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ORIGINAL RESEARCH

HEART FAILURE AND CARDIOMYOPATHIES

Renal Biomarkers in Heart Failure

Systematic Review and Meta-Analysis

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ABSTRACT

BACKGROUND Cystatin C, neutrophil gelatinase-associated lipocalin (NGAL), and kidney injury molecule (KIM)-1 are renal biomarkers increasingly appreciated for their role in the risk stratification and prognostication of heart failure (HF) patients. However, very few have been adopted clinically, owing to the lack of consistency.

OBJECTIVES The authors aimed to study the association between cystatin C, NGAL, and KIM-1 and outcomes, mortality, hospitalizations, and worsening renal function (WRF) in patients with acute and chronic HF.

METHODS We included peer-reviewed English-language articles from PubMed and EMBASE published up to December 2021. We analyzed the above associations using random-effects meta-analysis. Publication bias was assessed using funnel plots.

RESULTS Among 2,631 articles, 100 articles, including 45,428 patients, met the inclusion criteria. Top-tertile of serum cystatin C, when compared to the bottom-tertile, carried a higher pooled hazard ratio (pHR) for mortality (pHR: 1.59, 95% CI: 1.42-1.77) and for the composite outcome of mortality and HF hospitalizations (pHR: 1.49, 95% CI: 1.23-1.75). Top-tertile of serum NGAL had a higher hazard for mortality (pHR: 2.91, 95% CI: 1.49-5.67) and composite outcome (HR: 4.11, 95% CI: 2.69-6.30). Serum and urine NGAL were significantly associated with WRF, with pHRs of 2.40 (95% CI: 1.48-3.90) and 2.01 (95% CI: 1.21-3.35). Urine KIM-1 was significantly associated with WRF (pHR: 1.60, 95% CI: 1.24-2.07) but not with other outcomes. High heterogeneity was noted between studies without an obvious explanation based on meta-regression.

CONCLUSIONS Serum cystatin C and serum NGAL are independent predictors of adverse outcomes in HF. Serum and urine NGAL are important predictors of WRF in HF. (JACC Adv 2024;3:100765) © 2024 The Authors. Published by Elsevier on behalf of the American College of Cardiology Foundation. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

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The authors attest they are in compliance with human studies committees and animal welfare regulations of the authors' institutions and Food and Drug Administration guidelines, including patient consent where appropriate. For more information, visit the Author Center.

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ABBREVIATIONS AND ACRONYMS

GFR = glomerular function rate

HF = heart failure

KIM = kidney injury molecule

LVEF = left ventricular ejection function

NGAL = neutrophil gelatinase-associated lipocalin

WRF = worsening renal failure

Heart failure (HF) is a growing healthcare issue contributing to significant morbidity, mortality, and cost of care.[1] By the year 2030, it is estimated that more than 8 million American adults will be affected by HF.[2] Renal dysfunction is a common accompaniment and an independent predictor of poor clinical outcomes in patients with HF.[3] Renal dysfunction in HF can present as acute kidney injury, worsening renal function (WRF), or as a gradual decline leading to chronic kidney disease.[4] It has been noted that even minor variations in cardiac output can significantly affect renal perfusion by decreasing renal blood flow and increasing renal venous pressure.[5] These hemodynamic changes, in turn, activate neuro-hormonal, sympathetic, oxidative, and inflammatory pathways leading to altered glomerular structure and function, tubulointerstitial damage, injury, and permanent loss of nephrons.[6,7] In addition, diagnostic and therapeutic interventions used in these patients can adversely affect renal function.[8] Conversely, renal disease and the resultant fluid and electrolyte imbalances, anemia, and proteinuria contribute to the development and progression of HF.[9] Hence, early identification of renal dysfunction can aid in risk stratification and prognostication of patients with HF.[10]

Appreciation for renal biomarkers in HF has grown remarkably in the last decade, yet very few have been adopted in routine clinical practice.[11] Serum creatinine, a breakdown product of skeletal muscle creatinine phosphate, is freely filtered through the glomerulus enabling glomerular filtration rate (GFR) estimation.[12] While serum creatinine is commonly used to assess renal function, several factors such as active tubular secretion, high interindividual variation in creatinine phosphate production, and an exponential relationship with renal function make its GFR estimation unreliable.[9] On the contrary, cystatin C, a cysteine protease inhibitor produced by all nucleated cells, is freely filtered through the glomerulus without any active secretion and is unaltered by age, sex, or body mass, thereby allowing a more sensitive and fairly accurate measurement of GFR.[13] Beyond its role in GFR estimation, cystatin C has also been associated with adverse cardiovascular events in various patient populations.[14]

It has been hypothesized that, in patients with HF, renal dysfunction secondary to tubular injury occurs without the loss of excretory function (or decline in GFR) and can have prognostic implications.[15] Neutrophil gelatinase-associated lipocalin (NGAL) is a protein, upregulated in neutrophils and tubular epithelial cells in response to renal ischemia.[16] Likewise, kidney injury marker (KIM)-1 is a transmembrane protein expressed by injured (ischemic or nephrotoxic) proximal tubules.[17] Both NGAL and KIM-1 predict acute tubular injury 48 to 72 hours before a rise in serum creatinine and have been shown to be more sensitive and specific.[18,19] The present meta-analysis aims to collate the current available evidence on the association of the above renal biomarkers with important outcomes in HF patients, such as WRF, HF hospitalizations, and mortality.

METHODS

SEARCH STRATEGY AND STUDY SELECTION. The systematic review and meta-analysis was conducted and reported according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines,[20] using the Covidence platform.[21] The protocol, although not preregistered, can be found in the Supplemental Appendix. We searched for eligible studies in PubMed and Embase from inception to December 21, 2021, using the search strategy noted in Supplemental Table 1. Additionally, we reviewed bibliographies of the included studies and relevant review articles to identify potentially eligible studies. Inclusion was limited to articles published in the English language.

We included cohort studies, case-control studies, and secondary data analyses of randomized controlled trials that met the following criteria: 1) the study included patients with HF defined using a combination of signs, symptoms, imaging, and laboratory parameters suggestive of HF or with an established diagnosis of HF in medical records; 2) the study examined the association of at least one of following biomarkers, namely serum cystatin C, serum NGAL, urine NGAL, serum KIM-1 or urine KIM-1 with the outcomes of interest; 3) the study assessed the following outcomes: WRF, all-cause mortality, or a composite outcome of all-cause mortality and HF hospitalizations. WRF was defined as any in-hospital rise in serum creatinine ≥0.3 mg/dL (26.5 mmol/L) or ≥25% increase from admission serum creatinine value.[4] For assessing the association with WRF, the renal biomarkers measured at hospital admission were considered.

LITERATURE SCREENING AND DATA EXTRACTION. All the studies were imported to the Covidence platform for screening and data extraction. At least 2 authors (A.K., H.S.G., and M.G.M.) independently reviewed the title and abstracts of the imported studies for eligibility. Then, the studies were

screened for full-text eligibility. Discrepancies, if any, were resolved by a third author (V.C.). This was followed by data extraction by atleast 2 independent reviewers (A.K., H.S.G., and M.G.M.) using a predesigned questionnaire in the Qualtrics platform, and discrepancies were cleared by a third author (V.C.). Data on study characteristics, sample size, biomarkers assessed, time points of biomarker collection, outcome definitions, and follow-up periods were collected. In addition, the baseline patient characteristics, including demographics, comorbidities, baseline blood pressure, heart rate, laboratory parameters, left ventricular ejection fraction (LVEF %), and medications were collected. The Quality in Prognosis Studies tool[22] was used to assess the risk of bias and quality of the included studies. The tool estimated the risk of bias through the 6 key domains: study participation, study attrition, prognostic factor measurement, outcome measurement, adjustment for other prognostic factors, and statistical analysis.[22]

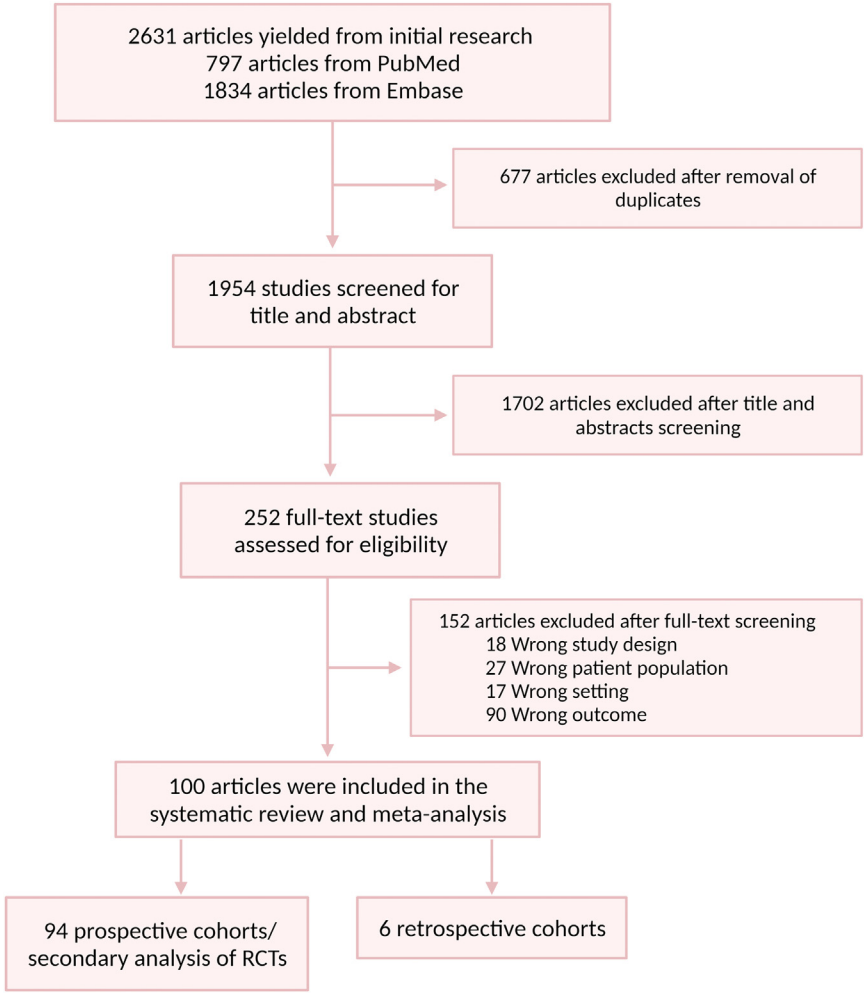
STATISTICAL ANALYSIS. We pooled the study-specific estimates using random effects meta-analysis. The HRs of the individual included studies were reported in different ways, such as HR per unit increase, 1 SD increase, 1 log change, 1 log SD change, or HR above and below a cut-off point. To enable a consistent approach to analysis and to allow for direct comparison and combination of the effect sizes of studies, the HRs from the studies were transformed to correspond to the HR for the top tertile with reference to the bottom tertile[23-25] of the biomarkers (cystatin C, NGAL, or KIM-1) for each of the outcomes (mortality, composite outcome, or WRF). We assumed that the biomarkers were log-normally distributed and that the association between the exposure and the outcome was log-linear. For a normal distribution, the difference between the means of the top and bottom tertile is 2.18 SD units. Thus, the scaling factor of 2.18 was used to transform log-risk estimates per SD to the log-risk estimates between the top and bottom tertiles.[23-25] The methodology is further detailed in the systematic review by Hemingway et al.[26] We pooled adjusted and unadjusted effect sizes separately. If multiple adjusted effect sizes were available, we used the 1 adjusted for most variables. We additionally pooled the mean difference (MD) of the biomarkers in patients with or without the above outcomes in separate analyses. We also performed random-effects meta-analyses of the c-statistics for the biomarkers with each outcome of interest. We assessed the between-study heterogeneity using the I^2 statistic. We performed meta-regression analysis using study-specific characteristics to assess statistical heterogeneity. We performed subgroup analysis using meta-regression for categorical study-specific characteristics such as geographic location based on the continent, acute or chronic HF, studies published before and after the year 2015, and adjustment for baseline serum creatinine. For continuous study-specific characteristics such as age, LVEF, serum creatinine, and proportions of female sex, diabetes mellitus, and hypertension, we fitted a meta-regression line for each biomarker and the outcome of interest. Publication bias for each outcome was inspected using a funnel plot, and Egger's test was performed if more than ten studies were present. If publication bias was detected, we followed the trim and fill procedure,[27] which calculates estimates adjusted for possible publication bias by imputing missing studies. All statistical analyses were performed using Stata version 16 (StataCorp),[28] and a 2-sided P value of <0.05 was deemed to represent statistical significance.

RESULTS

LITERATURE SEARCH, STUDY CHARACTERISTICS, AND QUALITY ASSESSMENT. We identified 1,954 distinct potentially relevant studies from 2 databases (PubMed and Embase) using our search strategy available in Supplemental Table 1. Of these, 1,702 studies were removed in the title and abstract review, and 152 were removed during the full-text review for reasons mentioned in the study selection flowchart (**Figure 1**). A total of 100 studies involving 45,428 patients were included in the systematic review and meta-analysis. Six included studies were retrospective, and the remaining 94 were prospective cohorts or cohorts nested within randomized controlled trials. In terms of biomarkers, serum cystatin C was assessed by 58 studies, serum NGAL by 30, urine NGAL by 25, serum KIM-1 by 2, and urine KIM-1 by 12 studies. With respect to clinical outcomes, 45, 44, and 36 studies reported data on all-cause mortality, composite outcome (all-cause mortality and HF readmissions), and WRF, respectively. Sixty-six studies included patients with acute HF, while the remaining 34 assessed patients with chronic HF. Thirteen studies were conducted in the Americas, 47 in Europe, 17 in Asia, and 23 across multiple continents. Further descriptions of the study characteristics, patient populations, biomarkers, and outcomes of the included studies are outlined in Supplemental Tables 2 and 3.

The risk of bias and quality assessment of the included studies using the Quality in Prognosis Studies tool is shown in Supplemental Table 4. Low

FIGURE 1 Flowchart for Study Selection



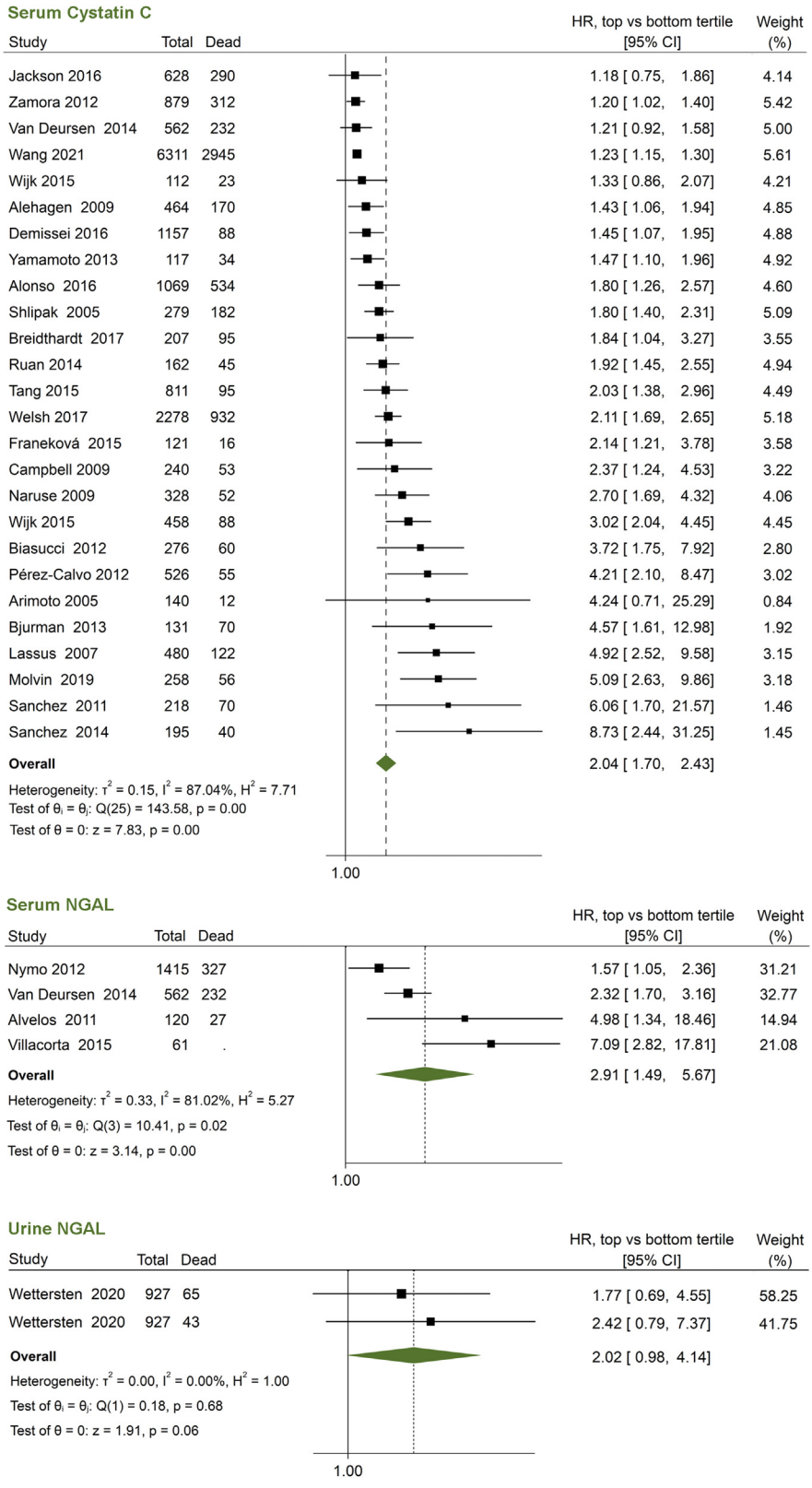
RCT = randomized controlled trial.

risk-of-bias scores were found in the majority of the studies for the domains of study participation, study attrition, and outcome measurement. Moderate risk of bias was found in most of the studies with respect to the domains of prognostic factor measurement and statistical analysis. Many studies have not adjusted adequately for the confounders; thus, for the domain of study confounding, a high risk of bias was detected in 26% of studies and moderate risk of bias in 23% of studies. The level of adjustment for confounders in each of the studies is detailed in Supplemental Tables 5 and 6.

ASSOCIATION OF SERUM CYSTATIN C WITH HF OUTCOMES. The pooled adjusted HR for all-cause mortality and the composite outcome for the top-tertile of serum cystatin C when compared with the bottom-tertile were 2.04 (95% CI: 1.70-2.43) ($I^2 = 87\%$) among 26 studies and 2.26 (95% CI: 1.73-2.96) ($I^2 = 90\%$) among 17 studies, respectively (**Figures 2 and 3**, **Table 1**). The adjusted HR for WRF comparing the top-tertile of serum cystatin to the bottom-tertile was 1.30 (95% CI: 0.54-3.14) in 1 study (**Table 1**).

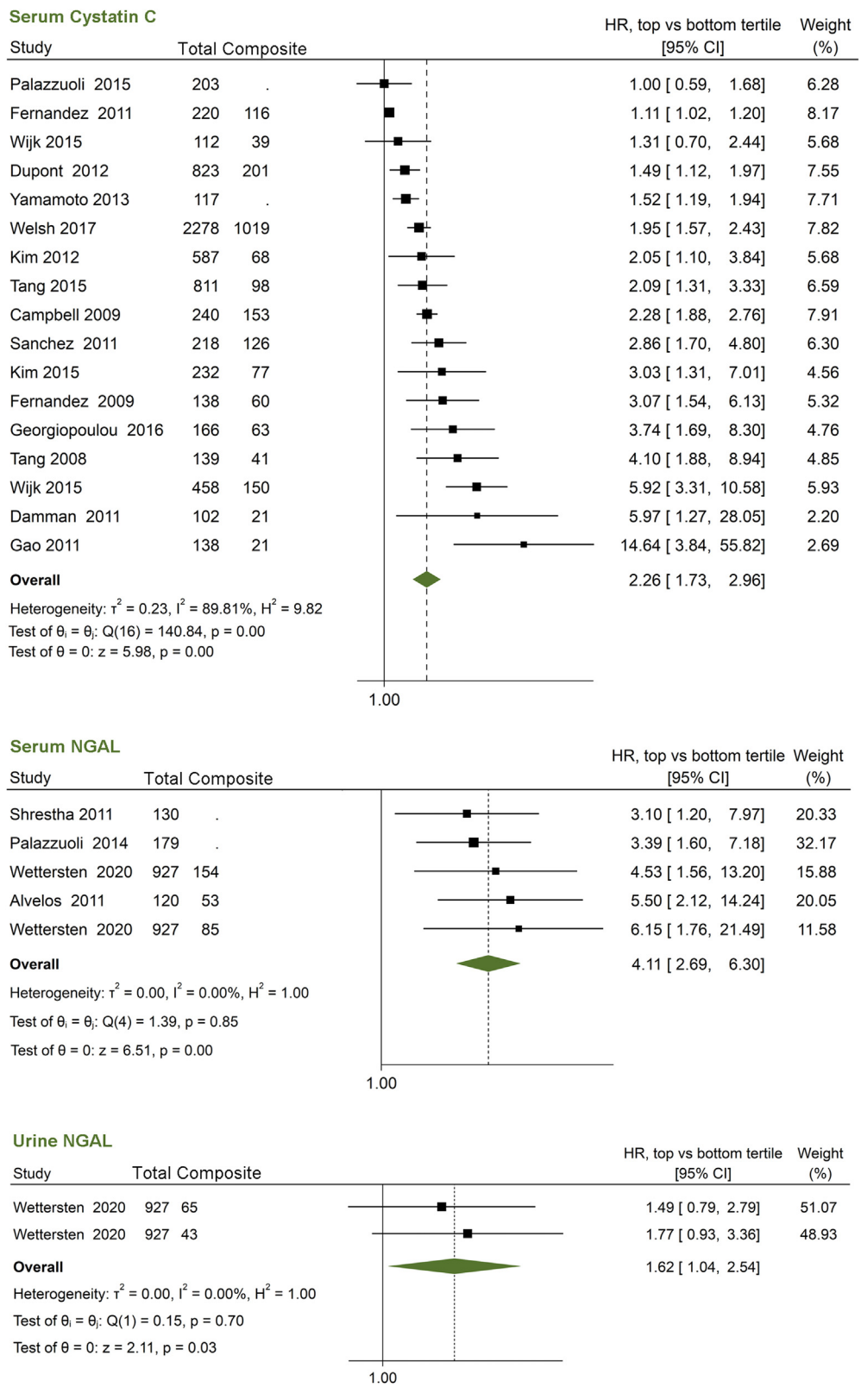
The pooled MD comparing serum cystatin (mg/dL) levels between patients who died and survived was 0.40 (95% CI: 0.31-0.48) ($I^2 = 76\%$) from 14 studies. Likewise, patients with a composite outcome had significantly higher serum cystatin levels when compared to those who did not (pooled mean difference [pMD] 0.33, 95% CI: 0.25-0.41) ($I^2 = 72\%$) from 12 studies (Supplemental Figures 1 and 2, **Table 2**). The pooled MD of serum cystatin C (mg/dL) between

FIGURE 2 **Forest Plots of Pooled HR for Serum Cystatin C, Serum NGAL, and Urine NGAL for All-Cause Mortality in Patients With Heart Failure**



NGAL = neutrophil gelatinase-associated lipocalin.

FIGURE 3 Forest Plots of Pooled HR for Serum Cystatin C, Serum NGAL, and Urine NGAL for the Composite Outcome of All-Cause Mortality and Hospitalizations in Patients With Heart Failure



NGAL = neutrophil gelatinase-associated lipocalin.

TABLE 1 Association of Renal Biomarkers With All-Cause Mortality, Composite Outcome, and Worsening Renal Function Using Random-Effects Meta-Analysis of HRs

		Pooled HR*					
Outcome	Biomarkers	No. of Studies	Unadjusted HR (95% CI)	I^2 Statistic	No. of Studies	Adjusted HR (95% CI)	I^2 Statistic
All-cause mortality	Serum Cystatin C	22	2.61 (2.16-3.16)	92.49%	26	2.04 (1.70-2.43)†	87.0%
	Serum NGAL	4	2.95 (1.09-7.96)	96.33%	4	2.91 (1.49-5.67)†	81.0%
	Urine NGAL	3	2.10 (1.36-3.25)	1.49%	2	2.02 (0.98-4.14)	0%
	Serum KIM-1	2	1.59 (1.19-2.11)	0%	2	1.31 (0.80-2.13)	58.3%
	Urine KIM-1	1	1.74 (0.96-3.22)	-	1	1.33 (0.98-1.80)	-
Composite outcome	Serum Cystatin C	18	2.41 (1.82-3.18)	97.09%	17	2.26 (1.73-2.96)†	89.8%
	Serum NGAL	5	2.91 (1.44-5.90)	82.35%	5	4.11 (2.69-6.30)†	0%
	Urine NGAL	2	2.12 (1.35-3.34)	0%	2	1.62 (1.04-2.54)†	0%
	Serum KIM-1	3	2.09 (0.57-7.58)	80.86%	2	1.61 (0.61-4.22)	0%
	Urine KIM-1	3	2.16 (1.80-2.59)	0%	2	1.22 (0.84-1.77)	84.1%
Worsening renal function	Serum Cystatin C	1	3.25 (1.69-6.25)	-	1	1.30 (0.54-3.14)	-
	Serum NGAL	10	3.86 (2.40-6.21)	61.31%	7	2.40 (1.48-3.90)†	73.6%
	Urine NGAL	7	2.43 (1.48-3.99)	89.94%	6	2.01 (1.21-3.35)†	64.6%
	Serum KIM-1	0	-	-	0	-	-
	Urine KIM-1	2	2.22 (1.77-2.80)	1.41%	2	1.60 (1.24-2.07)†	0%

*The pooled HRs represent the random-effects meta-analysis of HRs from the studies to represent the HR between the top and bottom tertile of the biomarkers for each outcome. †Refers to statistical significance ($P < 0.05$).

NGAL = neutrophil gelatinase-associated lipocalin; KIM = kidney injury molecule.

patients who developed WRF vs those who did not was 0.37 (95% CI: -0.01 to 0.74) (I^2 = 98%) from 6 studies (Figure 4, Table 2).

ASSOCIATION OF SERUM AND URINE NGAL WITH HF OUTCOMES. Patients in the top-tertile of serum NGAL had significantly higher pooled hazards of all-cause mortality (HR: 2.91, 95% CI: 1.49-5.67) (I^2 = 81%) and composite outcome (HR: 4.11, 95% CI: 2.69-6.30) (I^2 = 0%) when compared to the patients in the bottom-tertile (Figures 2 and 3, Table 1). The pooled adjusted HR for WRF comparing the top-tertile of NGAL to the bottom-tertile was 2.40 (95% CI: 1.48-3.90) (I^2 = 74%) among 7 studies (Table 1). Likewise, serum NGAL (ng/mL) was significantly higher among patients who died (pMD 38.65,

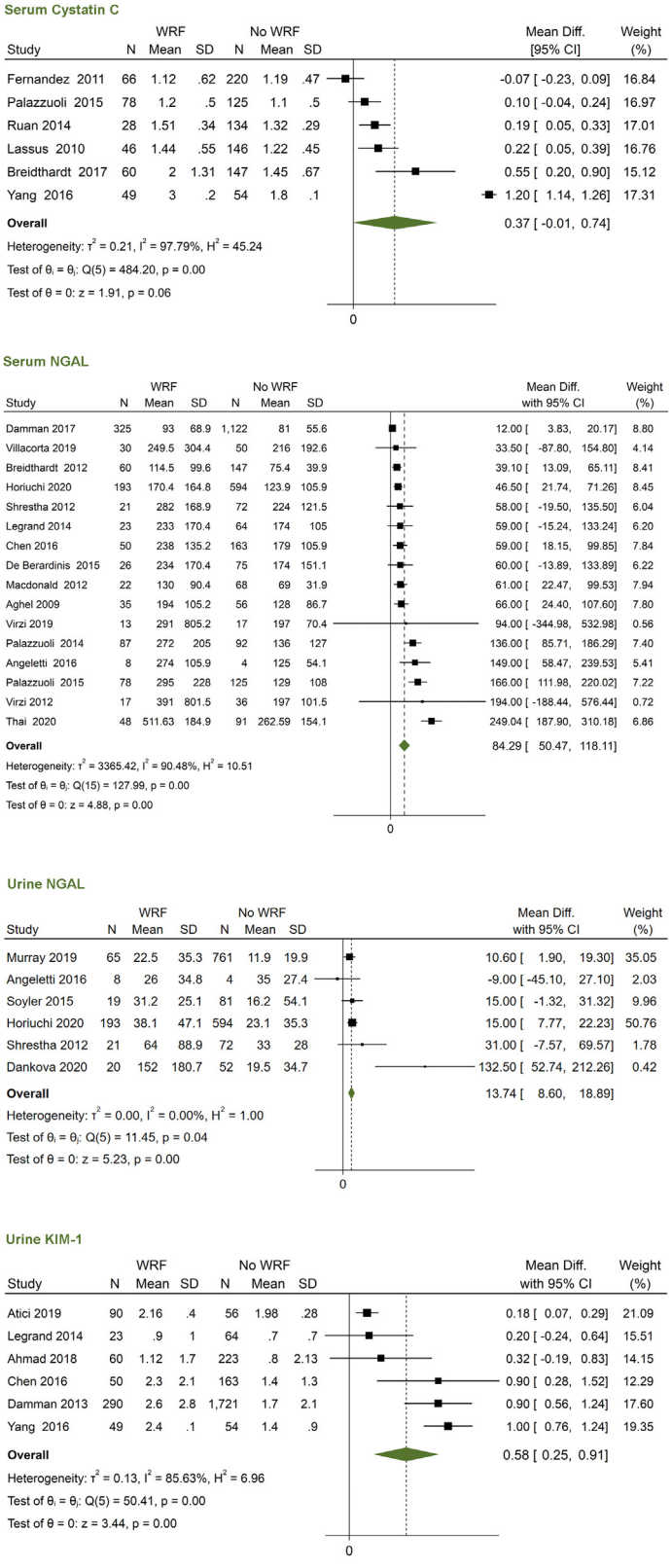
TABLE 2 Association of Renal Biomarkers With All-Cause Mortality, Composite Outcome, and Worsening Renal Function Using Random-Effects Meta-Analysis of Mean Difference of the Biomarkers in Patients With and Without the Outcome

		Pooled Mean Difference*			
Outcome	Biomarkers	No. of Studies	Mean Difference (95% CI)	*P* Value	I^2 Statistic
All-cause mortality	Serum Cystatin C (mg/L)	14	0.40 (0.31-0.48)†	<0.001	75.9%
	Serum NGAL (ng/mL)	4	38.65 (26.09-51.22)†	<0.001	17.7%
	Urine NGAL (ng/mL)	0	-	-	-
	Serum KIM-1 (pg/mL)	0	-	-	-
	Urine KIM-1 (ng/gCr)	0	-	-	-
Composite outcome	Serum Cystatin C (mg/L)	12	0.33 (0.25-0.41)†	<0.001	71.6%
	Serum NGAL (ng/mL)	6	44.60 (12.98-76.21)†	0.010	75.9%
	Urine NGAL (ng/mL)	3	7.86 (−2.05-17.76)	0.120	66.8%
	Serum KIM-1 (pg/mL)	0	-	-	-
	Urine KIM-1 (ng/gCr)	2	448.02 (−198.76-1094.8)	0.170	95.8%
Worsening renal function	Serum Cystatin C (mg/L)	6	0.37 (−0.01-0.74)	0.060	97.8%
	Serum NGAL (ng/mL)	16	84.29 (50.47-118.11)†	<0.001	90.5%
	Urine NGAL (ng/mL)	6	13.74 (8.60-18.89)†	<0.001	0%
	Serum KIM-1 (pg/mL)	0	-	-	-
	Urine KIM-1 (ng/gCr)	6	0.58 (0.25-0.91)†	<0.001	85.6%

*The pooled HRs represent the random-effects meta-analysis of mean difference (MD) from the studies to represent the HR between the top and bottom tertile of the biomarkers for each outcome. †Refers to statistical significance ($P < 0.05$).

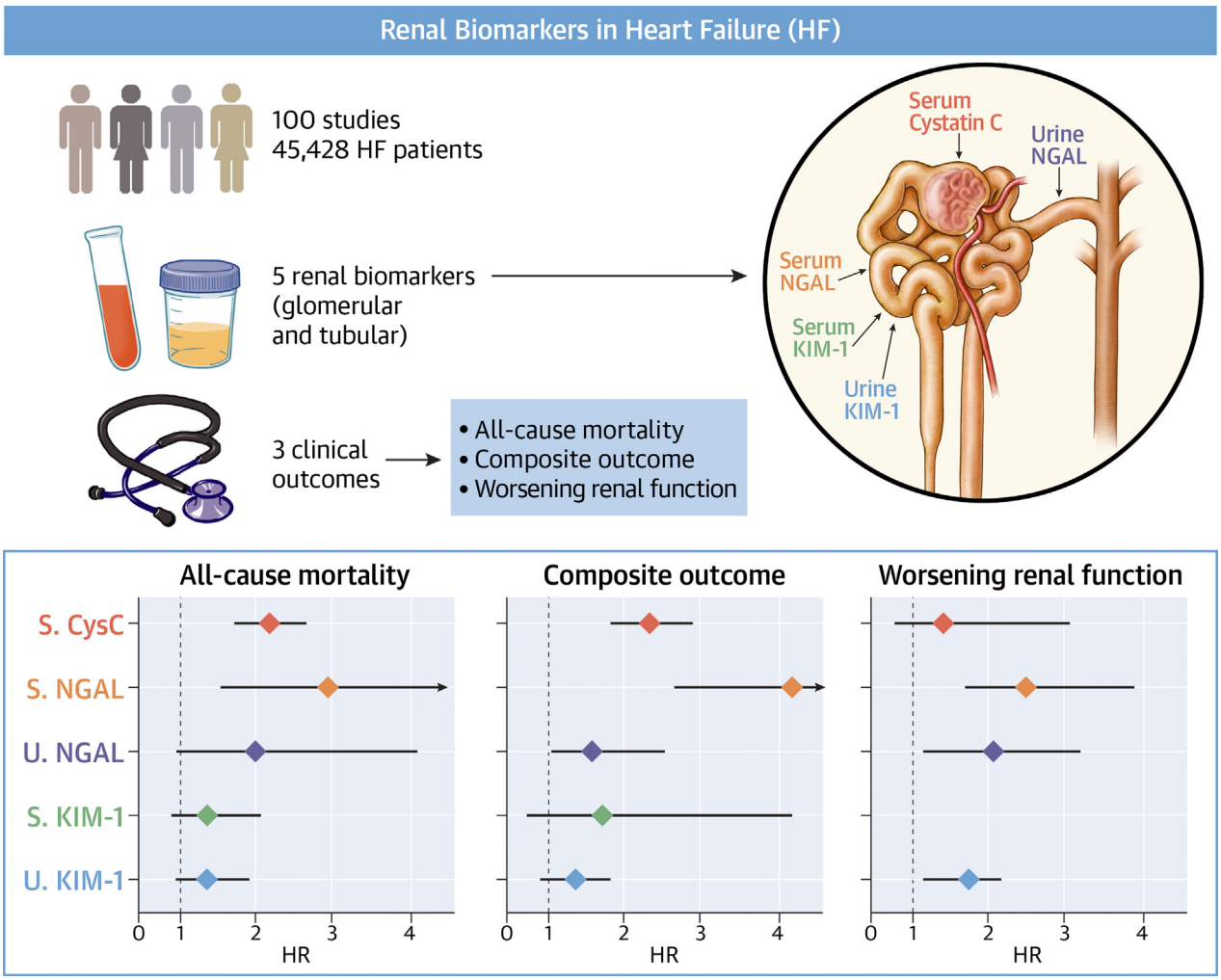
NGAL = neutrophil gelatinase-associated lipocalin; KIM = kidney injury molecule.

FIGURE 4 Forest Plots of Pooled Mean Difference for Serum Cystatin C, Serum NGAL, Urine NGAL, and Urine KIM-1 for Worsening Renal Function in Patients With Heart Failure



KIM = kidney injury molecule; NGAL = neutrophil gelatinase-associated lipocalin.

CENTRAL ILLUSTRATION **Association of Renal Biomarkers With Clinical Outcomes in Patients With Heart Failure**



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CysC = cystatin C; KIM = kidney injury molecule; NGAL = neutrophil gelatinase-associated lipocalin; S. = serum; U. = urine.

95% CI: 26.09-51.22) (I^2 = 18%), who had a composite outcome (pMD 44.60, 95% CI: 12.98-76.21) (I^2 = 76%) (Supplemental Figures 3 and 4, **Table 2**) and who developed WRF (pMD 84.29, 95% CI: 50.47-118.11) (I^2 = 90%) (**Figure 4**, **Table 2**).

The pooled adjusted HR comparing the top-tertile to the bottom-tertile of urine NGAL for all-cause mortality was 2.02 (95% CI: 0.98-4.14) (I^2 = 0%) and for composite outcome was 1.62 (95% CI: 1.04-2.54) (I^2 = 0%) (**Figure 2 and 3**, **Table 1**). Among 7 studies, patients in the top tertile of urine NGAL had a significantly higher hazard for WRF (HR: 2.01, 95% CI: 1.21-3.35) (I^2 = 65%) (**Table 1**). The studies assessing MD showed a similar association between urine NGAL (ng/mL) and HF outcomes (**Figure 4**, **Table 2**).

ASSOCIATION OF SERUM AND URINE KIM-1 WITH HF OUTCOMES. The pooled adjusted HR comparing the top-tertile to the bottom-tertile of serum KIM-1 for all-cause mortality was 1.31 (95% CI: 0.80-2.13) (I^2 = 58%), and for composite outcome, it was 1.61 (95% CI: 0.61-4.22) (I^2 = 0%) (**Table 1**). We did not find any studies assessing the effect of serum KIM-1 on the development of WRF. With reference to urinary KIM-1, 1 study reported data on adjusted hazards for all-cause mortality (HR: 1.33, 95% CI: 0.98-1.80) and

2 studies for the composite outcome (HR: 1.22, 95% CI: 0.84-1.77) (I^2 = 84%) (Table 1). On the contrary, patients in the top-tertile of urinary KIM-1 had a significantly higher hazard for the development of WRF when compared to the bottom-tertile (HR: 1.60, 95% CI: 1.24-2.07) (I^2 = 0%) (Table 1). The studies assessing MD showed a similar association between urine KIM-1 (ng/gCr) and HF outcomes (Figure 4, Table 2).

SUBGROUP ANALYSIS AND META-REGRESSION. Subgroup analysis based on the continent of study, study population (acute or chronic HF), year of publication, biochemical assay manufacturer, and adjustment for serum creatinine are described in Table 3. None of the above subgroup analyses were statistically significant, suggesting that the association of serum cystatin C with all-cause mortality and composite outcome and the association of serum NGAL with WRF remained the same across the subgroups. Additionally, meta-regression analysis showed that none of the continuous study-level variables were associated with the HR for the association between serum cystatin C and NGAL and HF outcomes (Supplemental Table 7).

META-ANALYSIS OF C-STATISTICS. Using random-effects meta-analysis, pooled C-statistics for serum cystatin C were 0.76 (0.74-0.78) for all-cause mortality, 0.68 (0.64-0.71) for the composite outcome, and 0.66 (0.55-0.76) for WRF. The pooled C-statistics for serum NGAL were 0.64 (0.58-0.69) for all-cause mortality, 0.66 (0.58-0.73) for the composite outcome, and 0.71 (0.65-0.76) for WRF (Supplemental Figures 5 to 13, Supplemental Table 8).

PUBLICATION BIAS. The Egger tests indicated asymmetry or small-study effects for the HR for the association of serum cystatin C with all-cause mortality ($P < 0.001$) and composite outcome ($P < 0.001$). Trim and fill analysis was performed, which yielded corrected HRs of 1.73 (95% CI: 1.40-2.15) for all-cause mortality and 1.80 (95% CI: 1.31-2.48) for the composite outcome. The other associations showed no evidence of publication bias. The funnel plots and the trim and fill imputed plots are provided in the Supplemental Figures 14 to 18.

DISCUSSION

The present systematic review and meta-analysis comprising 100 studies and 45,428 participants demonstrates a strong association between serum cystatin C levels and higher all-cause mortality and HF hospitalizations. Serum NGAL, urine NGAL, and KIM-1 at baseline were strong predictors of WRF. We also found that serum NGAL, but not urine NGAL and urine KIM-1, was associated with all-cause mortality and composite endpoints (Central Illustration).

Since its introduction in 1985, serum cystatin C has been shown to be an accurate marker of GFR in various patient populations.[29] It is well established that renal dysfunction is a strong predictor of adverse outcomes in patients with HF.[3] This aligns with our results that serum cystatin C is a predictor of all-cause mortality and composite outcome in patients with HF. This association was consistent even in the pooled creatinine-adjusted HR for mortality and composite outcome for cystatin C (Table 3). While it is possible that serum cystatin C's ability to estimate renal function better than serum creatinine contributed to its independent predictive ability, mechanisms unrelated to renal function cannot be discounted. For instance, cystatin C has been hypothesized to play a key role in vascular biology,[30] and serum cystatin C levels have been independently associated with increased risk of coronary artery disease, ischemic stroke, and HF.[31] However, serum cystatin C failed to predict WRF in patients admitted for HF in our meta-analysis. One of the key explanations is that WRF, defined based on serum creatinine rise in-hospital, is not necessarily a result of loss of excretory function.[15] Renal dysfunction in patients admitted for HF is often a result of a tubular injury caused by reduced renal blood flow secondary to volume overload, decreased intravascular volume, and nephrotoxins.[15,32] In addition, extra-renal factors such as hemoconcentration, initiation of renin-angiotensin-aldosterone system inhibitors, and concurrent illness could contribute to the creatinine rise.[4,32]

This concept of tubular injury contributing to WRF is further supported by our results that both serum and urine NGAL measured at admission were strong predictors of WRF in patients admitted for HF. After its initial discovery through genomic and proteomic analyses, NGAL has been studied in various animal and clinical models as a marker of tubular injury.[16,19] NGAL has been hypothesized to aid in epithelization and regeneration of tubular cells, providing biological plausibility for its induction after tubular injury.[33] Both serum and urine NGAL rise within hours after the tubular insult and rapidly decrease with resolution of the injury.[19] Early and accurate identification of tubular injury in HF patients will aid in risk stratification, thereby allowing reno-protective strategies for high-risk patients with tubular injury while avoiding unnecessary suspension of beneficial medical therapy in low-risk patients without tubular injury.[34] With respect to mortality and

TABLE 3 Meta-Regression of Categorical Study-Level Characteristics

Outcome	Biomarker	Study-Level Characteristic	Subgroup	No. of Studies	pHR (95% CI)	I^2 Statistic	*P* Value
All-cause mortality	Serum Cystatin-C	Continent	Asia	4	1.94 (1.42-2.64)	52.7%	0.605
			Europe	12	2.43 (1.70-3.47)	86.7%	
			North America	2	1.87 (1.48-2.35)	0%	
			Multicontinent	6	1.78 (1.25-2.52)	90.9%	
		Study population	Acute HF	16	2.36 (1.76-3.16)	88.5%	0.109
			Chronic HF	10	1.77 (1.46-2.15)	74.2%	
		Year of publication	Before 2015	15	2.32 (1.74-3.11)	86.2%	0.170
			After 2015	9	1.79 (1.41-3.26)	81.9%	
		Assay manufacturer	Abbott	5	1.72 (1.35-2.21)	51.7%	0.060
			Roche	4	2.47 (1.49-4.09)	84.8%	
			Siemens	6	2.96 (1.96-4.45)	62.0%	
			Dako	3	2.94 (1.24-6.95)	81.9%	
			Radim	2	1.42 (0.96-2.21)	76.5%	
		Creatinine adjustment	Creatinine adjusted	13	2.11 (1.59-2.80)	90.7%	0.794
			Creatinine unadjusted	13	2.01 (1.58-2.56)	79.1%	
		Duration of follow-up	≤12 mo	8	2.76 (1.87-4.09)	77.4%	0.069
			>12 mo	18	1.82 (1.51-2.20)	86.5%	
Composite outcome	Serum Cystatin-C	Continent	Asia	4	2.88 (1.27-6.51)	84.1%	0.727
			Europe	5	1.91 (1.06-3.43)	85.9%	
			North America	4	2.39 (1.53-3.73)	79.9%	
			Multicontinent	2	1.98 (1.63-2.41)	0%	
		Study population	Acute HF	8	1.92 (1.40-2.63)	85.9%	0.195
			Chronic HF	9	2.80 (1.74-4.49)	89.6%	
		Year of publication	Before 2015	10	2.29 (1.61-3.26)	91.5%	0.619
			After 2015	5	2.00 (1.35-2.97)	67.5%	
		Assay manufacturer	Abbott	3	2.02 (1.28-3.2)	64.0%	0.926
			Roche	3	2.46 (1.05-5.75)	89.9%	
			Siemens	8	2.10 (4.43-3.09)	89.4%	
		Creatinine adjustment	Creatinine adjusted	8	2.56 (1.71-3.81)	97.7%	0.437
			Creatinine unadjusted	9	2.05 (1.40-3.01)	81.6%	
		Duration of follow-up	≤12 mo	5	2.09 (1.47-2.98)	69.2%	0.678
			>12 mo	12	2.44 (1.67-3.56)	92.5%	
WRF	Serum NGAL (ng/mL)	Continent	Asia	3	121.0 (−0.51 to 242.5)	95.3%	0.105*
			Europe	7	106.57 (56.73-156.40)	76.6%	
			North America	1	58.0 (−19.5 to 135.5)	-	
			South America	1	33.5 (−87.8 to 154.8)	-	
			Multicontinent	4	33.7 (6.35-61.14)	65.7%	
		Year of publication	Before 2015	7	67.71 (40.03-97.38)	52.8%	0.406*
			After 2015	9	84.29 (50.47-118.11)	94.1%	
		Assay manufacturer	Abbott	10	67.91 (33.74-102.08)		0.217*
			Bioporto	4	107.34 (16.17-198.52)		
			Siemens	1	149.0 (58.48-239.52)		
		Creatinine adjustment	Creatinine adjusted	9	74.18 (30.91-117.45)	87.2%	0.456*
			Creatinine unadjusted	7	101.75 (43.59-159.90)	87.1%	

*P value calculated using meta-regression.

HF = heart failure; NGAL = neutrophil gelatinase-associated lipoprotein; pHR = pooled hazard ratio; pMD = pooled mean difference; WRF = worsening renal function.

hospitalizations, we found a significant association with serum NGAL but not with urine NGAL. The potential explanations lie in the distinct pathophysiologic processes represented by the markers. Serum NGAL is produced by renal (proximal tubules) and extra-renal sources in response to renal tubular injury, inflammation, and sepsis.[35] Serum NGAL is a compensatory protective mechanism in these acute

states, but chronic elevations are maladaptive and associated with myocardial remodeling, fibrosis, and atherosclerotic plaque instability.[36] Hence, the association of serum NGAL with adverse clinical outcomes might reflect a systematic process beyond renal injury. On the contrary, urinary NGAL is produced by distal nephrons solely in response to local tubular injury.[35] Pertaining to the above hypothesis, Mori et al[37] showed that tagged NGAL injected systemically accumulates in the proximal tubule but does not appear in urine.

Unlike NGAL, KIM-1 is solely produced in the proximal tubular cells in response to acute and chronic insults.[17] KIM-1 expression aids in phagocytosis of apoptotic tubular epithelial cells, which decreases inflammation and promotes tissue repair.[38] Urine KIM-1 represents the shed ectodomains, while plasma KIM-1 is obtained as a leak from compromised renal microvasculature and correlates with urine levels.[39] We found that serum and urine KIM-1 were associated with WRF, aligning with its role as a sensitive and specific marker of tubular injury in previous research models. We did not find a significant association between serum or urinary KIM-1 and mortality or composite outcomes.

STUDY STRENGTHS. Our study has several strengths. Our systematic review examined the association of 3 renal biomarkers and clinical endpoints, such as mortality, hospitalizations, and WRF, in patients with HF. We included over 100 studies with 45,428 HF patients from 15 countries with diverse patient populations, disease characteristics, healthcare access, and expertise, which allows generalizability of our results. The study not only assesses the prognostic role of these biomarkers in HF but also enables us to understand the complex pathophysiological process, as reflected by our results. We further assessed the strength of our associations through meta-regression of characteristics such as age, sex, blood pressure, heart rate, LVEF, comorbidities such as diabetes mellitus and hypertension, and serum creatinine. Our subgroup analysis based on the year of study was able to account for significant advancements in HF management in recent years. We were able to show that the prognostic role of the biomarkers was similar across different biochemical assays.

STUDY LIMITATIONS. However, our results should be interpreted in the context of the following limitations. Most of the included studies had a single timepoint measurement, restricting our ability to assess the effect of biomarker changes over time on the outcomes, especially mortality and the composite outcome. Factors such as steroid use, hyperthyroidism, inflammatory states, sepsis, and malignancies can influence renal biomarker levels but were not adjusted for in many included studies. Significant heterogeneity was observed in our analyses, but no single culpable variable was identified from our meta-regression, and thus it was likely multifactorial. In addition, inconsistencies in the reporting of covariates across studies restricted our ability to perform meta-regression for many variables. In the meta-analysis of adjusted effect sizes, individual studies were adjusted for comparable but different parameters, thus preventing uniform pooling of the results. Though we pooled prognostic c-statistics in our study, unlike diagnostic c-statistics, they cannot be relied upon to assess a biomarker's clinical utility[40] Biomarkers with good risk prediction could still have low prognostic c-statistics. All studies of WRF were conducted in patients hospitalized with acute HF, whereas mortality and composite outcomes were extracted from both acute and chronic HF. We did not obtain individual patient-level data, which might help address the possible clinical and methodological heterogeneity of the included studies. The decision to assume a log-normal distribution was made based on literature precedents and the nature of biomarker data, which often exhibits right-skewed distributions. However, we acknowledge that this is an assumption, and without access to individual patient data from the original studies, it's a limitation of our approach.

CONCLUSIONS

Our results have several important clinical implications. Serum cystatin C and serum NGAL are strong and independent predictors of adverse clinical outcomes in patients with HF, thus enabling risk stratification and therapeutic decision-making. In addition, we showed that serum and urine NGAL are important predictors of WRF in HF patients admitted to the hospital, thus aiding in the identification of patients at risk of WRF using tubular injury markers. A multimarker model that will differentiate normal functional kidneys from reduced GFR, loss in glomerular integrity, and tubulointerstitial injury is imperative in managing patients with HF.

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PERSPECTIVES

COMPETENCY IN PATIENT CARE AND PROCEDURAL SKILLS: Early and accurate identification of renal dysfunction in HF patients will aid in risk stratification, thereby allowing reno-protective strategies for high-risk patients while avoiding unnecessary suspension of beneficial medical therapy in low-risk patients.

TRANSLATIONAL OUTLOOK: Further efforts are needed to evaluate a multibiomarker model that will differentiate normal functional kidneys from reduced GFR, loss in glomerular integrity, and tubulointerstitial injury in managing patients with HF.

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KEY WORDS cystatin C, KIM-1, NGAL, tubular injury, worsening renal function

APPENDIX For supplemental tables and figures, please see the online version of this paper.