

Integrated TGF- β 1, Aldosterone and Genotype Profiling for Risk Stratification and Treatment Response in Chronic Heart Failure with Cardiorenal Syndrome

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Abstract Cardiorenal syndrome (CRS) in the setting of chronic heart failure (CHF) is driven by hemodynamic, neurohormonal and fibrotic mechanisms. Transforming growth factor- β 1 (TGF- β 1), aldosterone and collagen IV are key fibrosis-related biomarkers, while functional polymorphisms of TGF- β 1 (rs1800469, rs1800470, rs1800471, rs1800473) and aldosterone synthase (CYP11B2 -344 T/C) may modulate individual susceptibility and treatment response. To evaluate the clinical significance of circulating TGF- β 1, aldosterone and collagen IV, and their interaction with TGF- β 1 and CYP11B2 genotypes for risk stratification and response to guideline-directed therapy plus SGLT2 inhibition in CHF patients with and without CRS. We prospectively enrolled 200 patients with CHF of ischaemic origin (NYHA II–IV) and 40 apparently healthy controls. Patients were divided into two groups: CHF with CRS (CRS+, n=100) and CHF without CRS (CRS–, n=100). All patients received standard guideline-based therapy (ACEI/ARB, β -blocker, mineralocorticoid receptor antagonist, diuretics); a subset with CRS additionally received dapagliflozin 10 mg/day for 6 months. Serum TGF- β 1, aldosterone and collagen IV, creatinine, urea and cystatin C were measured by ELISA. Glomerular filtration rate (eGFR) was estimated. TGF- β 1 polymorphisms (rs1800469, rs1800470, rs1800471, rs1800473) and CYP11B2 -344 T/C were genotyped by PCR–RFLP. Clinical status (NYHA class, symptom score), 6-minute walk test (6MWT) and echocardiographic indices were assessed at baseline and after 6 months.

Keywords Cardiorenal syndrome, Chronic heart failure, TGF- β 1, Aldosterone, Collagen IV, CYP11B2, Genetic polymorphism, SGLT2 inhibitor, Dapagliflozin, Fibrosis biomarkers

1. Introduction

Cardiorenal syndrome (CRS) represents a bidirectional and self-perpetuating interaction between the heart and kidneys, in which acute or chronic dysfunction of one organ induces or aggravates dysfunction of the other [1,2,3]. In patients with chronic heart failure (CHF), CRS significantly increases morbidity, mortality, hospitalization rates and health-care costs [2,4]. Epidemiological data indicate that in end-stage renal disease up to 40% of patients have coronary artery disease and up to 75% have left ventricular hypertrophy, while in chronic kidney disease (CKD) of any stage cardiovascular events account for more than half of all deaths [3,5].

The pathophysiology of CRS is complex and involves haemodynamic compromise, activation of the renin-angiotensin-aldosterone system (RAAS) and sympathetic nervous system, inflammation, oxidative stress and progressive fibrosis in

both myocardium and renal parenchyma [1,4,6,7]. Among molecular mediators, transforming growth factor- β 1 (TGF- β 1) is recognized as a master regulator of fibrosis and extracellular matrix remodeling [8,9]; aldosterone promotes sodium retention, vascular and myocardial fibrosis and adverse cardiac remodeling [3,6,7,10]; collagen IV reflects extracellular matrix deposition and structural damage in the heart and kidneys [8,9].

Genetic background influences susceptibility to CHF, CKD and CRS. Functional polymorphisms in the TGF- β 1 gene (e.g. rs1800469, rs1800470, rs1800471, rs1800473) and in the aldosterone synthase gene CYP11B2 (-344 T/C) have been associated with altered cytokine or hormone production, blood pressure regulation, degree of cardiac and renal fibrosis, and variable treatment response in different populations [5,6,8,9,11–14]. In particular, -344 T/C polymorphism in the CYP11B2 promoter has been linked to enhanced transcriptional activity, higher aldosterone levels and increased risk of hypertension, diabetic kidney disease and adverse cardiovascular remodeling [11,12,13,14]. However, data on these variants in Central Asian, particularly Uzbek,

populations with CHF and CRS are scarce.

SGLT2 inhibitors such as dapagliflozin have emerged as key cardio-renal protective agents, improving outcomes in CHF with reduced and preserved ejection fraction, as well as in CKD, partly through haemodynamic, metabolic and antifibrotic mechanisms [15,16,17]. Nevertheless, inter-individual variability in clinical benefit is prominent, suggesting that underlying molecular and genetic factors, including profibrotic signalling and aldosterone activity, may modulate drug response [3,7,10,15,16,17,18,19,20].

The present study was designed to integrate circulating fibrosis-related biomarkers (TGF- β 1, aldosterone, collagen IV) with TGF- β 1 and CYP11B2 genotyping in order to (i) characterize their relationship with CRS and organ dysfunction in Uzbek patients with CHF, and (ii) assess genotype-dependent differences in response to guideline-directed therapy plus SGLT2 inhibition.

2. Materials and Methods

This prospective observational study was conducted between 2022 and 2025. We enrolled 200 consecutive patients with chronic heart failure of ischaemic origin (NYHA II–IV) and 40 age- and sex-matched apparently healthy volunteers as controls. Patients were divided into two groups:

- CRS+ group: 100 CHF patients with chronic cardiorenal syndrome (reduced eGFR and/or structural kidney damage according to KDIGO 2024 criteria).
- CRS– group**: 100 CHF patients without CRS (preserved renal function).

Controls (n=40) had no history or evidence of cardiovascular or renal disease. All participants provided written informed consent.

Inclusion and exclusion criteria

Inclusion criteria for CHF patients:

- CHF diagnosed according to ESC 2023 guidelines;
- ischaemic aetiology;
- stable clinical condition at enrolment;
- age ≥ 18 years.

Major exclusion criteria:

- recent acute myocardial infarction or unstable angina;
- acute stroke;
- severe valvular disease requiring surgery;
- active systemic inflammatory or autoimmune disease;
- advanced hepatic failure;
- active malignancy;
- severe psychiatric disorder or alcohol abuse.

Treatment protocol

All CHF patients received guideline-directed medical therapy including an ACE inhibitor or ARB, β -blocker, mineralocorticoid receptor antagonist and diuretics as indicated. In the CRS+ group, dapagliflozin 10 mg once daily was added and continued for 6 months.

Clinical and functional assessment

At baseline and at 6-month follow-up, the following were assessed:

- NYHA functional class and a clinical status score (including dyspnoea, peripheral oedema, pulmonary rales, hepatomegaly, heart rate and systolic blood pressure);
- quality of life using the Minnesota Living with Heart Failure Questionnaire (MLHFQ);
- exercise capacity by 6-minute walk test (6MWT);
- standard echocardiography: left ventricular end-diastolic and end-systolic diameters and volumes, left ventricular ejection fraction (LVEF).

Laboratory measurements. Venous blood samples were collected after an overnight fast. Serum creatinine, urea and electrolytes were measured by standard biochemical methods; eGFR was calculated using accepted equations. Serum cystatin C, TGF- β 1, aldosterone and collagen IV were determined by enzyme-linked immunosorbent assay (ELISA). NT-proBNP was measured by immunoassay.

Genotyping. Genomic DNA was isolated from peripheral blood leucocytes using standard kits. TGF- β 1 polymorphisms (rs1800469, rs1800470, rs1800471, rs1800473) and CYP11B2 –344 T/C were genotyped by polymerase chain reaction with restriction fragment length polymorphism (PCR–RFLP) using specific primers and restriction endonucleases. Genotype and allele frequencies were tested for Hardy–Weinberg equilibrium.

Statistical analysis

Data were analysed using IBM SPSS 26.0. Continuous variables are presented as mean $\pm$ standard deviation (SD) or standard error (SE); categorical variables as percentages. Between-group comparisons were performed with Student's t-test or ANOVA for continuous variables and χ^2 test for categorical variables. Paired t-test was used to compare baseline and 6-month values. Correlations between biomarkers and functional indices were assessed by Pearson or Spearman coefficients as appropriate. Odds ratios (OR) and 95% confidence intervals (CI) were calculated to estimate associations between genotypes and CRS. A p-value < 0.05 was considered statistically significant.

3. Results

Baseline clinical and biomarker profile

CRS+ patients had more pronounced symptoms (higher clinical score, more oedema and pulmonary congestion), higher NYHA class and lower 6MWT distance compared with CRS– patients, while controls were asymptomatic.

Serum creatinine, urea and cystatin C were significantly higher and eGFR significantly lower in CRS+ than in CRS– and control groups (all $p < 0.001$). NT-proBNP levels were markedly elevated in CHF groups, especially in CRS+.

Fibrosis-related biomarkers showed a graded increase

from controls to CRS– to CRS+:

- TGF- β 1: 35.5 ± 0.61 (controls) vs 57.9 ± 0.52 (CRS–) vs 85.6 ± 3.6 pg/mL (CRS+);
- Aldosterone: 95.9 ± 0.53 vs 170.2 ± 0.92 vs 212.2 ± 1.02 pg/mL;
- Collagen IV: 8.65 ± 0.18 vs 14.1 ± 0.14 vs 19.9 ± 0.2 ng/mL (all $p < 0.001$).

In the CRS+ group, TGF- β 1 correlated positively with creatinine and cystatin C and negatively with eGFR. Aldosterone correlated positively with creatinine, cystatin C, collagen IV and left ventricular mass, and negatively with eGFR and LVEF.

TGF- β 1 and CYP11B2 genotypes and CRS risk

Genotype distributions differed significantly between groups. For illustration:

- TGF- β 1 rs1800473 TT and rs1800471 GG genotypes were more frequent in CRS+ patients and associated with higher TGF- β 1 levels and more severe renal and cardiac dysfunction.
- “Protective” genotypes (e.g. rs1800469 CC, rs1800470 CC, rs1800471 CC, rs1800473 CC) were relatively under-represented in CRS+ and associated with lower biomarker levels.

CYP11B2 –344 TT genotype and T allele were significantly enriched in CRS+ compared with controls; carriers had higher aldosterone and collagen IV, higher creatinine and lower eGFR. Conversely, the CC genotype and C allele were associated with a lower fibrosis and neurohormonal burden.

Genotype-dependent treatment response

In CRS+ patients treated with dapagliflozin for 6 months, overall eGFR increased by approximately 10–11%, TGF- β 1 and aldosterone decreased by about 21%, and collagen IV declined significantly. NYHA class improved, 6MWT distance increased and LVEF rose modestly.

However, when stratified by genotype, the magnitude of improvement varied:

TGF- β 1 variants

- rs1800473 TT carriers showed greater increases in eGFR and larger reductions in TGF- β 1 than unfavourable genotypes in some models, whereas in other SNPs (e.g. rs1800471) CC carriers demonstrated better response than GG. Overall, haplotypes combining “low-producer” alleles were associated with stronger biomarker and functional improvements.

- CYP11B2 –344 T/C

- CC genotype carriers exhibited the largest decrease in aldosterone (~30% or more) and collagen IV, greater improvement in eGFR and better gains in 6MWT distance compared with TT carriers, who had more modest changes.

In all genotype groups, creatinine and cystatin C declined, but reductions were more pronounced in patients with “protective” TGF- β 1 and CYP11B2 genotypes.

4. Discussion

This study demonstrates that in Uzbek patients with CHF, particularly those with CRS, circulating TGF- β 1, aldosterone and collagen IV are not only markers of fibrosis and organ dysfunction, but also reflect underlying genetic susceptibility conferred by TGF- β 1 and CYP11B2 polymorphisms.

The strong association between high TGF- β 1, elevated aldosterone, increased collagen IV and reduced eGFR is consistent with experimental and clinical data implicating TGF- β 1 as a central mediator of myocardial and renal fibrosis, and aldosterone as a key driver of RAAS-dependent remodeling. Our findings extend these observations by showing that patients carrying specific functional variants of TGF- β 1 and CYP11B2 genes form distinct risk strata with respect to CRS development and progression.

Importantly, the study suggests that the benefit of SGLT2 inhibition is modulated by genetic background. While dapagliflozin improved renal function, fibrosis biomarkers and functional capacity in the overall CRS+ cohort, the treatment effect was substantially greater in carriers of “favourable” genotypes, particularly CYP11B2 CC and TGF- β 1 variants associated with lower cytokine expression. Conversely, “risk” genotypes showed attenuated responses, which may partly explain clinical heterogeneity seen in large outcome trials.

From a translational standpoint, integrating biomarker and genotype data could help:

- identify CHF patients at highest risk of CRS and accelerated fibrosis;
- select those most likely to benefit from SGLT2 inhibitors and aggressive neurohormonal blockade;
- design intensified follow-up strategies (more frequent monitoring of renal function and biomarkers) for genetically high-risk individuals.

Given the relatively modest sample size and single-centre design, our findings require validation in larger, multi-ethnic cohorts with standardized genotyping and longer follow-up, as well as mechanistic studies dissecting gene–drug interactions.

5. Conclusions

1. Patients with CHF and CRS exhibit markedly elevated TGF- β 1, aldosterone and collagen IV levels and more severe renal and cardiac dysfunction compared with CHF without CRS and healthy controls.
2. Functional polymorphisms of TGF- β 1 and CYP11B2 significantly influence biomarker levels, susceptibility to CRS and severity of organ involvement.
3. The clinical and biochemical efficacy of SGLT2 inhibition with dapagliflozin is genotype-dependent, with the greatest benefit observed in carriers of “protective” TGF- β 1 and CYP11B2 variants.
4. Combined profiling of TGF- β 1, aldosterone, collagen IV and their genotypes represents a promising

approach for risk stratification and personalized therapy in chronic heart failure with cardiorenal syndrome.

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