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Galectin-3 Serum Levels Are Independently Associated With Microalbuminuria in Chronic Heart Failure Outpatients

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Background: Galectin-3 (Gal-3) is a novel biomarker reflecting inflammation status and fibrosis involving worsening of both cardiac and renal functions.
Objectives: The aim of this study was to evaluate the relationship between Gal-3 serum levels and microalbuminuria in a group of chronic heart failure (CHF) outpatients.
Patients and Methods: We enrolled CHF outpatients having stable clinical conditions and receiving conventional therapy. All patients underwent clinical evaluation, routine chemistry analysis, echocardiography, and evaluation of the urinary albumin/creatinine ratio (UACR).
Results: Among the patients enrolled, 61 had microalbuminuria (UACR, 30-299) and 133 normoalbuminuria (UACR, < 30). Patients with normoalbuminuria showed significantly higher levels of Gal-3 than those without (19.9 ± 8.8 vs. 14.6 ± 5.5 ng/mL). The stepwise regression analysis indicated that Gal-3 was the first determinant of microalbuminuria (odds ratio [OR]: 1.08; 95% confidence interval [CI]: 1.02 - 1.14, P = 0.012), followed by diabetes (OR 2.14; 95% CI: 1.00 - 4.57; P = 0.049) and high central venous pressure (OR 2.80; 95% CI: 1.04 - 7.58; P= 0.042).
Conclusions: Our findings indicate an independent association between Gal-3 levels and microalbuminuria, an early marker of altered renal function. This suggests the possible role of Gal-3 in the progression of cardiorenal syndrome in CHF outpatients.

Keywords: Galectin 3, Diabetic Nephropathies, Renal Insufficiency, Heart Failure

1. Background

Galectin-3 (Gal-3) is a beta-galactoside-binding lectin that is associated with cardiac fibrosis in experimental studies (1, 2). In humans, Gal-3 has been shown to be a prognostic marker in acute (3) as well as in chronic heart failure (CHF) (4). Moreover, in patients with or without HF the presence of renal dysfunction is associated with higher serum levels of Gal-3 (5). Higher levels of Gal-3 has been also found being (5), and higher serum levels of Gal-3 are associated with renal fibrosis (6) and a greater incidence of renal dysfunction (7). Nevertheless, whether high Gal-3 levels are the cause or consequence of renal impairment in CHF has not been well established.

2. Objectives

In order to better clarify the relationship between Gal-3 levels and chronic kidney disease (CKD), we evaluated the relationship between Gal-3 serum levels and the urinary albumin/creatinine ratio (UACR) (i.e., a marker of alteration of size and/or charge selectivity of the glomerular basement membrane (8) in a group of CHF outpatients).

3. Patients and Methods

We enrolled outpatients with CHF who referred to the Heart Failure Unit of the University of Bari. At the time of enrolment, we included patients who were clinically stable for at least 30 days and who had been taking conventional medical and electrical therapy for at least 3 months. Patients with acute decompensated heart failure, acute worsening of kidney function, renal failure requiring dialysis or transplantation, and macroalbuminuria were excluded

from the study. The protocol was approved by local ethical committee, and all patients gave their informed consent.

At the time of enrolment, all patients underwent a medical visit and electrocardiography. An echocardiographic evaluation was performed to evaluate left ventricular volumes and ejection fraction (LVEF), the presence of transmitral restrictive pattern, systolic peak of tricuspid annular plane excursion (TAPSE), central venous pressure (CVP), and pulmonary systolic artery pressure (PAPs), as previously described (9). Blood samples were obtained to evaluate levels of amino-terminal brain natriuretic peptide (NT-proBNP; immunoassay Dade Behring, Eschborn, Germany), serum electrolytes (mEq/L), and serum creatinine (mg/dL). The glomerular filtration rate was calculated using the abbreviated CKD-EPI formula (GFR-EPI, mL/minute/1.73 m^2). Gal-3 levels were measured from the plasma using an enzyme-linked immunosorbent assay (BG Medicine, Waltham, MA, USA). Normoalbuminuria, microalbuminuria, and macroalbuminuria were defined as a urinary albumin/creatinine ratio (UACR) of <30, 30 to 299, and ≥ 300 mg/g, respectively.

Continuous variables are expressed as mean ± standard deviation. Categorical variables are reported as frequencies and percentages. Univariate and stepwise multivariate logistic regression analyses were used to assess the association among studied variables and the presence of microalbuminuria. $P < 0.05$ was considered statistically significant. The analyses were made using STATA software, Version 12 (StataCorp, College Station, Texas).

4. Results

Of 205 patients, 11 (6%) were excluded because of the presence of macroalbuminuria. The remaining 194 patients (81% male, 64 ± 13 years, New York Heart Association (NYHA) class 2.3 ± 0.6, LVEF 33% ± 9%) were evaluated. Seven (4%) patients among these had CHF with preserved ejection fraction, 37% had ischemic cardiomyopathy, 28% had diabetes mellitus, 54% had arterial hypertension, and 16% had atrial fibrillation. The mean estimated glomerular filtration rate (eGFR EPI) was 73 ± 25 mL/minute/1.73 m^2, the mean NT proBNP value was 2030 ± 2881 pg/mL, and the mean Gal-3 level was 16.4 ± 7.1 ng/mL. Patients received conventional medical (angiotensin-converting-enzyme inhibitors and or angiotensin receptor blocker, 79%; betablockers, 96%; diuretics, 92%; aldosterone antagonist, 73%) or electrical therapy (86%, automatic cardioverter/defibrillator; 39%, cardiac resynchronization therapy device).

Table 1. Baseline Patients' Clinical and Therapeutic Characteristics According to the Presence or Absence of Microalbuminuria [a]

Variables	Microalbuminuria	Normoalbuminuria	Univariate Regression Analysis	
			OR, 95% CI	P
Number	61	133		
Age, y	68 ± 12	62 ± 13	1.04 (1.01 - 1.07)	0.004
Male gender [b]	82	79	1.21 (0.56 - 2.63)	0.628
Ischemic cardiomyopathy [b]	47	32	1.89 (1.02 - 3.53)	0.042
Diabetes [b]	39	23	2.23 (1.16 - 4.29)	<0.001
Hypertension [b]	52	63	1.03 (0.56 - 1.89)	0.927
BMI , kg/m²	27 ± 4	28 ± 5	0.94 (0.87 - 1.01)	0.089
Systolic pressure, mmHg	120 ± 18	121 ± 16	0.99 (0.97 - 1.01)	0.443
NYHA class	2.5 ± 0.5	2.2 ± 0.6	1.99 (1.17 - 3.39)	0.011
LVEDV	166 ± 65	157 ± 59	1.00 (0.99 - 1.01)	0.353
LVEF [b]	31 ± 10	33 ± 9	0.96 (0.93 - 0.99)	0.042
TAPSE, mm	18 ± 4	19 ± 4	0.95 (0.89 - 1.02)	0.181
PAPs, mmHg	40 ± 18	32 ± 10	1.04 (1.02 - 1.07)	0.001
CVP, mmHg	6.3 ± 5.3	3.9 ± 3.0	1.15 (1.06 - 1.23)	< 0.001
CVP > 5, mmHg [b]	26	8	4.37 (1.85 - 10.3)	< 0.001
GFR-EPI,mL/minute/1.73 m^2	62 ± 24	78 ± 23	0.97 (0.96 - 0.98)	< 0.001
NT-proBNP, pg/mL [c]	3637 ± 4166	1298 ± 1584	1.92 (1.44 - 2.55)	< 0.001
Gal-3, ng/mL	19.9 ± 8.8	14.6 ± 5.5	1.12 (1.07 - 1.18)	< 0.001

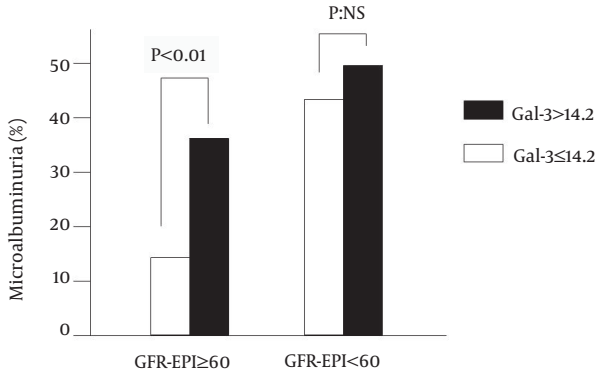
[a] Abbreviations: BMI, Body Mass Index; CVP, Central Venous Pressure; Gal-3, Galectin-3; GFR, Glomerular Filtration Rate; LVEDV, Left Ventricular End Diastolic Volume; LVEF, Left Ventricular Ejection Fraction; NT-proBNP, N-Terminal pro-Brain Natriuretic Peptide; PAPs, systolic peak of Pulmonary Arterial Pressure; TAPSE, peak of Tricuspid Annular Plane Systolic Excursion.
[b] The values are presented as %.
[c] Regression analyses performed after log-transformation.

One-hundred thirty-three of the enrolled patients (69%) had normal levels of urinary albumin excretion (8 ± 8 mg/g), whereas 61 (31%) had microalbuminuria (105 ± 72 mg/g). Table 1 shows the mean values of the studied variables in patients with and those without microalbuminuria. The univariate logistic regression analysis showed that age, ischemic cardiomyopathy, diabetes, NYHA class, LVEF, PAPs, CVP, GFR-EPI, logarithm of NTproBNP, and Gal-3 levels were significantly associated with microalbuminuria.

A stepwise multivariate logistic regression analysis including age, diabetes, ischemic cardiomyopathy, NYHA class, LVEF, CVP > 5 mm Hg, GFR-EPI, logNT-proBNP, and Gal 3, indicated that Gal-3 was the first variable that remained significantly associated with microalbuminuria (odds ratio [OR] 1.08, 95% confidence interval [CI]: 1.02 - 1.15, P = 0.012), followed by diabetes (OR 2.14; 95% CI: 1.00 - 4.57; P = 0.049) and PVC > 5 mmHg (OR 2.80; 95% CI: 1.04 - 7.58; P = 0.042). No significant association was found with the remaining variables. To exclude the confounding effects of diabetes on our results, we analyzed the association between Gal-3 levels and microalbuminuria among 140 non-diabetic patients by using the above-mentioned multivariate regression model, after excluding diabetes as an independent variable. In this model, Gal-3 remained significantly associated with microalbuminuria (OR: 1.18; 95% CI: 1.09 - 1.27; P < 0.001).

According to the ROC curve analysis, Gal-3 was significantly associated with microalbuminuria (AUC 0.69; 95% CI: 0.62 - 0.78). The best cutoff for Gal-3 (14.2 ng/mL) showed a sensitivity of 74% and a specificity of 56% in detecting the presence of microalbuminuria. However, as shown in the figure 1, this cutoff could detect the subgroup of patients having a higher prevalence of microalbuminuria only among patients with GFR-EPI ≥ 60 mL/minute/1.73 m^2, but not in those with GFR-EPI < 60 mL/minute/1.73 m^2.

Figure 1. Prevalence of Microalbuminuria in Patients with GFR-EPI ≥ 60 mL/Minute/Minute × 1.73 m^2 and in Those with GFR-EPI < 60 mL/minute/1.73 m^2 According to the Gal-3 Cutoff of 14.2 ng/mL



Using Gal-3, the subgroup that showed significantly higher prevalence of microalbuminuria could be detected only in patients with relatively preserved renal function. Gal-3: galectin 3. GFR-EPI: estimated glomerular filtration rate by EPI formula.

5. Discussion

To the best of our knowledge, our study findings are the first to show that Gal-3 serum levels are significantly and independently associated with microalbuminuria in CHF outpatients, thus suggesting a strong relationship between this new biomarker and CKD in this clinical setting.

Microalbuminuria plays a key role in the characterization of patients with CHF. Although estimation of GFR is considered the best overall measure of kidney function and is recommended for the routine evaluation in patients with CHF (10), it presents a number of limitations (11). As a consequence, other parameters have been proposed to better characterize renal function such as those providing integrative information on glomerular function or those reflecting tubular injury (11). In this setting, microalbuminuria is a biomarker that can integrate the information of GFR and offers additive prognostic information (12, 13). In fact, in patients with preserved GFR, microalbuminuria could represent an early sign of renal damage, reflecting a reduced number of nephrons and hyperfiltration (13, 14). Moreover, microalbuminuria could be the result of the leakage of albumin through the endothelium and glomerular basement membrane that is strictly associated to endothelial dysfunction and inflammatory cytokine activation.

By demonstrating that Gal-3 is the first determinant significantly associated with Microalbuminuria, as indicated in the forward stepwise multivariate regression analysis, our results support a possible pathophysiological link between Gal-3 and CKD. This hypothesis is further strengthened by the fact that high Gal-3 levels were significantly associated with a greater prevalence of microalbuminuria in the presence of preserved GFR. On the basis of experimental studies, Gal-3 is considered a biomarker that can promote cardiac fibrosis (1, 2) and is associated with worse prognosis in patients with CHF. However, there are few data concerning the possible relationship between Gal-3 levels and renal dysfunction in patients with CHF. Experimental studies suggest that it could prevent chronic tubular injury and attenuate fibrosis in response to ischemic and nephrotoxic injury (15), but could also promote fibrosis in cases of persistent tissue injury (16, 17). The possible involvement of Gal-3 in the genesis and progression of renal dysfunction has been also suggested by the results of Framingham Offspring Study, in which high levels of Gal-3 were associated with increased risk of GFR decline and incident CKD (7). In CHF patients, the available data have only demonstrated an independent and negative correlation between Gal-3 levels and GFR (5). However, it is unclear whether the increased Gal-3 serum levels reflect the consequence of CKD (reduced clearance and/or renal production) in patients with CHF or if these levels favor the onset and progression

of CKD by their profibrotic effects (5). The independent association between Gal-3 levels and microalbuminuria, particularly in patients with relatively preserved renal function, seems to support this last hypothesis. By promoting renal fibrosis, Gal-3 levels could mediate the progression of both renal and cardiac dysfunction, thus representing a biomarker that can better phenotyping of patients prone to progression of cardiorenal syndrome.

In our study, we did not evaluate the association between Gal-3 levels and biomarkers reflecting tubular injury that could further support our hypothesis. This is a limitation of our study. In conclusion, by demonstrating the independent association between Gal-3 levels and microalbuminuria in CHF outpatients, our study findings provide new useful data to better clarify the association between Gal-3 levels and CKD in these patients. Our results could also support the design of future studies aimed to prospectively evaluate the association among Gal-3 serum levels and the progression of renal dysfunction in CHF patients.

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Authors' Contributions

Massimo Iacoviello conceived and designed the study, critically reviewed intellectual content of the paper, and approved the final version to be submitted for publication. Nadia Aspromonte contributed towards the designing of the study, interpreting the data, critically reviewing the article's intellectual content, and approved the final version to be submitted for publication. Valeria Paradies reviewed the article's intellectual content, and approved the final version to be submitted for publication. Marta Leone analyzed and interpreted the data, reviewed the article's intellectual content, and approved the final version to be submitted for publication. Valeria Antoncecchi analyzed and interpreted the data, drafted the article and critically reviewed its intellectual content, and approved the final version to be submitted for publication. Roberto Valle analyzed and interpreted the data, reviewed the article's intellectual content, and approved the final version to be submitted for publication. Pasquale Caldarola analyzed and interpreted the data, drafted the article and critically reviewed its intellectual content, and approved the final version to be submitted for publication. Marco Matteo Ciccone analyzed and interpreted the data, drafted the article and critically reviewed its intellectual content, and approved the final version to be submitted for publication. Loreto Gesualdo designed the study, interpreted the data, critically reviewed the article's intellectual content, and approved the final version to be submitted for publication. Francesca Di Serio designed the study, analyzed and interpreted the data, drafted the article, and critically reviewed its intellectual content, and approved the final version to be submitted for publication.

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References

1. Sharma UC, Pokharel S, van Brakel TJ, van Berlo JH, Cleutjens JP, Schroen B, et al. Galectin-3 marks activated macrophages in failure-prone hypertrophied hearts and contributes to cardiac dysfunction. *Circulation.* 2004;**110**(19):3121–8.
2. Yu L, Ruifrok WP, Meissner M, Bos EM, van Goor H, Sanjabi B, et al. Genetic and pharmacological inhibition of galectin-3 prevents cardiac remodeling by interfering with myocardial fibrogenesis. *Circ Heart Fail.* 2013;**6**(1):107–17.
3. Shah RV, Chen-Tournoux AA, Picard MH, van Kimmenade RR, Januzzi JL. Galectin-3, cardiac structure and function, and long-term mortality in patients with acutely decompensated heart failure. *Eur J Heart Fail.* 2010;**12**(8):826–32.
4. Lok DJ, Van Der Meer P, de la Porte PW, Lipsic E, Van Wijngaarden J, Hillege HL, et al. Prognostic value of galectin-3, a novel marker of fibrosis, in patients with chronic heart failure: data from the DEAL-HF study. *Clin Res Cardiol.* 2010;**99**(5):323–8.
5. Gopal DM, Kommineni M, Ayalon N, Koelbl C, Ayalon R, Biolo A, et al. Relationship of plasma galectin-3 to renal function in patients with heart failure: effects of clinical status, pathophysiology of heart failure, and presence or absence of heart failure. *J Am Heart Assoc.* 2012;**1**(5):e000760.
6. Henderson NC, Mackinnon AC, Farnworth SL, Kipari T, Haslett C, Iredale JP, et al. Galectin-3 expression and secretion links macrophages to the promotion of renal fibrosis. *Am J Pathol.* 2008;**172**(2):288–98.
7. O'Seaghdha CM, Hwang SJ, Ho JE, Vasan RS, Levy D, Fox CS. Elevated galectin-3 precedes the development of CKD. *J Am Soc Nephrol.* 2013;**24**(9):1470–7.
8. Stehouwer CD, Nauta JJ, Zeldenrust GC, Hackeng WH, Donker AJ, den Ottolander GJ. Urinary albumin excretion, cardiovascular disease, and endothelial dysfunction in non-insulin-dependent diabetes mellitus. *Lancet.* 1992;**340**(8815):319–23.
9. Iacoviello M, Puzzovivo A, Guida P, Forleo C, Monitillo F, Catanzaro R, et al. Independent role of left ventricular global longitudinal strain in predicting prognosis of chronic heart failure patients. *Echocardiography.* 2013;**30**(7):803–11.
10. Fallahzadeh MK, Sagheb MM, Fallahzadeh MH. In memorandum of world kidney day: chronic kidney disease: a common but often unnoticed major health problem. *Iran Red Crescent Med J.* 2011;**13**(3):164–6.
11. Iacoviello M, Leone M, Antoncecchi V, Ciccone MM. Evaluation of chronic kidney disease in chronic heart failure: From biomarkers to arterial renal resistances. *World J Clin Cases.* 2015;**3**(1):10–9.
12. Jackson CE, Solomon SD, Gerstein HC, Zetterstrand S, Olofsson B, Michelson EL, et al. Albuminuria in chronic heart failure: prevalence and prognostic importance. *Lancet.* 2009;**374**(9689):543–50.
13. Niizeki T, Takeishi Y, Sasaki T, Kaneko K, Sugawara S, Watanabe

T, et al. Usefulness of albuminuria as a prognostic indicator in patients with chronic heart failure secondary to ischemic or idiopathic dilated cardiomyopathy. *Am J Cardiol.* 2013;**111**(8):1180–6.
14. Pinto-Sietsma SJ, Janssen WM, Hillege HL, Navis G, De Zeeuw D, De Jong PE. Urinary albumin excretion is associated with renal functional abnormalities in a nondiabetic population. *J Am Soc Nephrol.* 2000;**11**(10):1882–8.
15. Okamura DM, Pasichnyk K, Lopez-Guisa JM, Collins S, Hsu DK, Liu FT, et al. Galectin-3 preserves renal tubules and modulates extracellular matrix remodeling in progressive fibrosis. *Am J Physiol Renal Physiol.* 2011;**300**(1):F245–53.
16. Henderson NC, Sethi T. The regulation of inflammation by galectin-3. *Immunol Rev.* 2009;**230**(1):160–71.
17. Varin R, Mulder P, Tamion F, Richard V, Henry JP, Lallemand F, et al. Improvement of endothelial function by chronic angiotensin-converting enzyme inhibition in heart failure : role of nitric oxide, prostanoids, oxidant stress, and bradykinin. *Circulation.* 2000;**102**(3):351–6.