Identification of microRNAs as potential novel regulators of *HSD11B1* expression

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11β-Hydroxysteroid dehydrogenase type 1 (11β-HSD1, gene name *HSD11B1*) is a ubiquitously expressed enzyme that converts glucocorticoid receptor-inert cortisone to receptor-active cortisol. *HSD11B1* expression is regulated in a highly tissue-specific manner by immunomodulatory and metabolic regulators. Multiple evidences support a causal role for 11β-HSD1 in the current obesity epidemic. In obese people, *HSD11B1* expression is increased in adipose tissue, but typically decreased in liver, and the underlying tissue-specific mechanisms are largely unknown.

In this context, a potential role of microRNAs (miRNAs) was investigated. Four different miRNA target prediction tools were used to choose possible candidates and a publicly available miRNA expression atlas to further select candidates expressed in hepatocytes. Using a luciferase reporter assay, where the complete 3'UTR of *HSD11B1* mRNA was inserted downstream of the gene for firefly luciferase, three potential miRNAs, hsa-miR-561, -579 and -340 were identified as potential negative regulators of *HSD11B1* expression. Moreover, disruption of the corresponding microRNA response elements (MREs) abolished repression of luciferase activity for hsa-miR-561 and -579, but not completely for hsa-miR-340. Therefore, hsa-miR-561 and hsa-miR-579-mediated downregulation of *HSD11B1* expression are strictly dependent on the binding of miR-561- and miR-579-MRE in the 3'UTR of *HSD11B1* mRNA. Levels of firefly luciferase mRNA were not changed by miRNA-561 and -579; and levels of endogenous *HSD11B1* mRNA were as well unchanged by miRNA-561 and -579, indicating a mechanism based on translational repression rather than on mRNA degradation. Interestingly, hsa-miR-579 was still performing downregulation of *HSD11B1* expression after treatment with glucocorticoids to induce *HSD11B1* expression, due to different regulatory mechanisms for *HSD11B1* expression by glucocorticoids and miRNAs in a dual luciferase assay system. The function of miRNA-561 and -579 could be blocked by anti-microRNA oligonucleotides (AMOs). MiRNA-561 and -579 were amplified by specific stem-loop reverse transcription primers and specific PCR primers from human hepatocytes and HepG2 cells. Although their relative contribution to *HSD11B1* expression remains unclear, literature findings and a pathway enrichment analysis of miRNA-561 and -579 target mRNAs support a role of these miRNAs in glucocorticoid metabolism/signalling and associated diseases.