Effects of Sacubitril/Valsartan vs Irbesartan on Urine Tubular Biomarkers in CKD: Findings from the UK HARP-III Trial

Writing Committee and Affiliations

Michelle A Goonasekera* MD [a,b], Greco B Malijan* MD [a], Rebecca J Sardell PhD [a], Natalie Staplin PhD [a], Benjamin S Storey MD [b], Daniel Chapman PhD [c], Michael Hill DPhil [c], Stewart Moffat BSc [c], Abdulrahman Al-Mohammad MBBChir [a], Doreen Zhu MD [a,b], Nigel J Brunskill PhD [d,e], John JV McMurray MD [f,g], Maarten W Taal MD [h,i], William G Herrington MD [a,b], Martin J Landray PhD [a], Colin Baigent FMedSci [a], Richard Haynes† DM [a,b], Parminder K Judge† PhD [a,b]

*Joint first author contribution

†Joint senior authorship

[a] Renal Studies Group, Clinical Trial Service Unit and Epidemiological Studies Unit, Nuffield Department of Population Health, University of Oxford, Oxford, United Kingdom

[b] Oxford Kidney Unit, Oxford University Hospitals NHS Foundation Trust, Oxford, United Kingdom

[c] Wolfson Laboratory, Clinical Trial Service Unit and Epidemiological Studies Unit, Nuffield Department of Population Health, University of Oxford, Oxford, United Kingdom

[d] College of Life Sciences, University of Leicester

[e] Department of Nephrology, University Hospitals of Leicester NHS Trust

[f] British Heart Foundation Glasgow

[g] Cardiovascular Research Centre, School of Cardiovascular and Metabolic Health, University of Glasgow, Glasgow, United Kingdom

[h] Centre for Kidney Research and Innovation, Academic Unit for Translational Medical Sciences, School of Medicine, University of Nottingham, United Kingdom

[i] Department of Renal Medicine, University Hospitals of Derby and Burton NHS Foundation Trust, Derby, United Kingdom

Short Title

Urine Tubular Biomarkers in UK HARP III

Correspondence

Parminder K Judge

Renal Studies Group at the University of Oxford, Clinical Trial Service Unit and Epidemiological Studies Unit (CTSU), Nuffield Department of Population Health, University of Oxford, Richard Doll Building, Old Road Campus, Roosevelt Drive, Oxford OX3 7LF, UK.

Email: parminder.judge@ndph.ox.ac.uk. Tel: +44 1865 743743

Keywords

sacubitril/valsartan, chronic kidney disease, tubular biomarkers, neutrophil gelatinase-associated lipocalin

Word count, figures and tables

Abstract: 188, Body text word count: 915,

References: 10, Figures: 1

Supplementary Material:

Supplementary Methods, Supplementary Tables: 1, Supplementary Figures: 2

ABSTRACT

Introduction: Sacubitril/valsartan shows benefits in heart failure and may have kidney protective effects. Its impact on kidney tubular health in chronic kidney disease (CKD) remains unclear. We evaluated the effects of sacubitril/valsartan on urinary markers of tubular dysfunction and injury in UK Heart and Renal Protection-III (HARP III, ISRCTN:11958993).

Methods: Urine tubular biomarkers were measured at baseline, 3 and 6 months using first morning void or spot urine samples among 411 participants from UK HARP III. A mixed model repeated measures approach was used to quantify the study average effect of treatment on the urine biomarkers.

Results: Compared to allocation to irbesartan, allocation to sacubitril/valsartan reduced neutrophil-gelatinase associated lipocalin (NGAL), a marker secreted in the distal tubules after ischemia and reperfusion, by 18% (95% CI: -32% to -1%). No significant changes were observed for the other biomarkers, and there was no evidence of effect modification by key baseline characteristics across all biomarkers studied.

Conclusion: In UK HARP-III, sacubitril/valsartan reduced urinary NGAL compared with irbesartan but did not affect other tubular biomarkers of injury, ischemia and fibrosis, suggesting limited tubular benefits, consistent with no observed effect on kidney function.

INTRODUCTION

Neprilysin inhibition prevents breakdown of natriuretic peptides mediating natriuresis, diuresis, renin-angiotensin system inhibition, and anti-inflammatory processes, and may benefit the kidneys. [1] Sacubitril/valsartan, an angiotensin receptor-neprilysin inhibitor, was shown to reduce the risk of cardiovascular death or hospitalization for heart failure by 20% (hazard ratio 0.80 [95%CI 0.71-0.89]), compared with enalapril, in 8442 patients with heart failure with reduced ejection fraction, and is a guideline-recommended treatment for this group. [2] Allocation to sacubitril/valsartan, compared to enalapril, delayed progression of estimated glomerular filtration rate (eGFR) decline (-1.61 vs -2.04 mL/min/1.73m^2/year respectively) but increased albuminuria (1.2 vs 0.9 mg/mmol). [3]

The UK Heart and Renal Protection (HARP)-III trial (ISRCTN:11958993) compared the effects of sacubitril/valsartan 97/103 mg twice daily with irbesartan 300 mg once daily in 414 patients with chronic kidney disease (CKD) at risk of progression. [4] Sacubitril/valsartan, compared to irbesartan, had no significant effect on the primary outcome of measured GFR at 12 months (29.8 [SE 0.5] vs 29.9 [0.5] mL/min/1.73m^2 respectively; difference, -0.1 [0.7] mL/min/1.73m^2, p=0.86). Sacubitril/valsartan reduced systolic blood pressure by 5.4 (3.4-7.4) mmHg, troponin I by 16% (8-23%), and NT-proBNP by 18% (11-25%). In contrast to heart failure populations, urine albumin-to-creatinine ratio (uACR), was non-significantly reduced by 9% (-18, 1%, p=0.08). [4]

Effects of sacubitril/valsartan on urine biomarkers kidney injury molecule-1 (KIM-1; a transmembrane glycoprotein shed by proximal tubular cells during injury) and neutrophil gelatinase-associated lipocalin (NGAL; a lipocalin secreted from damaged distal tubular cells after ischemia and reperfusion) were pre-specified exploratory outcomes. Effects on these biomarkers, and additional urine biomarkers reflecting kidney tubular health and dysfunction measured in post-hoc exploratory analyses, [5] are presented in this brief report.

METHODS

The UK HARP-III trial methods and main results have been reported previously. [4,6] Participants provided first morning void urine (or if unavailable spot urine) samples at randomization, 3 and 6 months for urine biomarker measurements. All measurements were indexed to urine creatinine. In addition to uACR, KIM-1, and NGAL, urine biomarkers

measured included markers of: proximal tubular dysfunction (alpha-1 microglobulin [α1M]); functional reserve (epidermal growth factor, uromodulin); tubular stress and fibrosis (dickkopf-3 [DKK-3], monocyte chemoattractant protein-1, human cartilage glycoprotein-40 [YKL-40]); and inflammation and injury (interleukin-18 [IL-18]).[5]

A linear mixed model repeated measures (MMRM) approach was used to quantify the effect of treatment on study average urine biomarkers. No formal correction for multiplicity of testing was made. In post-hoc explorations, interaction terms for key subgroups (diabetes, eGFR, level of albuminuria, and primary kidney disease) were used to evaluate any effect modification. Relative differences in treatment effect at each time point were also estimated. All analyses were performed using R version 4.4.2 and SAS version 9.4. Details of biomarker measurements and statistical approach are in the Supplementary Methods.

RESULTS

Of 414 UK HARP-III participants, 411 had at least one valid biomarker measurement at 3 or 6 months. Mean (SD) eGFR was 35.5±10.9 mL/min/1.73m^2 and median (Q1-Q3) uACR was 478 (97-1354) mg/g. The most abundant creatinine-indexed urine tubular biomarker at baseline was α1M (39 [21-62] mg/g), and the least abundant was IL-18 (98 [41-191] ng/g). Characteristics of included participants were comparable across the two randomized arms, including urine biomarker levels at baseline (Supplementary Table S1).

Sacubitril/valsartan reduced NGAL by 18% (-32, -1%) compared with irbesartan (Figure 1). This change was evident at 3 months and persisted at 6 months (Supplementary Fig. S1). There was no significant effect on the other biomarkers studied, including KIM-1 (-2% [-10, 8%]). Post-hoc subgroup analyses showed no evidence of effect modification by key characteristics across the pre-specified tubular biomarkers (Supplementary Fig. S2).

DISCUSSION

In observational studies, higher urine NGAL is associated with distal tubular injury and lower eGFR. [5,7] Our findings are consistent with studies in animal models of cardiorenal syndrome, which show a reduction in urine NGAL with sacubitril/valsartan compared to valsartan, possibly related to improvements in cardiac and kidney perfusion and reduced

inflammation and injury. [8] Neprilysin inhibition is proposed to attenuate kidney injury in models of kidney disease via several mechanisms: suppression of pro-inflammatory cytokines (including pro-IL-1β), which drives NGAL expression upon activation; promotion of regulators of oxidative pathways; and reduced fibrosis. [8,9] However, in UK HARP-III, sacubitril/valsartan, compared with irbesartan, had no demonstrable effect on measured or estimated GFR, which would be indicative of improved kidney perfusion. There was also no significant effect on other urine markers of tubular injury (IL-18 and KIM-1) or fibrosis (DKK-3), nor were there any significant changes in the other urine biomarkers studied. Despite the small increase in albuminuria observed in the heart failure trials of sacubitril/valsartan, urine biomarker results from UK HARP-III suggest there is no evidence of adverse effects on kidney tubular health.

Study limitations include the relatively small sample size, short duration of follow-up, and use of biomarkers with large within-person biological variability (e.g., YKL-40), resulting in considerable uncertainty in effect estimates. Therefore, small effects on urine biomarkers other than NGAL may be undetectable. As data on initial storage temperatures were unavailable, the MMRM analysis could not be adjusted for potential biomarker instability at higher freezer temperatures. We assumed that both angiotensin receptor blockers (valsartan and irbesartan) would have comparable effects on urine tubular biomarkers, but it is possible small differences exist due to their distinct pharmacological profiles. [10] Lastly, the effect on NGAL is imprecisely quantified and could represent a chance finding.

In UK HARP-III, sacubitril/valsartan reduced NGAL by 18% compared to irbesartan. However, sacubitril/valsartan did not affect other urine tubular biomarkers of functional reserve, injury, or fibrosis consistent with the lack of effect observed on kidney function.

STATEMENTS

Acknowledgements

We thank the participants, the local site staff, and all members of the UK HARP-III steering and data monitoring committees.

Statement of Ethics

UK HARP-III was reviewed and approved by the Nottingham Research Ethics Committee 2 (approval number 13/EM/0434). All participants provided written informed consent to participate in the study.

Conflict of Interest Statement

MAG, GBM, RJS, NS, DC, MH, AAM, DZ, MJL, CB, RH, WGH, and PKJ report institutional grant funding from Boehringer Ingelheim and Eli Lilly for the EMPA-KIDNEY trial. MAG reports institutional grant funding from Health Data Research UK, Novartis and Novo Nordisk. NS reports institutional grant funding from Novo Nordisk. JJVM reports institutional grant funding from the British Heath Foundation, the National Institute for Health – National Heart, Lung and Blood Institute, Alnylam Pharmaceuticals, AstraZeneca, Bayer, Cardurion, Novartis, Roche, and Cytokinetics paid to the University of Glasgow as well as personal consulting for Alnylam Pharmaceuticals, AnaCardio, AstraZeneca, Bayer, Cardurion, Cytokinetics, Novartis, River BioMedics, Biohaven Pharmaceuticals, Chugai Pharmaceuticals, Protherics Medicine Developments Ltd., and DalCor Pharmaceuticals. MWT reports consulting fees from Boehringer Ingelheim; honoraria from Bayer, Wiley Publishers, and Boehringer Ingelheim; support to attend conference from Bayer; royalties from Elsevier as an Editor of The Kidney; and a leadership role in the International Society of Nephrology. CB reports grant funding from the Medical Research Council, National Institute for Health and Care Research (NIHR) Health Technology Assessment (HTA) (17/140/02) and Health Data Research UK; and advisory roles for NIHR HTA, the British Heart Foundation, and the European Society of Cardiology. WGH reports advisory roles for the UK Kidney Association, European Renal Association, European Society of Cardiology, and KDIGO. RH reports institutional grant funding from Novartis and trial drug supply from Roche and Regeneron. MWT, WGH, and PKJ were members of the journal's Editorial Board at the time of submission. The other authors have no conflicts of interest to declare.

Funding Statement

The UK HARP-III trial was funded by a grant from Novartis to the University of Oxford to perform the trial.

Author Contributions

Richard Haynes and Parminder K Judge selected the urine biomarkers and designed the substudy. Richard Haynes, Martin J Landray and Colin Baigent secured trial funding. Richard Haynes, Parminder K Judge, Benjamin S Storey, Doreen Zhu, Nigel J Brunskill, John JV McMurray, and Maarten W Taal collected the data. Daniel Chapman, Michael Hill, and Stewart Moffat conducted the laboratory analyses. Greco B Malijan, William G Herrington, Parminder K Judge, Rebecca Sardell, Natalie Staplin & Richard Haynes developed the data analysis plan. Greco B Malijan performed the statistical analyses under supervision from Rebecca Sardell & Natalie Staplin. Michelle A Goonasekera and Greco B Malijan wrote the first draft of the manuscript, with support from Abdulrahman Al-Mohammad. All authors contributed to interpretation of the results and revision of the manuscript.

Data Availability Statement

Data used in this publication are available in line with the policy and procedures described at: https://www.ndph.ox.ac.uk/data-access. For further information, contact the corresponding author.

REFERENCES

1 Campbell DJ. Long-term neprilysin inhibition - implications for ARNIs. Nat Rev Cardiol. 2017 Mar;14(3):171–86.

2 McMurray JJV, Packer M, Desai AS, Gong J, Lefkowitz MP, Rizkala AR, et al. Angiotensin–Neprilysin Inhibition versus Enalapril in Heart Failure. New England Journal of Medicine. 2014 Sept;371(11):993–1004.

3 Damman K, Gori M, Claggett B, Jhund PS, Senni M, Lefkowitz MP, et al. Renal Effects and Associated Outcomes During Angiotensin-Neprilysin Inhibition in Heart Failure. JACC Heart Fail. 2018 June;6(6):489–98.

4 Haynes R, Judge PK, Staplin N, Herrington WG, Storey BC, Bethel A, et al. Effects of Sacubitril/Valsartan Versus Irbesartan in Patients With Chronic Kidney Disease. Circulation. 2018 Oct;138(15):1505–14.

5 Ix JH, Shlipak MG. The Promise of Tubule Biomarkers in Kidney Disease: A Review. American Journal of Kidney Diseases. 2021 Nov;78(5):719–27.

6 UK HARP-III Collaborative Group. Randomized multicentre pilot study of sacubitril/valsartan versus irbesartan in patients with chronic kidney disease: United Kingdom Heart and Renal Protection (HARP)- III—rationale, trial design and baseline data. Nephrol Dial Transplant. 2016;32(12):2043–51.

7 Catanese L, Siwy J, Mischak H, Wendt R, Beige J, Rupprecht H. Recent Advances in Urinary Peptide and Proteomic Biomarkers in Chronic Kidney Disease: A Systematic Review. Int J Mol Sci. 2023 May;24(11):9156.

8 Rahman MM, Rahman A, Nishiyama A. Potential renoprotective effects and possible underlying mechanisms of angiotensin receptor-neprilysin inhibitors in cardiorenal syndrome. Front Med (Lausanne). 2024;11:1451450.

9 Jing W, Vaziri ND, Nunes A, Suematsu Y, Farzaneh T, Khazaeli M, et al. LCZ696 (Sacubitril/valsartan) ameliorates oxidative stress, inflammation, fibrosis and improves renal function beyond angiotensin receptor blockade in CKD. Am J Transl Res. 2017 Dec;9(12):5473–84.

10 Belz GG, Breithaupt-Grögler K, Butzer R, Fuchs W, Hausdorf C, Mang C. The pharmacological potency of various AT(1) antagonists assessed by Schild regression technique in man. J Renin Angiotensin Aldosterone Syst. 2000 Dec;1(4):336–41.

FIGURE LEGENDS

Fig. 1. Effect of sacubitril/valsartan versus irbesartan on study average creatinine-indexed urine biomarker concentrations.

The mixed model repeated measures model included treatment allocation, time, treatment-by-time interaction, baseline biomarker concentration, and baseline-by-time interaction. The study average values include estimates at 3 and 6 months. The dashed line indicates the transition of units from mg/g to ng/g. The size of the boxes represents the amount of information estimated.