The expression profile of microRNAs in CKD patients with vascular calcification

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Background

Chronic kidney disease (CKD) patients have a high risk of developing vascular media calcification, which is associated with significant cardiovascular morbidity and mortality. Our objective was to find key molecules, which regulate the phenotypic modulation of vascular smooth muscle cells. We focused on the role of microRNAs (miRNAs) and analysed the expression profile of 20 miRNAs in 53 patients receiving dialysis treatment. The respective miRNAs were further analysed in a mouse model of uremic media calcification.

Methods

Peripheral blood samples were obtained from 53 hemodialysis patients and 7 healthy controls. We determined the expression levels of several miRNAs with RT-qPCR. For our mouse experiments, we used the DBA/2NCrl mouse strain, which develop severe media calcification within days when put on high phosphate diet (HPD). C57BL/6 mice were used as controls. We measured the expression levels of the investigated miRNAs in the blood and in the aortic tissue in order to determine whether changes in their expression levels were associated with vascular calcification. Synthetic mmu-miR-142-3p mimic was administered via tail vein injection. The effects of the mimic were investigated by RT-qPCR, mass spectrometry and wire myography.

Results

microRNA	Patients	DBA/2NCrl	C57BL/6J
21	DOWN	DOWN	NOT
26b	DOWN	DOWN	NOT
98	DOWN	DOWN	NOT
98-3p	DOWN	NOT	NOT
103a	DOWN	DOWN	NOT
142-3p	DOWN	DOWN	NOT
146a	DOWN	DOWN	NOT
155	DOWN	DOWN	NOT

Table 1. The expression of microRNAs in human patients and mice.
The expression profile of several miRNAs in human patients receiving hemodialysis treatment compared to healthy controls (Patients) was similar to the expression of the respective microRNAs in the DBA/2NCrl mouse model for uremic media calcification. Control C57BL/6 mice had no regulation of these miRNAs. (DOWN: significant downregulation, UP: significant upregulation, NOT: not regulated)

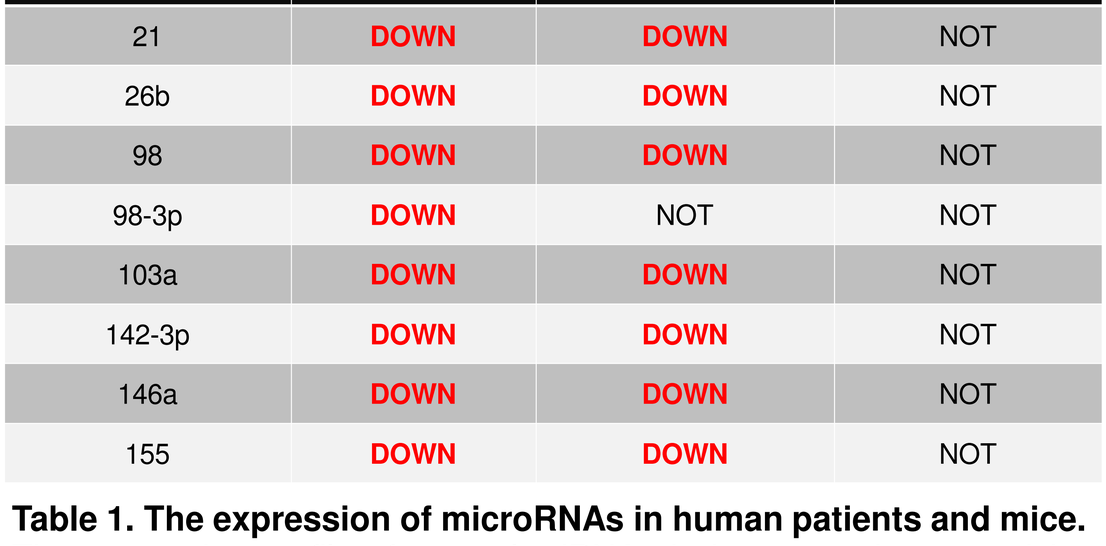

Figure 1. miR-142-3p expression.
miR-142-3p was significantly downregulated in the blood of the hemodialysis patients and in the blood of the DBA/2 mice on high phosphate diet. miR-142-3p was also downregulated in the aorta of these mice (A). C57BL/6 mice show no regulation of this miRNA (B).

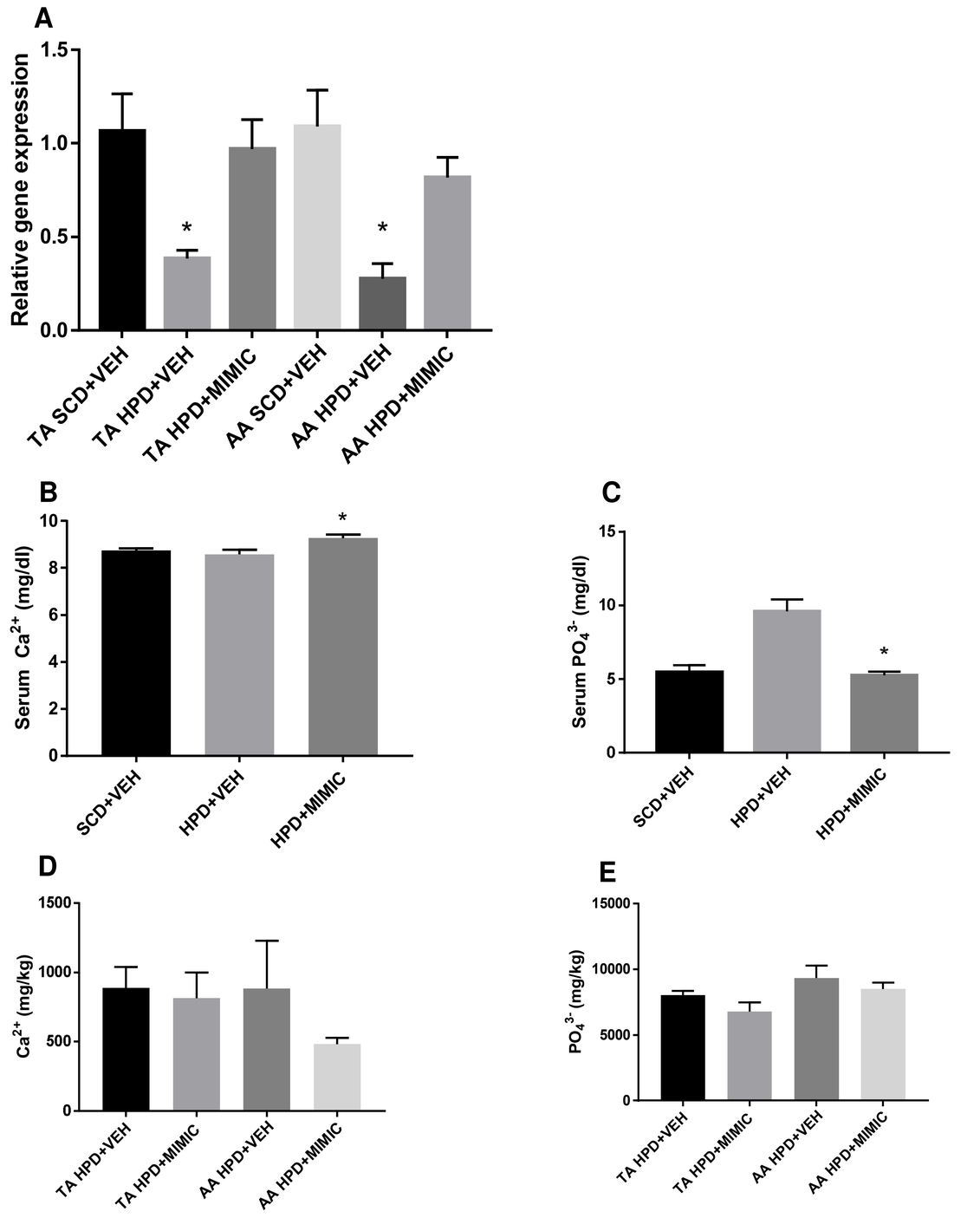

Figure 2. The effects of the syn-mmu-miR-142-3p injection.
48 hours after administration, the syn-mmu-miR-142-3p was detectable in the aorta of the mice and its expression was similar to the healthy mice and it was significantly different from the mice on HPD injected with vehicle (A). The mimic increased the serum calcium (B) and decreased the serum phosphate (C) levels. However after 48 hours there were no significant differences in the calcium (D) or phosphorus (E) content of the aortas.

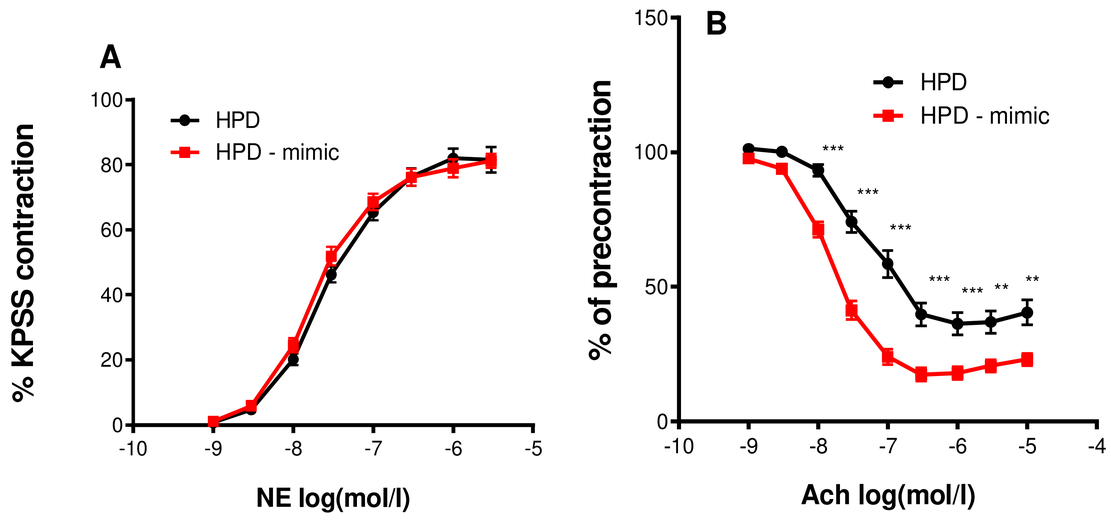

Figure 3. miR-142-3p affects relaxation.
The contraction of the aortic rings did not differ between the vehicle and the mimic treated groups (A). The rings isolated from the mice injected with the miR-142-3p mimic relaxed better compared to mice receiving vehicle (B).

Conclusion

These findings may indicate that miRNAs play a role in the process of uremic media calcification and their regulation is not an effect of the dietary phosphate. The expression level of miR-142-3p in the blood reflects its expression in the aorta and it is likely to play a functional role in the regulation of relaxation in the aorta.

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