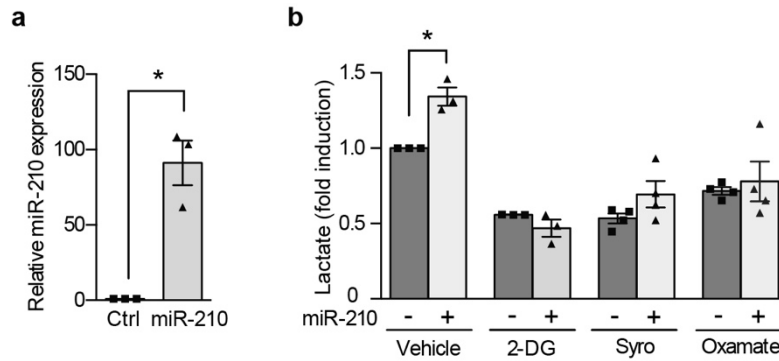
Supplementary figure and figure legends



Supplementary Figure 1: HbA1c (**a**) and blood glucose (**b**) were measured from db/db mice for control mimic (Ctrl, n=6) and miR-210 mimic (miR-210, n=7) treatment before wounding. (**c**) Eight days after wounding, kidney, heart, liver and spleen tissues were collected and miR-210 levels were measured (n=4). Statistical differences were calculated using Student's t-test. Data is represented as mean $\pm$ s.e.m. No significant difference was found between groups (ns).



Supplementary figure 2: ATP was measured from wounds of db/db mice injected with control mimic (Ctrl) or miR-210 mimic (miR-210) (n=11) and was normalized to protein concentration. Data are represented as mean $\pm$ s.e.m. Statistical difference was calculated using Student's t-test. No significant difference was found between groups (ns).



Supplementary figure 3: (a) miR-210 expression was measured by quantitative RT-PCR in HDF cells following the transfection of control mimic (Ctrl) or miR-210 mimic (miR-210) (n=3). **(b)** Lactate was measured in HDF cells transfected with control mimic or miR-210 mimic and was treated with high glucose concentrations (30 mM) in the presence of 2-deoxyglucose (15 mM, n=3), Syrosingopine (10 μ M, n=4), Oxamate (45 mM, n=4), or vehicle (n=3) in hypoxia. Statistical differences were calculated by paired Student's t-test. Data is represented as mean $\pm$ s.e.m. *, $P < 0.05$.

Supplementary methods

Measurement of ATP levels in wound tissue

ATP levels in the wounds were measured using ATP detection assay kit (Cayman Chemical) according to the manufacturer's instructions. Briefly, the tissues were lysed in ATP detection sample buffer and the lysate was added to a reaction mix containing ATP detection assay buffer, D-luciferin and luciferase. Following 20 minutes incubation protected from light, luminescence was recorded in Glomax Luminometer (Promega). The concentration of ATP in the lysates were determined by plotting the absorbance values against serially diluted ATP detection standards. The final ATP levels were finally normalized to protein concentrations.