Evaluation of CHAC1 as a biomarker of hyperacute kidney injury after renal ischemia

P. Halbeisen[1,2]; K. M. Schmidt-Ott[1,2]; T. Westhoff[3]; E. Schrezenmeier[1,2,*]

1 Charité Universitätsmedizin Berlin, Department of Medicine/ Nephrology and Intensive care medicine, Berlin, Germany
2 Max Delbrück Center for Molecular Medicine in the Helmholtz Association, Berlin, Germany
3 Medical Department I, Universitätsklinikum Marien Hospital Herne, Ruhr University of Bochum, Germany
* E.S. is supported by the Charité Junior Clinical Scientist Program (Charité-Universitätsmedizin Berlin and the Berlin Institute of Health).

Purpose: Several novel biomarkers such as neutrophil gelatinase-associated lipocalin (NGAL) improve the early detection of acute kidney injury (AKI), but they are sensitive to stimuli other than kidney injury and remain elevated for long periods. We identified glutathione specific gamma-glutamylcyclotransferase 1 (CHAC1) as a potential new biomarker for early AKI in a mouse model of renal ischemia-reperfusion injury (IRI). In mice, CHAC1 is produced in injured proximal tubule cells in the early phase after ischemia (6 hours after IRI). Our goal was to evaluate the utility of urinary CHAC1 in a cohort of patients who underwent unilateral clamping of the renal artery during partial nephrectomy (nephron sparing surgery, NSS).

Table 1: Patient characteristics

	Group 1: NSS with ischemia (n=27)	**Group 2**: NSS without ischemia (n=10)	p-value
Epidemiology			
Male/Female	20 (74,1%) / 7 (25,9%)	8 (80%) / 2 (20%)	1
Age (in years)	65 (53-77)	63 (58-66,5)	0,39
Concomitant diseases			
Diabetes mellitus	7 (25,9%)	2 (20%)	1
Hypertension	18 (66,7%)	5 (50%)	0,45
Renal data on admission			
Plasma creatinine (mg/dl)	0,91 (0,81-1,07)	0,94 (0,86-1,21)	0,45
eGFR (ml/min/1,73m²)	82,6 (63,7-97,5)	76,53 (61,26-87,2)	0,37
Surgical data			
Tumour diameter (mm)	35 (25-45)	25 (14,5-49,8)	0,28
Operation time (min)	247 (185-275)	184,5 (160-264,8)	0,19
Ischemia time (min)	18 (11,5-21)	-	

Methods: The study population consisted of 37 patients. 27 patients underwent partial nephrectomy with unilateral clamping of the renal artery inducing IRI (group 1). 10 patients underwent partial nephrectomy without renal ischemia (group 2). Urinary samples were collected before surgery and serially after surgery. Urinary CHAC1 was detected by Western blotting using anti-CHAC1 antibody (Abcam ab155533).

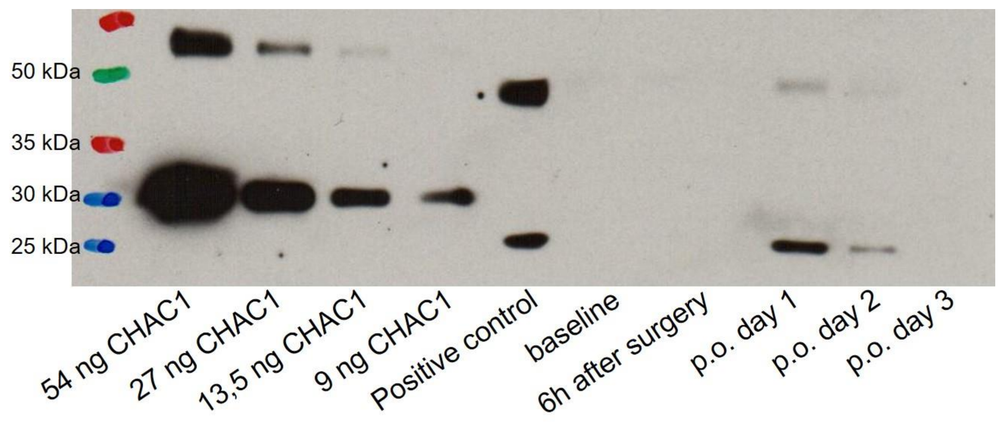

Figure 1: Western blot analysis of urinary CHAC1 in a group1 patient.

Semiquantitative analysis of band intensity was performed using ImageJ calibrating to known quantities of recombinant CHAC1 loaded onto adjacent lanes. Urinary NGAL and calprotectin were measured using enzyme-linked immunosorbent assay.

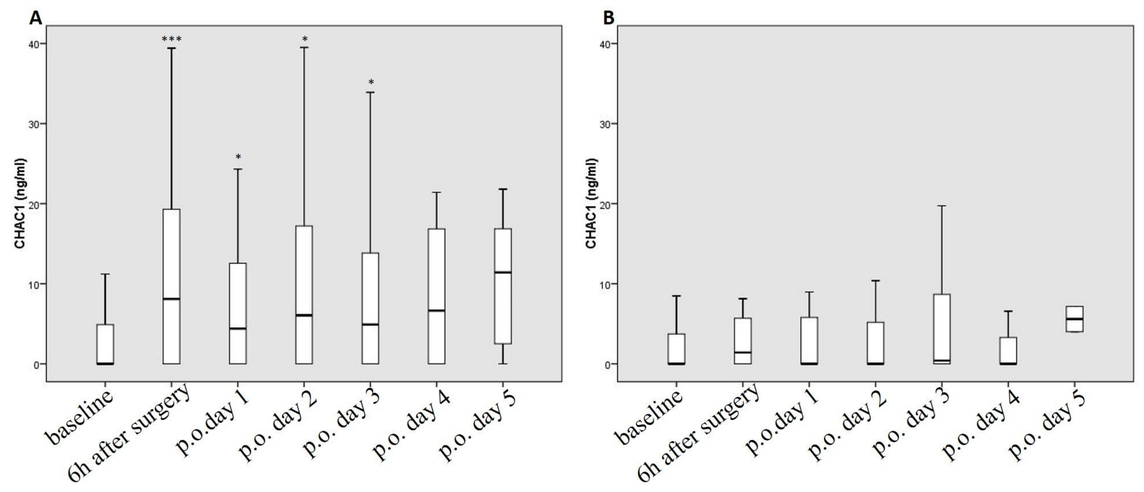

Figure 2: Pre- and postoperative (p.o.) concentrations of urinary CHAC1 in group 1 (A) and in group 2 (B). Asterisks indicate significant changes from baseline. *p<0.05, **p<0.01, ***p<0.001.

Results: According to the AKIN criteria, 12 out of 27 patients in group 1 (44,4%) and 2 out of 10 patients in group 2 (20%) developed AKI. Urinary CHAC1 concentration was significantly increased compared to preoperative baseline in group 1 from 6h up to 3 days after surgery, but not in group 2 (see Figure 2 for details). Highest CHAC1 concentrations in group 1 were reached 6h after surgery. CHAC1 on postoperative day 1 correlated positively with NGAL and Calprotectin in group 1.

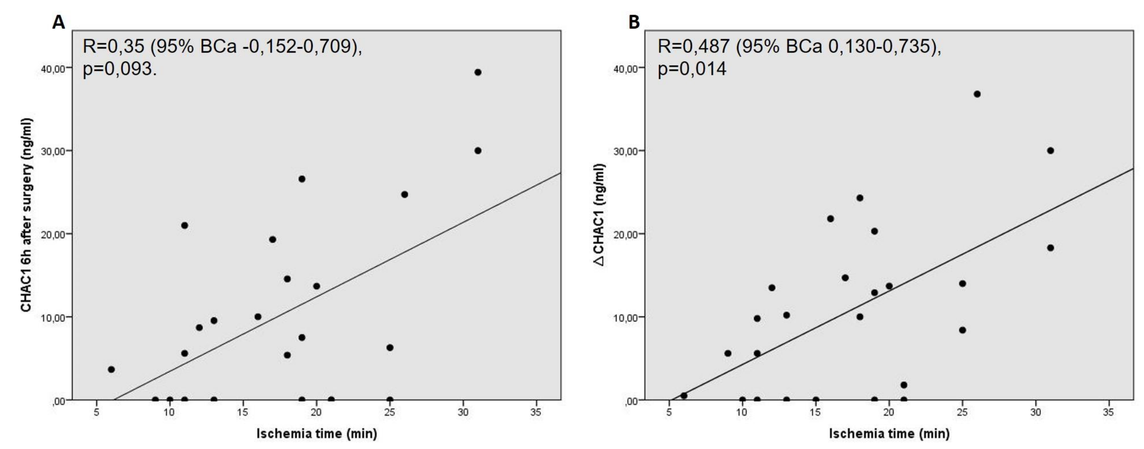

Figure 3: Correlation between ischemia time and (A) CHAC1 concentration 6h after surgery and (B) maximal change from baseline CHAC1 (ΔCHAC1) in group 1 (assessed by Spearman´s correlation test).

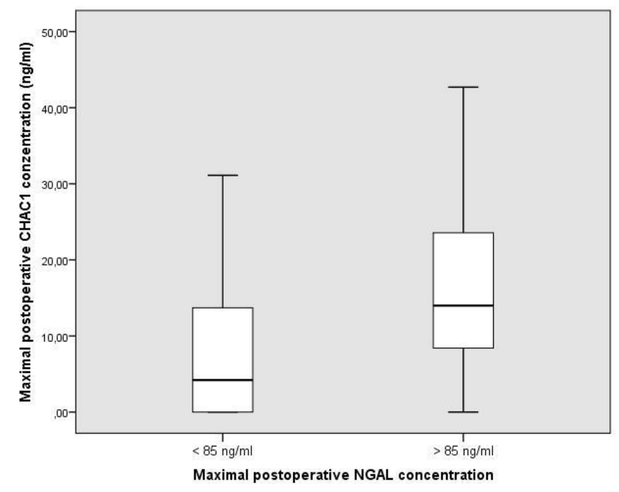

Figure 4: Maximal postoperative CHAC1 concentration in patients having a maximal postoperative NGAL concentration lower (N=18) or higher (N=19) than 85ng/ml. A non-parametric analysis of variance showed a significant difference between the two groups (p=0,031).

Conclusion: Urinary CHAC1 is a promising new biomarker for the detection of AKI after renal ischemia. CHAC1 increases early after renal ischemia and shows a rapid normalization. Future studies in different clinical settings must now further evaluate the diagnostic value of urinary CHAC1.