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## INTRODUCTION

Despite its limitations, serum creatinine remains the most widely used test to estimate renal function, while microalbuminuria adds additional information on renal damage. Beta-trace protein (BTP) is a low molecular weight protein that has been proposed as an earlier biomarker of decreased glomerular filtration rate (GFR) and renal damage, than the traditional biomarkers, but its utility has not been characterized in animal models.

This study aimed to determine serum and urine patterns of renal markers alongside the occurrence of renal lesions in rat models of early and advanced chronic kidney disease (CKD) induced by nephrectomy.

## METHODOLOGY

### Experimental design

Male Wistar rats, 12 weeks old, were randomly divided in three surgical groups: Sham (n = 8, subjected to surgical process without kidney mass reduction), Early CKD (n = 8, subjected to 1/2 nephrectomy), and Advanced CKD (n = 7, subjected to 5/6 nephrectomy).

The project received approval (7/2020) from the local Organization Responsible for Animal Welfare (ORBEA).

### Biochemical analysis

After five weeks, rats were euthanized, blood and kidneys were collected. Circulating BTP levels were measured with a rat specific-ELISA kit (MyBiosource), and serum Cr was evaluated using automated methods (Roche Diagnostics).

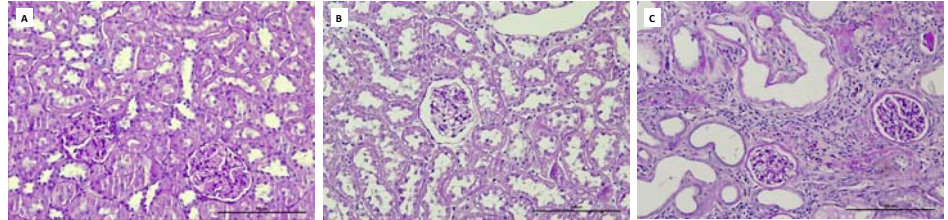
### Histopathological analysis

Renal tissue samples were formalin-fixed, embedded in paraffin wax, and 4µm thick sections were stained with Periodic acid–Shiff. Kidney lesions were identified and semi-quantitatively evaluated, according to the extension occupied by the lesion, as described [1].

## REFERENCE

[1] Lousa I., et al. TNFR2 as a Potential Biomarker for Early Detection and Progression of CKD. *Biomolecules*. 2023;13(3).

## RESULTS

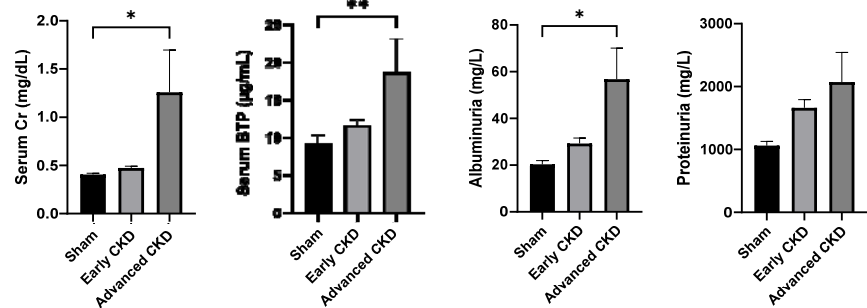


**Figure 1.** Representative images of the renal lesions observed in rats under study (PAS staining, 200x).

A. Sham group; showing no significant histopathological changes

B. Early CKD group; showing dilatation of Bowman's space, glomerular basement membrane thickening, tubular membrane and epithelial irregularities and tubular dilatation.

C. Advanced CKD group; showing Interstitial fibrosis and tubular atrophy (IFTA) and interstitial inflammatory infiltration.



**Figure 2.** Levels of serum creatinine, serum BTP, albuminuria and proteinuria in Sham (n = 8), Early CKD (n = 8) and Advanced CKD (n = 7) groups.

Results are presented as mean ± standard error of the mean (S.E.M.). The Kruskal-Wallis test was used to compare groups, followed by Bonferroni correction for multiple comparisons (\* p < 0.05, \*\* p < 0.01).

**Table 1.** Correlations between renal function markers with glomerular filtration rate (GFR) and the total score of histopathological lesions

		Serum Cr	Serum BTP	Albuminuria	Proteinuria
GFR	rs	-0.680	-0.453	-0.524	-0.135
	p	<0.001	0.030	0.010	0.539
Mild glomerular lesions	rs	0.805	0.541	0.448	0.528
	p	<0.001	0.008	0.032	0.010
Mild tubular lesions	rs	0.690	0.610	0.385	0.384
	p	<0.001	0.020	0.069	0.070
Advanced glomerular lesions	rs	0.724	0.455	0.486	0.491
	p	<0.001	0.029	0.019	0.017
Advanced tubular lesions	rs	0.639	0.517	0.413	0.169
	p	0.005	0.011	0.080	0.442

Spearman rank correlation coefficient (rs) test was used to determine the association between variables.

## CONCLUSIONS

In rat models of early and advanced CKD induced by nephrectomy:

- Histological changes in the early-stage group are not accompanied by significant variations in traditional renal function markers, neither BTP.
- In advanced CKD, serum creatinine and BTP seem to reflect the histopathological changes observed with disease progression.

The combined use of creatinine and BTP might improve disease staging.

## ACKNOWLEDGEMENTS

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