

URINARY microRNA-21 EXPRESSION IN NEPHROPATHIES



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OBJECTIVES

MicroRNAs (miRNAs) are a class of noncoding RNA acting at a post-transcriptional level to control the expression of large sets of target mRNAs. While there is evidence that miRNAs deregulation plays a causative role in various complex disorders, their role in fibrotic kidney diseases is largely unexplored. However some data suggest, that increased circulating miR-21 levels are associated with kidney fibrosis. In this regard, we attempted to compare the level of miR-21 expression in the urine with signs of kidney lesions in patients (Pts) with nephropathy (Neph).

METHODS

Seventeen Pts with different Neph confirmed by kidney biopsy were examined: IgA nephropathy (n=4), focal segmental glomerulosclerosis (n=3), minimal change disease (n=2), membranoproliferative glomerulonephritis (n=2), AL amyloidosis (n=2), other (n=4). miR-21 expression in the urine was determined by a RT-PCR assay (EvaGreen fluorescence dye) and calculated using the $2^{-\Delta\Delta Ct}$ protocol. In Pts glomerular filtration rate (GFR) was assessed by creatinine clearance (CCr), Cockcroft&Gault, MDRD7 and CRD-EPI formulas. Daily urinary protein (UPE, g/24), creatinine (UCr, mmol/24), urea (UUr, mmol/24), potassium (UK, mmol/24), sodium (UNa, mmol/24), calcium (UCa, mmol/24), chloride (UCI, mmol/24) and inorganic phosphorus (UP, mmol/24) has been established. Fractional excretion (relative to CCr; FE, %) of all the previously mentioned chemical components of urine, except for protein and creatinine, was calculated. Total protein (TP, g/l) and albumin (Alb, g/l) serum concentrations also been taken into account. Percent of total sclerosed glomeruli and glomeruli with segmental sclerosis were calculated in the kidney biopsies (late microscopy). The degree of tubular atrophy (TA), diffuse (DF) and segmental (SF) interstitial fibrosis were evaluated semi-quantitatively on a scale from one to three stages (1 - mild changes, 2 - moderate changes, 3 - severe changes). miR-21 expression in the urine of healthy donors (n=11) was taken as control (C). All data are presented as mean \pm SE. Unpaired Student t-test and Spearman rank correlation coefficient (RS) were used.

RESULTS

The urine levels of miR-21 expression in Pts with Neph were significantly higher than in C (0.370 ± 0.0002 vs 0.031 ± 0.017 ; respectively, $p=0.0011$). There was strong positive correlation between urine miR-21 (RS=0.570, $p=0.017$) in Pts. In Neph the value of RS (-0.463, $p=0.061$) between miR-21 and Alb did not achieve statistical significance. In third stage (0.568 ± 0.122 ; n=7) of TA urine miR-21 was significant more than second (0.202 ± 0.067 ; n=7, $p=0.021$). Pts with first stage of TA were only three, that did not allow perform adequate statistical comparisons. There were no any significant associations between urine miR-21 and other functional or morphological parameters studied.

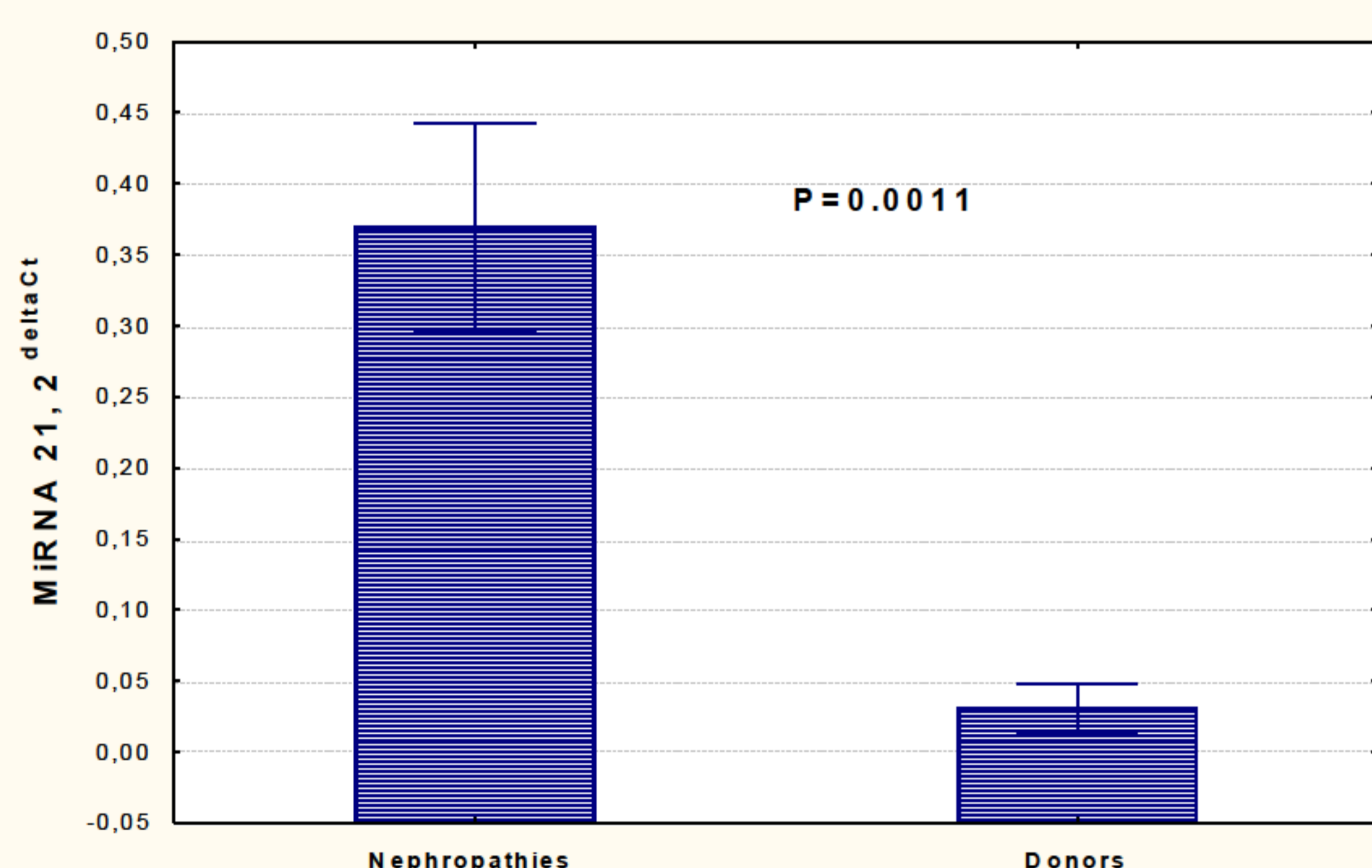


Fig.1
MiRNA 21 urinary expression in patients with nephropathies and healthy donors

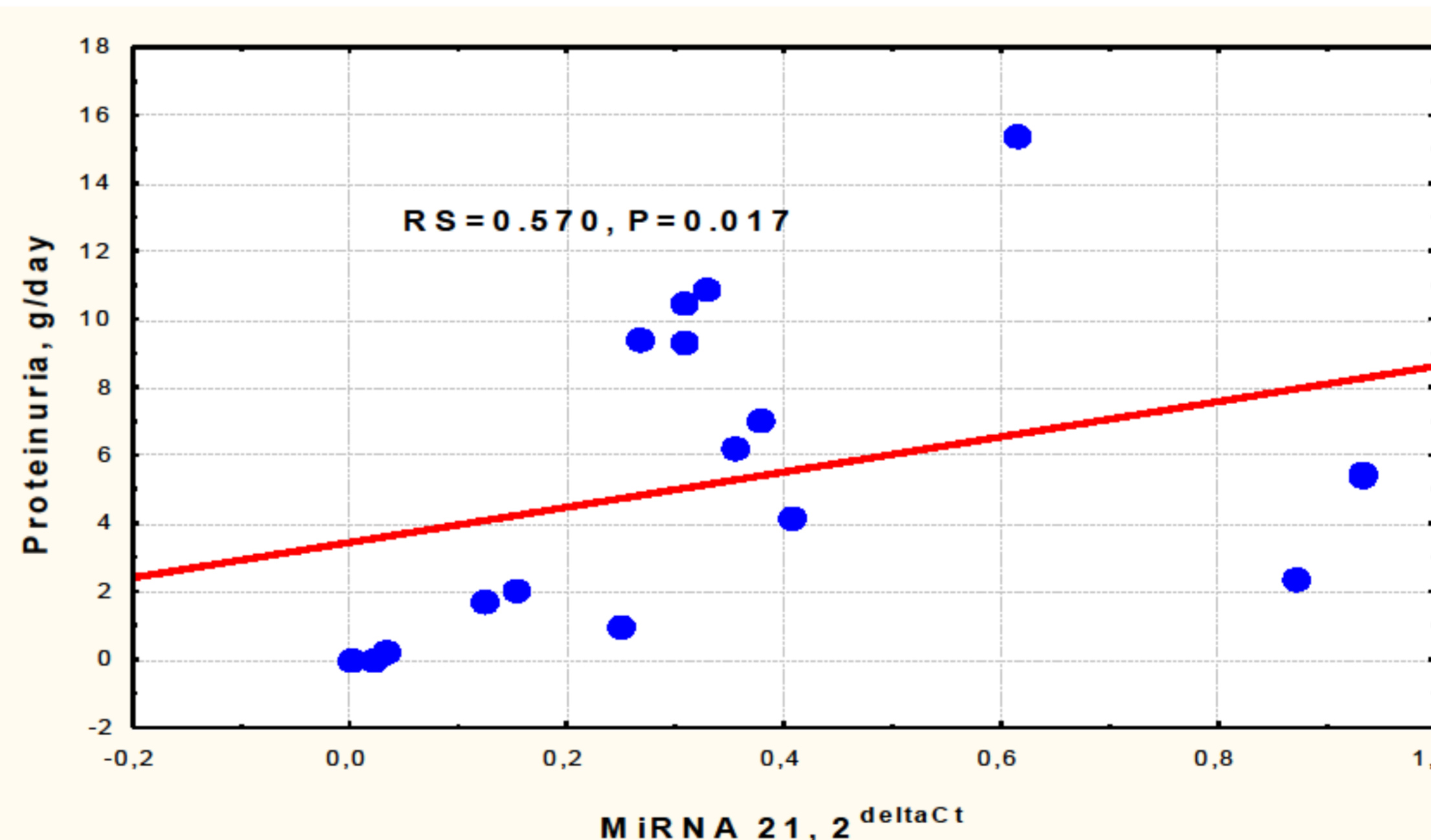


Fig. 2. Correlation between MiRNA urinary expression and proteinuria in pts with nephropathies

CONCLUSIONS

These data suggest that the level of miR-21 expression in the urine to a certain degree can be associated with the severity of renal damage in patients with nephropathies. However, to clarify the role of miR-21 in the development and progression of renal lesions further investigations are needed.

