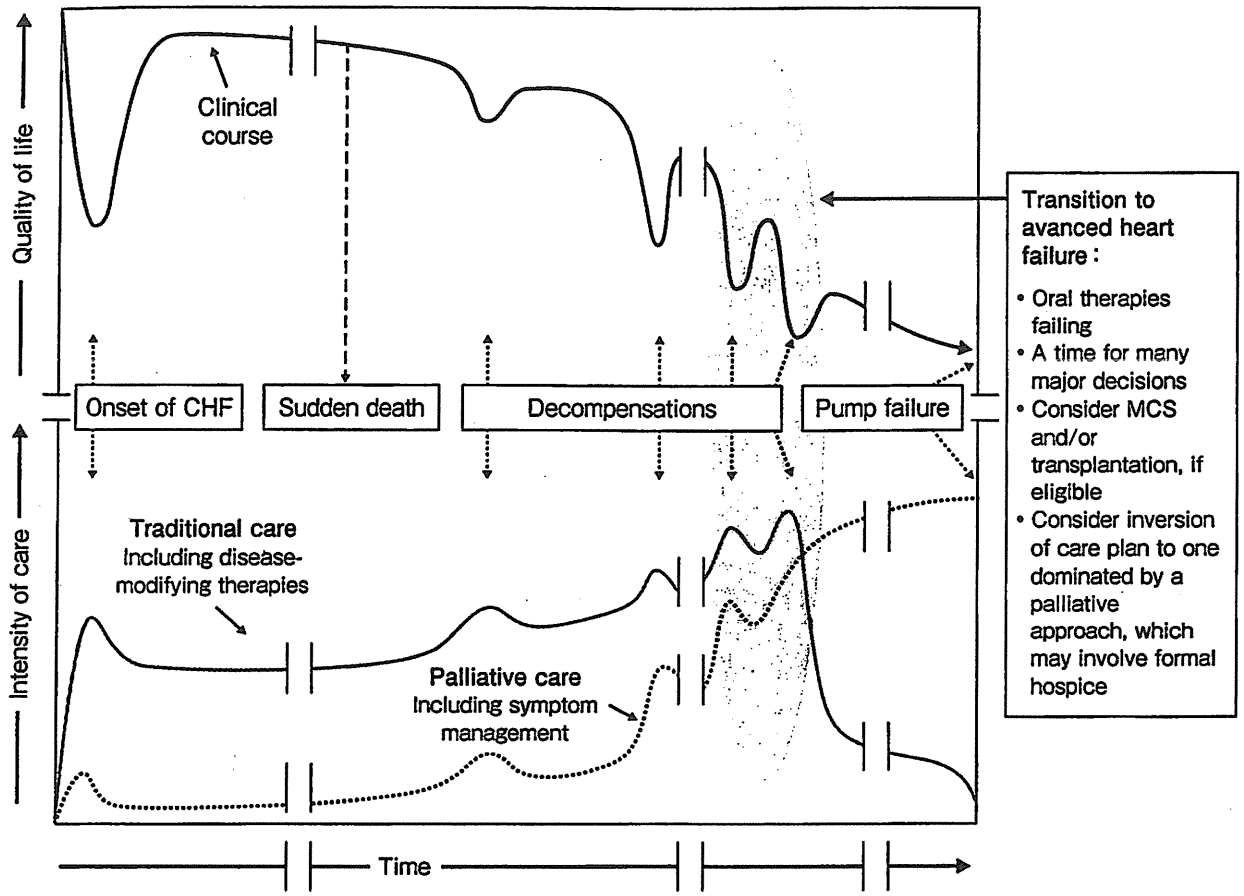

図 1　心不全の病状と選択できる治療手段の推移[4]

ある．急性例に対してはBTD，BTR，あるいはBTBとしてIABP，PCPS，VABあるいは体外設置型VASの適応が考慮される．また，慢性心不全に対して現在わが国では，心臓移植適応例へのBTTとして体外設置型VASあるいは植込み型LVASの適応が考慮される．なお，心臓移植適応がない症例に対するDTとしての植込み型LVASの適応については，現在検討されている状況である．

補助循環の適応検討

心不全の病状の推移は図1に示すように，発症から時間的経過を経てポンプ失調に至る[4]．このなかで，致死性不整脈などによる突然死の可能性もあるが，病状に応じた治療がなされる．治療を継続して行っているにもかかわらず，内科的な治療抵抗性の重症心不全に移行した状況は今後の治療方針を決定する時期であり，適応があれば機械的補助循環や心臓移植を考慮する．さらに，最近わが国でも取り入れられつつあるホスピスを含めた苦痛を軽減するケアプランへの変更を熟慮する．

このなかで，機械的補助循環の適応検討においては，補助循環が今後の治療につながることが重要である．また，①回復不能な腎機能障害，②回復不能な肝機能障害，③呼吸不全(循環不全に伴うものは除く)，④高度な血液障害(出血傾向など)，あるいは⑤重症感染症を伴う場合は，心臓ポンプ機能の補助・代行を継続して行うことが困難あるいはできなくなるため，補助循環の適応とならない．さらに，本人・家族に対する補助循環治療に関するインフォームドコンセントにおいて，図2に示すように“生存”のみならず，“人生の質”さらには“費用/負担”についても多職種が参加し，十分な考慮・検討を行い，治療について同意を得ることが必要である．とくに慢性心不全例においては，患者本人による補助循環や心臓移植治療に関する同意が求められる[5-7]．

補助循環の中止を検討すべき状況とは？

補助循環を用いることにより全身循環が改善し，さらに良好に維持され，BTD，BTR，あるい

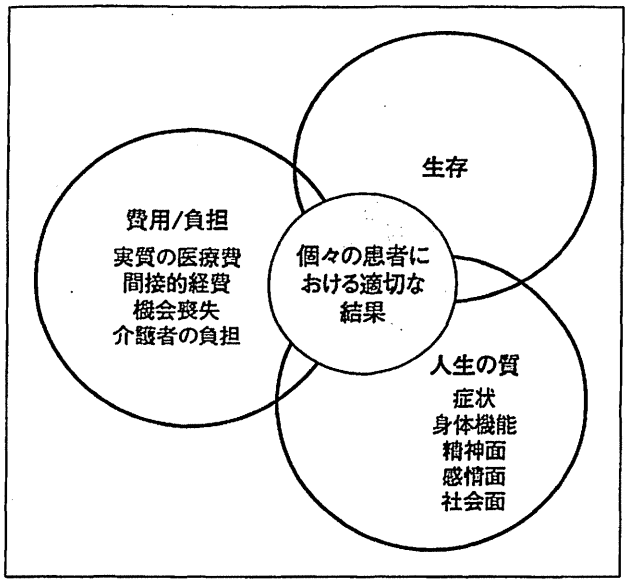

図 2　心不全における治療選択[4]

はBTTをめざしている状況や，DTとして長期在宅治療を行っている場合では，補助循環の継続は妥当である．しかし，表1に示すような補助循環による効果を期待できない状態においては，補助循環の適応について再度検討する必要がある．

補助循環の適応決定

難治性心不全における補助循環治療選択の決定は困難な場合が多い．致死性不整脈や，急性心筋炎あるいは急性心筋梗塞による急性発症例では，患者の意思確認ができず家族による判断が求められる．また，各種心筋症による慢性心不全で前述の重症心不全となったが，今後の治療方針が明確になっていない状況における急性増悪においても，短時間での本人・家族の判断が求められることになる．医療における意思決定においては，これまで医師に決定をゆだねるパターナリズムや，患者・家族主体で自律的に決める場合が多くみられた．近年，医師・看護師を含む多職種の医療者が意思決定に関与するようになり，患者・家族とともに医師・看護師を含む医療者が共同で意思決定するshared decision makingが望ましいとされている．

補助循環治療の選択を行う場合は直接死につながる状況である．また，図2で示した事柄の検討に加え，後述する補助循環に基づく出血，感染，脳血管障害などの合併症などにより，治療中止を考慮すべき状況となりうることも踏まえる必要ある．多くは短時間での決断が必要なため，とくに死をタブー視するわが国においては精神的および心理的に危機的状況に陥りやすい(表1)[8]．このため，多職種の関与が重要となる．

難治性心不全において補助循環を効果的に組み込んだ治療を進めるためには，慢性心不全においてタイミングを逸することなく，患者・家族が望む治療と生き方を事前に対話するアドバンスケアプランニング(advance care planning：ACP)[4]が，さらには治療選択の前段階から終末期までの治療選択の意思決定支援(preparedness planning)が推奨されている[9]．

補助循環の継続

補助循環の目的は前述したように，心臓ポンプ

表 1　終末期における診療方針決定を困難にしている要因[8]

- 医学的な不確定要素が多い(不確実性が高い)
- 延命措置・生命維持治療の著しい進歩(ゼロにならない希望)
- 患者の揺れる気持ち
- 死に対する恐怖・嫌悪・忌避(誰も死にたくない，家族を失いたくない)
- 不十分なアドバンスケアプラニング(患者の意思推定が困難)
- 患者本人の意思決定能力低下
- 関係者が抱く“何もしないという選択肢”の心理的困難さ
- 関係者間の意見の不一致，価値観の対立
- 倫理的不確実性(どうすることが正しいのかわからない)
- Quality of life(QOL)，人間の尊厳，そして治療の利益と害に関する関係者の異なる評価
- 他人の生き死にを決める代理決定者の心理的負担
- 曖昧な医学的介入中止の基準(治療義務の限界)
- “世間”の救命・延命技術に対する過大な期待
- 法の空白

表 2　補助循環における終末期の定義[5]

(1) Bridge to decision(BTD)として適応されている場合
・心臓に対する治療手段(冠動脈再建，心臓弁への加療など)がなく，自己心機能の回復が期待できない場合
・諸臓器(腎，肝など)機能障害が高度で回復不能と判断される場合
・高度な脳神経障害を認める場合
・呼吸不全(循環不全に伴うものは除く)を認める場合
・高度な血液障害(出血傾向など)を認める場合
・重症感染症を認める場合
(2) Bridge to recovery(BTR)として適応されている場合
・各種治療を加えても自己心機能の回復が期待できず，心臓移植の適応とならない場合
・諸臓器(腎，肝など)機能障害が高度で回復不能と判断される場合
・高度な脳神経障害を認める場合
・呼吸不全(循環不全に伴うものは除く)を認める場合
・高度な血液障害(出血傾向など)を認める場合
・重症感染症を認める場合
(3) Bridge to transplantation(BTT)として適応されている場合
・諸臓器(腎，肝など)機能障害が高度で回復不能と判断される場合
・高度な脳神経障害を認める場合
・呼吸不全(循環不全に伴うものは除く)を認める場合
・高度な血液障害(出血傾向など)を認める場合
・重症感染症を認める場合
・心臓移植の適応から外れるようになった場合(一時的な状態で適応に復帰できる可能性がある場合は除く)
(4) Destination therapy(DT)あるいは長期在宅治療として適応されている場合
・諸臓器(腎，肝など)機能障害が高度で回復不能と判断される場合
・高度な脳神経障害を認める場合
・呼吸不全(循環不全に伴うものは除く)を認める場合
・高度な血液障害(出血傾向など)を認める場合
・重症感染症を認める場合
・在宅治療を行うことができない状態(一時的な状態で加療により在宅治療の継続が可能な場合は除く)

機能を補助または代行し良好に全身循環を維持することにより時間的猶予を得てつぎの手段に移ることをめざすものである．さらに，DTにおいては補助循環による生活をめざすものである．循環が良好に維持された状況において，心臓以外の脳を含む諸臓器機能不全などでその治療目的が達成できないと考えられる場合(終末期)には，補助循環の継続について検討すべきである(表2)．この場合，あらたな治療を行わないことや，施行中のPCPSなどの回路交換を実施しないことなども考慮する必要がある．

わが国における補助循環法(IABP，PCPS，VAB，およびVAS)は補助能力や施行可能期間が異なっている．このため，良好な循環が行えない場合に，より高度な補助が行えるシステムや，長期施行可能なシステムへの変更が検討されることがある．しかし，システム変更，とくにVASへの変更は手術侵襲が大きく，またVASにおいても期待できるのは循環動態改善に基づく諸臓器機能の回復で，その効果は限られており，タイミングも重要である．とくに，すでに高度の臓器障害を伴っている場合は回復を期待しがたい．このため，システム変更は慎重に検討する必要がある．

補助循環適応の意思確認において前述したように，急性心不全例では本人意思を確認できない状態で補助循環を開始せざるをえない場合が多く，また，治療の継続についても本人の意思を確認できない場合が多い．このため，家族に説明し同意を得て治療を行うこととなる．家族に対し補助循環継続による治療効果の見込みについて説明し，治療効果が得がたい末期的状況であることを十分説明し，理解が得られ家族が受容すれば，補助循環の中止を検討する．なお，末期的状況であることについては多職種チームによりコンセンサスを得ることが必要である．また，末期的状況において家族が治療継続を希望した場合には，病状およびその対応について十分に説明し，理解を得る．この場合，それぞれのシステムの限界を超えた治

療(PCPSなどの回路交換など)，あらたな治療，あるいは他の臓器不全に対する補助手段(透析など)は行わないのは妥当と考えられる．

慢性心不全例では，適応において治療に関するインフォームドコンセントを行うので，その際に本人および家族に末期的状況となった場合には十分な説明と同意を得たうえで，補助循環の中止を行うことにも同意を得る[10,11]．末期的状況となったとの判断は多職種チームによる検討により行い，本人および家族(本人の意思が確認できない場合は家族のみ)に病状について十分説明を行い，本人・家族(本人の意思が確認できない場合は家族のみ)が受容した段階で補助循環を中止する．家族が補助循環の継続を希望した場合においても，あらたな治療を加えることは医学的適応がないことについて再度説明したうえで行わないのが妥当と考えられる．なお，末期状態に及ぶと想定される状況となれば，末期における対応について本人・家族と相談し，対応方法を決定しておくことが望ましい．

補助循環施行中に末期状態でないにもかかわらず本人および家族から継続中止の要望があった場合，本人・家族も含めて多職種チームで協議し，そのうえで倫理委員会にはかる．

文献/URL

1) Rose, E. A. et al.：Randomized Evaluation of Mechanical Assistance for the Treatment of Congestive Heart Failure(REMATCH)study group. Long term mechanical left ventricular assistance for end-stage heart failure. *N. Engl. J. Med.*, **345**：1435-1443, 2001.
2) Slaughter, M. S. et al.：Advanced heart failure treated with continuous-flow left ventricular assist device. *N. Engl. J. Med.*, **361**：2241-2251, 2009.
3) 中谷武嗣：わが国における補助人工心臓治療の歴史．今日の移植，**26**：299-306，2015.
4) Allen, L. A. et al.：Decision making in advanced heart failure. *Circulation*, **125**：1928-52, 2012.
5) 日本循環器学会・他：循環器疾患における末期医療に関する提言(循環器病の診断と治療に関するガイドライン(2008-2009年度合同研究班報告)．2010，pp.36-38.
6) 日本循環器学会・他：重症心不全に対する植込型補助人工心臓治療ガイドライン．日本循環器学会・日本心臓血管外科学会合同ガイドライン(2011-2012年度合同研究班報告)．2013，p180.
7) 補助人工心臓治療関連学会協議会，Destination Therapy(DT)研究会：我が国における植込型補助人工心臓適応適正化の考え方—Destination Therapyについて．2015．(http://www.jacvas.com/view_dt.html)
8) 浅井　篤：緩和ケアに関する倫理的問題．心不全の緩和ケア—心不全患者の人生に寄り添う医療(大石醒悟・他編)．南山堂，2014，pp.213-223.
9) Swetz, K. M. et al.：Preparedness planning before mechanical circulatory support：A "How-to" guide for palliative medicine clinicians. *J. Pain Symptom. Manage.*, **47**：926-935, 2014.
10) Brush, S. et al.：End-of-life decision making and implementation in recipients of a destination left ventricular assist device. *J. Heart Lung Transplant.*, **29**：1337-1341, 2010.
11) Blumenthal-Barby, J. S. et al.：Assessment of patient's and caregivers'informational and decisional needs for left ventricular assist device placemtn：Implications for informed consent and shared decision-making. *J. Heart Lung Transplant.*, **34**：1182-1189, 2015.

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ORIGINAL ARTICLE
Heart Failure

Plasma Renin Activity Is a Strong and Independent Prognostic Indicator in Patients With Acute Decompensated Heart Failure Treated With Renin-Angiotensin System Inhibitors

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Background: The renin-angiotensin system (RAS) is activated in heart failure (HF) as a compensatory mechanism, being related to cardiac remodeling and poor prognosis. Although RAS inhibitors are used as first-line drugs for HF, plasma renin activity (PRA) is upregulated by RAS inhibitors via a negative feedback mechanism. The clinical significance of PRA during RAS inhibitor therapy is poorly understood in acute decompensated HF (ADHF). Therefore we examined the impact of PRA in HF patients already receiving RAS inhibitors.

Methods and Results: Of 611 consecutive patients with ADHF and emergency admission to hospital, we studied the impact of PRA on the prognosis of ADHF in 293 patients already receiving RAS inhibitors before admission. The patients were divided into 2 groups according to median PRA (≥ vs. <3.4 $ng \cdot ml^{-1} \cdot h^{-1}$). During a mean follow-up of 29.0 months, there were 124 deaths from all causes. Kaplan-Meier analysis showed that all-cause and cardiovascular mortality were significantly higher in patients with high PRA than low PRA (log-rank P=0.0002 and P<0.0001, respectively). Log PRA was an independent predictor of all-cause and cardiovascular death (HR, 1.194; 95% CI: 1.378–2.678, P<0.0001; and HR, 2.559; 95% CI: 1.610–4.144, P<0.0001, respectively).

Conclusions: PRA was associated with an increased risk of all-cause and cardiovascular mortality in ADHF patients already receiving RAS inhibitors, suggesting that PRA would be a useful biomarker during ADHF treatment. (*Circ J* 2015; **79:** 1307–1314)

Key Words: Acute decompensated heart failure; Plasma renin activity; Prognosis; Renin-angiotensin system blocker

In spite of great advances in the management of acute decompensated heart failure (ADHF), morbidity and mortality are still high and patient quality of life is impaired.[1–3] To improve the prognosis of ADHF, more sensitive and accurate diagnostic tools and more effective therapeutic approaches are necessary. The renin-angiotensin system (RAS) is fundamentally involved in the development and progression of heart failure (HF), which is initially upregulated in HF[4,5] to maintain cardiac output in order to maintain sufficient perfusion of vital organs. Overactivation of the RAS, however, ultimately results in increased afterload and body fluid retention, which leads to a vicious cycle of decompensated HF. Given that renin is the rate-limiting enzyme of the RAS, it is reasonable that measurement of plasma renin activity (PRA) helps to determine the degree of RAS activation in the clinical setting of HF. In fact, some earlier studies reported a strong inverse correlation between survival and PRA.[6–8]

Editorial p1206

After seminal clinical trials demonstrating that angiotensin-converting enzyme inhibitors (ACEI), angiotensin receptor blockers (ARB) and β-adrenergic receptor blockers can effectively improve the prognosis of HF,[9–12] however, they have been routinely used as first-line treatment for HF. During RAS inhibitor therapy, PRA is elevated due to decreased production of angiotensin II, which negatively regulates renin release.

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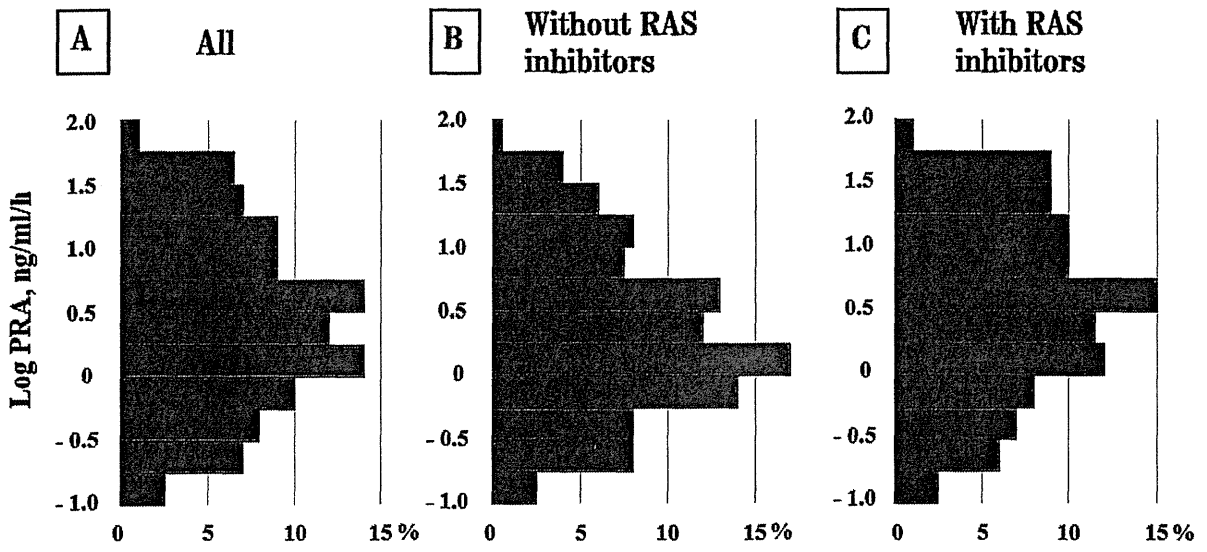

Figure 1. Distribution of log plasma renin activity (PRA) for (**A**) all patients (n=505), (**B**) patients without renin-angiotensin system (RAS) inhibitors (n=212), and (**C**) patients with RAS inhibitors (n=293).

β-blockers directly suppress PRA via inhibition of renal sympathetic activity. Moreover, loop diuretics, which block the Na+/K+/2Cl− co-transporter and stimulate renin release, are widely used to treat HF. Therefore, PRA is considerably altered by HF treatment. There is a paucity of data on the clinical interpretation of PRA as a biomarker in ADHF and its implications, although renin is the rate-limiting step in RAS activation. Compared to the large body of literature concerning brain natriuretic peptide (BNP) or BNP-related peptide as a prognostic marker of ADHF, very little is known about PRA.

Here we show for the first time the clinical impact of PRA on prognosis in patients with ADHF, all of whom were already being treated with ACEI, ARB, or both in the Nara Registry and Analyses for Heart Failure 2 (NARA-HF 2 study) cohort study.

Methods

Patient Selection

The NARA-HF study is a dynamic cohort study.[13] The NARA-HF 2 study recruited 611 consecutive patients with emergency admission to the internal medicine or cardiology wards or the coronary care unit at Nara Medical University Hospital with documented ADHF (either acute new-onset or acute-on-chronic HF) between January 2007 and December 2012. The diagnosis of HF was based on the Framingham criteria for HF.[14] Patients with acute myocardial infarction (AMI), acute myocarditis, and acute HF with acute pulmonary embolism were excluded.

Of the 611 patients, 505 patients had PRA measurement on admission. Among them, 293 patients had already received ACEI, ARB, or a combination of RAS inhibitors before admission but the remaining 212 patients had not been previously treated. We investigated the impact of PRA on the prognosis of ADHF in the 293 patients who had already received RAS inhibitors, but not direct renin inhibitors. Patients were divided into low PRA (n=147) and high PRA (n=146) groups based on median PRA (3.4 $ng \cdot ml^{-1} \cdot h^{-1}$). For each patient, baseline data included age, sex, body mass index (BMI), cause of HF, medical history, vital signs, laboratory and echocardiographic data, and medications on admission and at discharge.

Outcomes

The primary endpoints were all-cause and cardiovascular mortality. Cardiovascular death was defined as death due to HF, myocardial infarction, sudden death, stroke, and vascular disease such as aortic dissection. We checked medical records to determine vital status and the cause of death. When this information was unavailable in the medical record, we telephoned patients or their families. Information regarding cardiovascular events such as non-fatal AMI, stroke, and rehospitalization due to recurrence of ADHF was also obtained.

Statistical Analysis

Continuous variables are expressed as mean±SD and were compared using Student's t-test. Categorical variables are summarized with frequency percentages and were analyzed using chi-squared test. Cumulative event-free rates during follow-up were derived using the Kaplan-Meier method. Univariate and multivariate analyses of mortality were performed using Cox proportional hazards models. We utilized 4 models for the adjustment of covariates: model 1, unadjusted; model 2, adjusted for age and sex; model 3, adjusted for all factors in model 2 plus hemoglobin concentration (Hb), estimated glomerular filtration rate (eGFR), and sodium and BNP; and model 4, adjusted for all factors in model 3 plus left ventricular ejection fraction (LVEF) and systolic blood pressure (SBP). Multiple linear regression was performed to determine the variables that affected PRA.

Results are reported as hazard ratios (HR), coefficients, 95% confidence intervals (95% CI), and P-value. The HR for outcomes in the high PRA group was compared with those for

Table 1. Baseline HF Patient Characteristics

Characteristics	Total (n=293)	Low PRA (n=147)	High PRA (n=146)	P-value
Demographic				
Age (years)	73.4±11.9	75.4±9.9	71.4±13.3	0.0303
Female	38.2	42.2	34.2	0.1625
BMI (kg/m^2)	23.7±4.1	23.6±4.0	23.9±4.2	0.3491
Cause of HF				
Ischemic	43.3	40.1	46.6	0.2661
Valvular	17.1	17.7	16.4	0.7763
Dilated cardiomyopathy	16.0	12.9	19.2	0.1448
Hypertensive	6.1	8.8	3.4	0.0534
Medical history				
Diabetes mellitus	49.2	44.2	54.1	0.0904
Dyslipidemia	44.3	44.8	43.8	0.8750
Old MI	36.9	34.0	39.7	0.3109
Dialysis	5.5	6.1	4.8	0.6169
Procedures				
PCI	31.9	27.9	35.9	0.1438
CABG	5.1	4.1	6.2	0.4186
CRT/ICD	3.1	2.0	4.1	0.3048
NYHA class on admission				
III or IV	88.4	89.8	87.0	0.4528
Vital sign on admission				
SBP (mmHg)	145.0±36.5	155.9±34.5	134.0±35.2	<0.0001
DBP (mmHg)	80.4±21.9	85.3±23.2	75.3±19.4	<0.0001
Heart rate (beats/min)	92.1±25.6	89.4±26.1	94.8±24.9	0.0416
Echocardiographic parameters				
LVEF (%)	46.6±16.7	50.5±15.4	42.6±17.0	<0.0001
EF ≥50%	45.4	52.4	38.2	0.0151
LVEDD (mm)	55.7±10.6	53.8±8.8	57.7±12.0	0.0064
Laboratory data on admission				
Hemoglobin (g/dl)	11.1±2.3	10.8±2.3	11.4±2.3	0.0072
eGFR (ml/min/1.73 m^2)	38.6±23.3	38.7±24.1	38.4±22.5	0.9291
CKD stage 4 or 5	38.9	38.8	39.4	0.9628
Sodium (mmol/L)	139.3±4.4	140.3±3.4	138.4±5.0	0.0003
Potassium (mmol/L)	4.23±0.83	4.13±0.77	4.33±0.88	0.1273
PRA (ng·ml^{-1}·h^{-1})	3.4 (1.0–12.1)	1.0 (0.5–1.9)	12.1 (5.4–25.5)	<0.0001
Aldosterone (pg/ml)	63.2 (35.9–108.6)	56.4 (31.3–81.0)	84.1 (44.4–148.6)	<0.0001
Plasma BNP (pg/ml)	892 (457–1,658)	972 (518–1,706)	757 (364–1,569)	0.1007
Medication				
Admission				
ACEI	47.1	40.1	54.1	0.0166
ARB	66.6	71.4	61.6	0.0759
ACEI or ARB	100	100	100	1.0000
β-blockers	35.2	38.1	32.2	0.2900
Loop diuretics	60.1	56.5	63.7	0.2060
MR blockers	21.8	17.0	26.7	0.0444
Ca channel blockers	42.0	48.3	35.6	0.0278
Statin	28.7	27.9	29.5	0.7677
Discharge				
ACEI	54.5	53.7	55.2	0.7972
ARB	52.1	56.5	47.6	0.1289
ACEI or ARB	91.4	94.6	88.1	0.0505
β-blockers	54.5	51.7	57.3	0.3348
Loop diuretics	77.6	78.2	76.9	0.7894
MR blockers	30.0	28.6	31.5	0.5904
Ca channel blockers	34.1	40.8	27.3	0.0150

Data given as %, mean±SD, or median (25th–75th percentile). ACEI, angiotensin-converting enzyme inhibitor; ARB, angiotensin receptor blocker; BMI, body mass index; BNP, B-type natriuretic peptide; Ca, calcium; CABG, coronary artery bypass grafting; CKD, chronic kidney disease; CRT, cardiac resynchronization therapy; DBP, diastolic blood pressure; EDD, end-diastolic diameter; EF ejection fraction; eGFR, estimated glomerular filtration rate; HF, heart failure; ICD, implantable cardioverter defibrillator; LV, left ventricular; MI, myocardial infarction; MR, mineralocorticoid receptor; NYHA, New York Heart Association; PCI, percutaneous coronary intervention; PRA, plasma renin activity; SBP, systolic blood pressure.

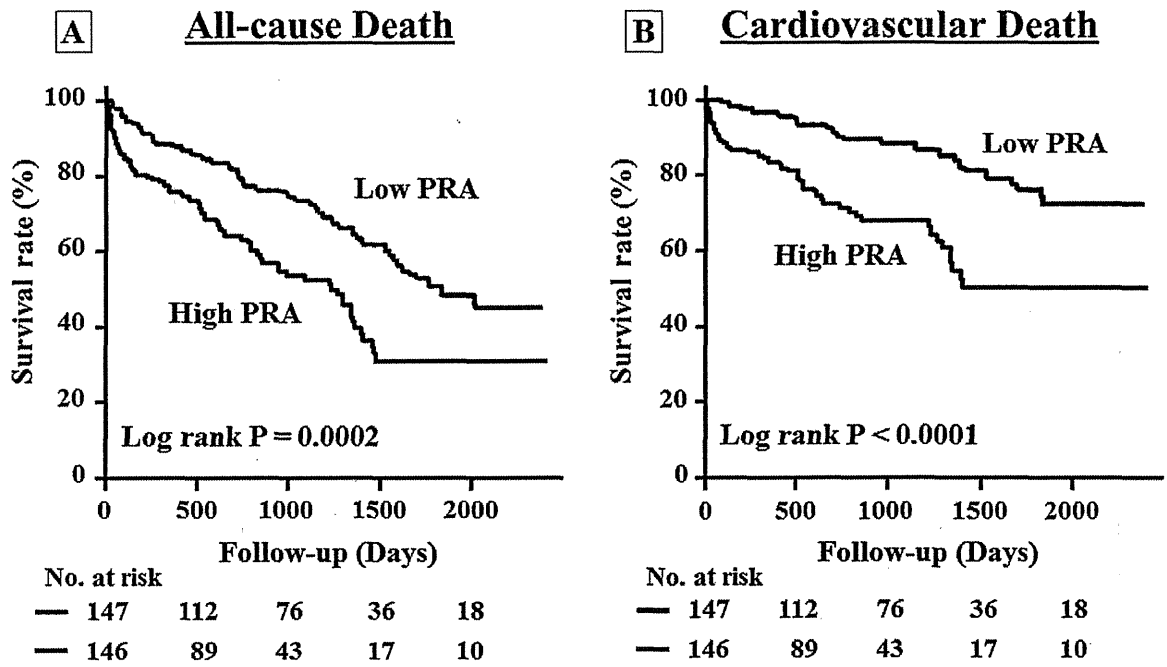

Figure 2. Kaplan-Meier event-free survival curves for (**A**) all-cause death and (**B**) cardiovascular death in patients with plasma renin activity (PRA) ≥3.4 ng·ml^{-1}·h^{-1} (blue line, high PRA group; n=146) compared with patients with PRA <3.4 ng·ml^{-1}·h^{-1} (red line, low PRA group; n=147).

the low PRA group, which served as the reference group. Variables with P<0.05 were retained in the model. JMP version 10 for Windows (SAS Institute, Cary, NC, USA) was used for all statistical analyses.

Results

PRA and RAS Inhibitor Therapy

Among the 611 patients who participated in this registry study, PRA was measured in 505 patients at the time of admission. Among them, 293 patients had already been treated with ACEI, ARB, or both, but 212 had not been on RAS inhibitors. Both PRA and logarithmically transformed PRA were significantly higher in patients treated with ACEI, ARB, or both than those who were not (mean±SD, 9.1±12.6 ng·ml^{-1}·h^{-1} vs. 6.0±10.3 ng·ml^{-1}·h^{-1}, P=0.0011; and 0.51±0.69 ng·ml^{-1}·h^{-1} vs. 0.31±0.64 ng·ml^{-1}·h^{-1}, P=0.0011, respectively). As shown in **Figure 1**, the histogram of logarithmically transformed PRA was shifted up in patients treated with RAS inhibitors. Age, proportion of women, proportion of New York Heart Association (NYHA) class III or IV patients, LVEF, and plasma BNP were similar between the groups. We investigated whether or not PRA at admission is associated with all-cause or cardiovascular mortality in the group of 293 patients who were already being treated with RAS inhibitors.

Baseline Characteristics

Mean age of the 293 patients was 73.4±11.9 years, and the proportion of women was 38.2% (**Table 1**). To investigate the impact of PRA on prognosis of ADHF, we divided patients into 2 groups according to median PRA on admission. **Table 1** lists baseline clinical characteristics vs. high and low PRA. Compared with patients in the low PRA group, the patients in the high PRA group were significantly younger, but the proportion of men and women and BMI were similar. There were no significant differences in the cause of HF or the proportion of comorbidities between the 2 groups. Laboratory findings except Hb, sodium, and aldosterone were similar between the groups, as shown in **Table 1**. Although NYHA functional class and plasma BNP were similar between the groups, patients with high PRA had significantly lower SBP and diastolic blood pressure (DBP), larger left ventricular end-diastolic diameter (LVEDD), and lower LVEF compared with those with low PRA.

The proportion of patients treated with β-blockers or loop diuretics was similar in the 2 groups both on admission and at discharge. Calcium (Ca) channel blockers were less frequently used in the high PRA group on admission and at discharge. Mineralocorticoid receptor blockers were more frequently used in the high PRA group on admission but the rates of use were similar between the groups at discharge.

Prognosis and Outcome

During the mean follow-up period of 29.0 months, 124 patients (42.3%) died; 68 (23.2%) from cardiovascular causes. As shown in the Kaplan-Meier survival curves, the high PRA

Table 2. Effect of PRA on Adverse Outcome

	PRA <3.4 ng·ml^{-1}·h^{-1} (n=147)	PRA ≥3.4 ng·ml^{-1}·h^{-1} (n=146)	P-value
All-cause death			
Unadjusted HR (95% CI)	1	1.965 (1.375–2.830)	0.0002
Adjusted HR (95% CI)	1	2.259 (1.530–3.353)	<0.0001
Cardiovascular death			
Unadjusted HR (95% CI)	1	3.243 (1.950–5.597)	<0.0001
Adjusted HR (95% CI)	1	3.668 (2.120–6.547)	<0.0001

The Cox proportional hazards model adjusted for the following covariates: age, sex, hemoglobin, eGFR, LVEF, BNP, and sodium. CI, confidence interval; HR, hazard ratio. Other abbreviations as in Table 1.

Table 3. Significant Factors in All-Cause and CV Death

	All-cause death		CV death	
	HR (95% CI)	P-value	HR (95% CI)	P-value
Model 1				
Log PRA (ng·ml^{-1}·h^{-1})	1.803 (1.373–2.380)	<0.0001	2.660 (1.815–3.960)	<0.0001
Model 2				
Log PRA (ng·ml^{-1}·h^{-1})	2.059 (1.550–2.752)	<0.0001	2.917 (1.953–4.433)	<0.0001
Age (years)	1.036 (1.020–1.054)	<0.0001	1.022 (1.002–1.044)	0.0300
Male	1.284 (0.887–1.880)	0.1862	1.233 (0.742–2.100)	0.4235
Model 3				
Log PRA (ng·ml^{-1}·h^{-1})	2.175 (1.604–2.964)	<0.0001	3.242 (2.100–5.095)	<0.0001
Age (years)	1.034 (1.018–1.052)	<0.0001	1.018 (0.998–1.040)	0.0755
Male	1.308 (0.894–1.932)	0.1673	1.295 (0.769–2.229)	0.3343
Hemoglobin (g/dl)	0.914 (0.833–1.004)	0.0602	0.87 (0.771–0.984)	0.0271
eGFR (ml/min/1.73 m^2)	0.998 (0.988–1.006)	0.5593	1.006 (0.993–1.017)	0.3649
Sodium (mmol/L)	0.971 (0.929–1.017)	0.2079	0.959 (0.907–1.020)	0.1760
Plasma BNP (100 pg/ml)	1.016 (1.003–1.028)	0.0159	1.022 (1.004–1.038)	0.0161
Model 4				
Log PRA (ng·ml^{-1}·h^{-1})	1.914 (1.378–2.678)	<0.0001	2.559 (1.610–4.144)	<0.0001
Age (years)	1.033 (1.015–1.052)	0.0001	1.015 (0.994–1.038)	0.1595
Male	1.326 (0.900–1.974)	0.1546	1.390 (0.821–2.407)	0.2229
Hemoglobin (g/dl)	0.913 (0.826–1.007)	0.0699	0.881 (0.773–1.002)	0.0535
eGFR (ml/min/1.73 m^2)	0.995 (0.986–1.004)	0.2974	1.002 (0.990–1.013)	0.7809
Sodium (mmol/L)	0.972 (0.930–1.019)	0.2380	0.958 (0.906–1.019)	0.1689
Plasma BNP (100 pg/ml)	1.015 (1.001–1.028)	0.0310	1.022 (1.004–1.039)	0.0191
LVEF (%)	1.002 (0.988–1.016)	0.8224	1.008 (0.989–1.026)	0.4185
SBP (mmHg)	0.992 (0.987–0.998)	0.0080	0.989 (0.981–0.997)	0.0042

CV, cardiovascular. Other abbreviations as in Tables 1,2.

group had a much higher rate of all-cause death (log-rank P=0.0002) and cardiovascular death (log-rank P<0.0001; **Figure 2**). **Table 2** shows unadjusted and adjusted HR for outcomes in the 2 groups. Compared with the low PRA group, the unadjusted HR for all-cause and cardiovascular death were significantly higher in the high PRA group (HR, 1.965; 95% CI: 1.375–2.830, P=0.0002; and HR, 3.243; 95% CI: 1.950–5.597, P<0.0001, respectively). Even after adjustment for covariates (age, sex, Hb, eGFR, LVEF, BNP, and Na) in multivariate Cox proportional hazard models, these findings remained significant (**Table 2**). In addition, rehospitalization due to HF recurrence was significantly higher in the high PRA group (P=0.0369). There were no differences, however, in the frequency of non-fatal acute MI or stroke between the 2 groups.

As shown in **Table 3**, PRA predicted all-cause death and cardiovascular death (P<0.0001 and P<0.0001, respectively). Even after adjusting for age, sex, and cardiovascular risk factors, these findings remained significant (**Table 3**). These results were similar when patients on chronic dialysis were excluded.

Factors Affecting PRA

We also performed multiple linear regression to identify factors affecting PRA. As shown in **Table 4**, PRA was associated with age, sodium, SBP, and LVEF, but not sex, Hb, BNP, aldosterone, or medication.

Table 4. Variables Affecting PRA

	Coefficient	95% CI	P-value
Age (years)	−0.161	−0.278 to −0.044	0.0073
Male	2.373	−0.481 to 5.227	0.1028
Hemoglobin (g/dl)	−0.023	−0.698 to 0.652	0.9468
Sodium (mmol/L)	−0.381	−0.687 to −0.076	0.0145
Plasma BNP (100 pg/ml)	−0.060	−0.176 to 0.055	0.3061
Aldosterone (pg/ml)	0.005	−0.001 to 0.010	0.0559
SBP (mmHg)	−0.085	−0.124 to −0.046	<0.0001
LVEF (%)	−0.102	−0.200 to −0.004	0.0407
β-blocker	−1.972	−4.866 to 0.922	0.1808
Loop diuretic	2.159	−0.668 to 4.985	0.1338
Calcium channel blocker	−1.487	−4.365 to 1.390	0.3098

If a patient was male or treated with medicine, the variable was assigned a value of 1; otherwise, 0 was assigned. Abbreviations as in Tables 1,2.

Discussion

Earlier studies showed that PRA is a risk factor for poor prognosis in patients with essential hypertension or chronic HF,[15–18] but they do not stratify patients according to RAS inhibitor status. In the present study, we demonstrate for the first time that PRA is a strong risk factor associated with all-cause and cardiovascular mortality in patients with ADHF already being treated with RAS inhibitors. This risk was still significant after adjustment for other risk factors such as age, anemia, eGFR, LVEF, and BNP. As with eGFR or BNP, PRA is a stronger predictor of all-cause and cardiovascular mortality. In contrast to earlier works, however, in the NARA-HF2 study, we could show only that high PRA tended to be associated with poor prognosis in ADHF patients who had not been treated with RAS blockers (log rank P=0.0841, data not shown). Current guidelines for the management of HF strongly recommend RAS inhibitors and β-blockers as first-line drugs with the goal of improving prognosis.[19–21] Most patients with HF receive RAS inhibitors and β-blockers if they do not have any contraindications. In this context, studying biomarkers, which are possibly altered by the use of these drugs, is becoming more important, to better understand the meaning of biomarkers.

In this study, we compared two groups based on median PRA (3.4 $ng \cdot ml^{-1} \cdot h^{-1}$), but it is not clear which cut-off point is clinically proper. We therefore also examined two other criteria: the upper reference value of PRA (2.0 $ng \cdot ml^{-1} \cdot h^{-1}$) and the best cut-off point according to receiver operating characteristic curve analysis (8.2 $ng \cdot ml^{-1} \cdot h^{-1}$). As shown in the Kaplan-Meier survival curves in **Figures S1,S2**, the higher PRA group had a much higher rate of all-cause death and cardiovascular death for both evaluations (log-rank P<0.0001 for both) as well as for median PRA, indicating that higher PRA is an predictor of poorer outcome in ADHF patients being treated with RAS blockers. Moreover, as shown in **Figure 2**, patients in the high PRA group were lost mostly at 100–200 days after admission. Within 200 days after admission, the proportion of cardiovascular death was higher in patients with high PRA than in those with low PRA (19.2% vs. 8.8%, P=0.0100). It is possible, therefore, that high PRA is more related to severe HF.

Although PRA is generally upregulated in HF as a reflection of RAS activation, there was a wide distribution of PRA, ranging from 0.1 to >60 $ng \cdot ml^{-1} \cdot h^{-1}$ in patients with ADHF who were already being treated with RAS inhibitors. To date it has not been well investigated as to which factors determine the higher PRA in patients who were treated with RAS inhibitors. Generally, expression and secretion of renin is upregulated by decreases in arterial pressure detected by baroreceptors, decreases in sodium chloride influx into the juxtaglomerular apparatus through the Na+/K+/2Cl− co-transporter, and activation of renal sympathetic nerve activity, and downregulated by angiotensin II in a negative feedback loop. Thus, β-blockers lower PRA, but RAS inhibitors and loop diuretics increase PRA. In the setting of HF, RAS regulation is more complex. For example, negative feedback is blunted[22] and alternative pathways such as the chimase-dependent pathway are activated.[23]

As shown in **Table 1**, there was no significant difference in the proportion of β-blockers or loop diuretics used. LVEDD was significantly larger, whereas LVEF, blood pressure, and serum sodium were significantly lower in the high vs. low PRA group. Moreover, on multivariate regression analysis SBP, LVEF, and serum sodium concentration were inversely related to PRA (**Table 4**). These findings suggest that LV remodeling was more advanced in the high PRA group. Significantly lower serum sodium may be the result of high doses of loop diuretics in the high PRA group, despite similar numbers of patients on loop diuretics in both groups. To confirm this hypothesis, loop diuretics other than furosemide were converted to furosemide equivalent doses: 4 mg of torasemide and 30 mg of azosemide were considered equivalent to 20 mg of furosemide. After conversion, there were no significant differences in furosemide equivalent dose between the high and low PRA groups. In patients with PRA ≥12.1 $ng \cdot ml^{-1} \cdot h^{-1}$ (top quartile), the furosemide equivalent dose was significantly higher than in the remaining patients (55.7±37.9 mg vs. 40.8±24.2 mg; P=0.0298). Although more detailed study is needed, the present findings suggest that high PRA may be correlated with the severity of HF itself rather than the effect of drugs used to treat it.

Aldosterone, an end-product of RAS, is involved in the pathophysiology of HF, as evidenced by recent clinical trials demonstrating that aldosterone blockers reduce mortality rates in patients with moderate–severe chronic HF and acute HF.[24–26] In this study, plasma aldosterone was significantly higher in the high PRA group compared with the low PRA group, suggesting insufficient suppression of RAS in the present patients. Another explanation is so called aldosterone

breakthrough phenomenon. In contrast with PRA, plasma aldosterone was not a risk factor for worse prognosis in the present patients (data not shown), as in past studies.[7] The precise reason for the discrepancy in prognostic ability between PRA and plasma aldosterone concentration in patients with ADHF treated with RAS inhibitors is unclear. One intriguing hypothesis is that renin itself may play a role in the development of HF via renin receptor-mediated pathways independent of the classical RAS.[27,28]

Some earlier studies reported the clinical significance of plasma active renin concentration (PARC) instead of PRA in HF patients. One study showed that PARC was superior to PRA in predicting outcome. In that study, patients with preserved EF (≥45%) or renal failure (serum creatinine >2.0 mg/dl) were excluded, but such patients were included in the present study. In the present study we did not measure PARC. Therefore, further studies are needed to investigate whether PRA or PARC is a better biomarker for survival.

In the NARA-HF 2 study, as described here, PRA >2.0 ng·ml^{-1}·h^{-1} was not significantly associated with poor prognosis in patients who had not been treated with RAS blockers, not consistent with previous work reported in the 1970s–1990s. At that time therapy with β-blockers as well as RAS blockers was not accepted as an effective therapy for HF. In the present study approximately 20% of patients had been treated with β-blocker, although they had not been treated with RAS blockers. Moreover the RAS blocker and β-blocker treatment was started during hospitalization and continued after discharge. It is possible that these factors more strongly affect the prognosis.

Study Limitations

There are several limitations to this study. The major limitation is that the sample size was moderate, the study was retrospective in nature, and it was based at a single center. We did not collect data on variables that can potentially influence prognosis in ADHF, such as respiratory function and QRS complex width on admission. We could not compare the doses of ACEI or ARB between the 2 groups because there are no official dose conversion formulas for RAS inhibitors.

With respect to PRA, there were also some limitations. First, it is generally recommended that PRA is measured while in the supine position for >30 min, but the supine position might have exacerbated HF in the present patients with emergency admission for ADHF. Therefore, most blood samples were not obtained after 30 min at rest. Second, we did not collect data on factors that could influence PRA, such as sympathetic activity and intravascular volume depletion, because we had no data on catecholamine level or serum osmolality.

Conclusions

PRA is associated with increased risk for all-cause and cardiovascular mortality in ADHF patients on RAS inhibitors, suggesting that PRA is a useful biomarker in ADHF.

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References

1. Pang PS, Komajda M, Gheorghiade M. The current and future management of acute heart failure syndromes. *Eur Heart J* 2010; **31:** 784–793.
2. Bueno H, Ross JS, Wang Y, Chen J, Vidan MT, Normand SL, et al. Trends in length of stay and short-term outcomes among Medicare patients hospitalized for heart failure, 1993–2006. *JAMA* 2010; **303:** 2141–2147.
3. Sato N, Kajimoto K, Keida T, Mizuno M, Minami Y, Yumino D, et al. Clinical features and outcome in hospitalized heart failure in Japan (from the ATTEND Registry). *Circ J* 2013; **77:** 944–951.
4. Schrier RW, Abraham WT. Hormones and hemodynamics in heart failure. *N Engl J Med* 1999; **341:** 577–585.
5. Mann DL, Bristow MR. Mechanisms and models in heart failure: The biomechanical model and beyond. *Circulation* 2005; **111:** 2837–2849.
6. Vergaro G, Emdin M, Iervasi A, Zyw L, Gabutti A, Poletti R, et al. Prognostic value of plasma renin activity in heart failure. *Am J Cardiol* 2011; **108:** 246–251.
7. Latini R, Masson S, Anand I, Salio M, Hester A, Judd D, et al. The comparative prognostic value of plasma neurohormones at baseline in patients with heart failure enrolled in Val-HeFT. *Eur Heart J* 2004; **25:** 292–299.
8. Opie LH. Mechanisms of cardiac contraction and relaxation. *In*: Braunwald E, editor. Heart disease, 7th edn. Philadelphia: Saunders, 2004; 457–489.
9. Garg R, Yusuf S. Overview of randomized trials of angiotensin-converting enzyme inhibitors on mortality and morbidity in patients with heart failure: Collaborative Group on AE Inhibitor Trials. *JAMA* 1995; **273:** 1450–1456.
10. Pfeffer MA, Swedberg K, Granger CB, Held P, McMurray JJ, Michelson EL, et al. Effects of candesartan on mortality and morbidity in patients with chronic heart failure: The CHARM-Overall programme. *Lancet* 2003; **362:** 759–766.
11. The Cardiac Insufficiency Bisoprolol Study II (CIBIS-II): A randomised trial. *Lancet* 1999; **353:** 9–13.
12. Packer M, Coats AJ, Fowler MB, Katus HA, Krum H, Mohacsi P, et al. Effect of carvedilol on survival in severe chronic heart failure. *N Engl J Med* 2001; **344:** 1651–1658.
13. Ueda T, Kawakami R, Horii M, Sugawara Y, Matsumoto T, Okada S, et al. High mean corpuscular volume is a new indicator of prognosis in acute decompensated heart failure. *Circ J* 2013; **77:** 2766–2771.
14. Ho KK, Pinsky JL, Kannel WB, Levy D. The epidemiology of heart failure: The Framingham Study. *J Am Coll Cardiol* 1993; **22:** 6A–13A.
15. Brunner HR, Sealey JE, Laragh JH. Renin as a risk factor in essential hypertension: More evidence. *Am J Med* 1973; **55:** 295–302.
16. Francis GS, Cohn JN, Johnson G, Rector TS, Goldman S, Simon A. Plasma norepinephrine, plasma renin activity, and congestive heart failure: Relations to survival and the effects of therapy in V-HeFT II: The V-HeFT VA Cooperative Studies Group. *Circulation* 1993; **87:** VI40–VI48.
17. Rouleau JL, Packer M, Moye L, de Champlain J, Bichet D, Klein M, et al. Prognostic value of neurohumoral activation in patients with an acute myocardial infarction: Effect of captopril. *J Am Coll Cardiol* 1994; **24:** 583–591.
18. Cohn JN, Levine TB, Olivari MT, Garberg V, Lura D, Francis GS, et al. Plasma norepinephrine as a guide to prognosis in patients with chronic congestive heart failure. *N Engl J Med* 1984; **311:** 819–823.
19. McMurray JJ, Adamopoulos S, Anker SD, Auricchio A, Bohm M, Dickstein K, et al. ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure 2012: The Task Force for the Diagnosis and Treatment of Acute and Chronic Heart Failure 2012 of the European Society of Cardiology: Developed in collaboration with the Heart Failure Association (HFA) of the ESC. *Eur Heart J* 2012; **33:** 1787–1847.
20. Yancy CW, Jessup M, Bozkurt B, Butler J, Casey DE Jr, Drazner MH, et al. 2013 ACCF/AHA guideline for the management of heart failure: A report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines. *J Am Coll Cardiol* 2013; **62:** e147–e239, doi:10.1016/j.jacc.2013.05.019.

21. JCS Joint Working Group. Guidelines for treatment of acute heart failure (JCS 2011): Digest version. *Circ J* 2013; **77:** 2157–2201.
22. van de Wal RM, Plokker HW, Lok DJ, Boomsma F, van der Horst FA, van Veldhuisen DJ, et al. Determinants of increased angiotensin II levels in severe chronic heart failure patients despite ACE inhibition. *Int J Cardiol* 2006; **106:** 367–372.
23. Schroten NF, Gaillard CA, van Veldhuisen DJ, Szymanski MK, Hillege HL, de Boer RA. New roles for renin and prorenin in heart failure and cardiorenal crosstalk. *Heart Fail Rev* 2012; **17:** 191–201.
24. Pitt B, Zannad F, Remme WJ, Cody R, Castaigne A, Perez A, et al. The effect of spironolactone on morbidity and mortality in patients with severe heart failure: Randomized Aldactone Evaluation Study Investigators. *N Engl J Med* 1999; **341:** 709–717.
25. Pitt B, Remme W, Zannad F, Neaton J, Martinez F, Roniker B, et al. Eplerenone, a selective aldosterone blocker, in patients with left ventricular dysfunction after myocardial infarction. *N Engl J Med* 2003; **348:** 1309–1321.
26. Zannad F, McMurray JJ, Krum H, van Veldhuisen DJ, Swedberg K, Shi H, et al. Eplerenone in patients with systolic heart failure and mild symptoms. *N Engl J Med* 2011; **364:** 11–21.
27. Kinouchi K, Ichihara A, Sano M, Sun-Wada GH, Wada Y, Kurauchi-Mito A, et al. The (pro)renin receptor/ATP6AP2 is essential for vacuolar H+-ATPase assembly in murine cardiomyocytes. *Circ Res* 2010; **107:** 30–34.
28. Ando T, Ichihara A. Novel approach to cardiovascular diseases: A promising probability of (pro)renin receptor [(P)RR]. *Curr Pharm Des* 2014; **20:** 2371–2376.

Supplementary Files

Supplementary File 1

Figure S1. Kaplan-Meier event-free survival curves for (**A**) all-cause death and (**B**) cardiovascular death in patients with plasma renin activity (PRA) ≥2.0 ng·ml^{-1}·h^{-1} (blue line, high PRA group; n=180) compared with patients with PRA <2.0 ng·ml^{-1}·h^{-1} (red line, low PRA group; n=113).

Figure S2. Kaplan-Meier event-free survival curves for (**A**) all-cause death and (**B**) cardiovascular death in patients with plasma renin activity (PRA) ≥8.2 ng·ml^{-1}·h^{-1} (blue line, high PRA group; n=90) compared with patients with PRA <8.2 ng·ml^{-1}·h^{-1} (red line, low PRA group; n=203).

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An oxidative stress biomarker, urinary 8-hydroxy-2′-deoxyguanosine, predicts cardiovascular-related death after steroid therapy for patients with active cardiac sarcoidosis

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ABSTRACT

Background: We investigated whether urinary 8-hydroxy-2′-deoxyguanosine (U-8-OHdG), a marker of oxidative DNA damage, is a prognosticator of cardiovascular-related death in patients with cardiac sarcoidosis (CS).

Methods and results: In this prospective study, 30 consecutive patients were divided into the active CS (n = 20) and non-active CS (n = 10) groups, based on abnormal isotope accumulation in the heart on ^{18}F-fluorodeoxyglucose positron-emission tomography/computed tomography (^{18}F-FDG PET/CT) imaging. Nineteen patients in the active CS group underwent corticosteroid therapy. Before corticosteroid therapy initiation, U-8-OHdG, brain natriuretic peptide (BNP), other biomarkers, and indices of cardiac function were measured. Patients were followed-up for a median of 48 months. The primary endpoint was the incidence of cardiovascular-related death.

During the follow-up period, in the corticosteroid-treated active CS group, 7 of 19 patients experienced cardiovascular-related death. By contrast, in the non-active CS group, 1 of 10 patients died from cardiovascular-related causes. Univariate and multivariate analyses showed that U-8-OHdG and BNP were independent predictors for cardiovascular-related death. The cut-off values for predicting cardiovascular death in corticosteroid-treated patients with active CS were 19.1 ng/mg·Cr and 209 pg/mL for U-8-OHdG and BNP, respectively. Patients with a U-8-OHdG concentration ≥19.1 ng/mg·Cr or a BNP concentration ≥209 pg/mL had a significantly higher cardiovascular-related death risk, but U-8-OHdG had better predictive value compared with BNP.

Conclusion: These findings suggested that U-8-OHdG was a powerful predictor of cardiovascular-related death in patients with CS, suggesting that active CS patients with elevated U-8-OHdG levels might be resistant to corticosteroid therapy.

1. Introduction

Sarcoidosis is a multisystem disorder with an unknown etiology characterized by the accumulation of T lymphocytes, mononuclear phagocytes, and non-caseating granulomas in affected tissues [1–3]. The prognosis of patients with cardiac sarcoidosis (CS) is very poor because of progressive heart failure, advanced atrioventricular (AV) block, ventricular tachycardia (VT), and/or ventricular fibrillation [4–8].

Enhanced production of reactive oxygen species may occur in cardiac tissues during the active phase of CS, leading to nucleic and mitochondrial DNA oxidization with subsequent urinary (U) excretion of an oxidized nucleoside of DNA, 8-hydroxy-2′-deoxyguanosine (8-OHdG) [9–17]. Recently, we reported that U-8-OHdG reflected the inflammatory activity of CS [18]. The notion was based on the following aspects: (1) positive immunohistochemical staining for 8-OHdG in CS

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tissue was increased by oxidative stress; (2) increases in serum 8-OHdG in the coronary sinus than in the aortic root indicated that it was produced in cardiac tissues; (3) ^{18}F-fluorodeoxyglucose positron-emission tomography/computed tomography (^{18}F-FDG PET/CT) revealed that U-8-OHdG levels increased in proportion to the inflammatory activity of CS; and (4) U-8-OHdG was a biomarker for therapeutic response to corticosteroid treatment [18].

Despite several studies having evaluated prognostic factors, predicting prognosis in patients with CS remains problematic [5,19, 20]. This is because the CS-induced inflammation changes in response to corticosteroid therapy, and, in addition, CS characteristics change in a spatiotemporal manner. Yazaki et al. reported that patients with active CS had a significantly worse diagnosis when compared to patients with non-active CS [5]. Furthermore, they reported that corticosteroid treatment improved prognosis in patients with active CS [5]. Blankstein et al. reported that, in patients with suspected CS there was a significant association between high-risk cardiac events (death and VT) with abnormal accumulation of ^{18}F-FDG in cardiac tissues, detected on PET/CT images, and cardiac perfusion defects, detected by Rubidium-82 scintigraphy [19]. These studies suggest that the inflammatory activity in cardiac tissue might be associated with poor prognosis in patients with CS.

In our institute, we administer corticosteroid therapy when CS is diagnosed according to CS guidelines and when a patient with CS has complications such as progressive heart failure or lethal arrhythmia according to focal accumulation of ^{18}F-FDG in cardiac tissues. The aim of the present study was to investigate prognostic factors for cardiovascular-related death in patients with active CS who were undergoing corticosteroid therapy.

2. Methods and results

2.1. Patients and study design

Thirty consecutive patients diagnosed with CS (New York Heart Association [NYHA]: classes I, II, and III) at our institution between June 2008 and December 2013 were enrolled in this study. Patients with acute heart failure, acute coronary syndrome, cancer, systemic inflammatory diseases such as infection or collagen disease, severe renal disease (glomerular filtration rate [GFR] <30 mL/min/1.73 m^2), or those who smoked tobacco were excluded.

In the present study, CS was diagnosed according to the Japanese guidelines for diagnosing CS, which were revised in 2006 by the Japan Society of Sarcoidosis and Other Granulomatous Disorder [20–22]. The Japanese guideline for diagnosing CS is as follows.

1. Histological diagnosis group

Cardiac sarcoidosis is confirmed when myocardial biopsy specimens demonstrate noncaseating epithelioid cell granuloma with a histological or clinical diagnosis of extracardiac sarcoidosis.

2. Clinical diagnosis group

Although myocardial biopsy specimens do not demonstrate noncaseating epithelioid cell granuloma, extracardiac sarcoidosis is diagnosed histologically or clinically and satisfies the following conditions and more than one in six basic diagnostic criteria.

(1) ≥2 of 4 major criteria are satisfied.
(2) One in 4 major criteria and ≥2 in 5 minor criteria are satisfied.

Major Criteria

(a) Advanced atrioventricular block.
(b) Basal thinning of the interventricular septum.
(c) Positive cardiac ^{67}Gallium (Ga) uptake or ^{18}F-FDG uptake (Modified Criteria to include PET results).
(d) Depressed left ventricle ejection fraction (LVEF <50%).

Minor Criteria

(a) Abnormal electrocardiography findings: ventricular arrhythmias (ventricular tachycardia, multifocal or frequent PVCs), CRBBB, axis deviation or abnormal Q-wave.
(b) Abnormal echocardiography: Regional abnormal wall motion or morphological abnormality (ventricular aneurysm, wall thickening).
(c) Nuclear medicine: Perfusion defect detected by ^{201}Thallium (Tl) myocardial scintigraphy or ^{99}Technetium (Tc) myocardial scintigraphy.
(d) Gadolinium-enhanced magnetic resonance imaging: Delayed myocardial enhancement.
(e) Endomyocardial biopsy: Moderate grade interstitial fibrosis or monocyte infiltration.

The Institutional Review Board of Yamaguchi University Hospital approved this study (#H19-87) at April 16, 2008, and all patients provided written informed consent before participating in the study.

2.2. Standard clinical evaluation

All patients underwent 12-lead electrocardiography (ECG), echocardiography, ^{18}F-FDGPET/CT [23], and at least 1 dedicated cardiac imaging study, including ^{67}Ga scintigraphy, ^{201}Tl myocardial scintigraphy, ^{99}Tc myocardial scintigraphy, or cardiac gadolinium-enhanced magnetic resonance imaging [24,25]. All patients were monitored by Holter ECG and/or by an ECG monitor (Nihon Kohden, Tokyo). Thirty CS patients underwent coronary angiography and 19 CS patients underwent endomyocardial biopsy.

2.3. Evaluation of inflammatory activity in CS

All patients underwent ^{18}F-FDG PET/CT examination on a 16-slice hybrid PET/CT scanner (Gemini GXL16, Philips Medical System) under identical conditions before corticosteroid therapy with whole-body and cardiac acquisitions as described previously [18]. These patients were divided to the active CS (n = 20) and non-active CS (n = 10) groups based on the ^{18}F-FDG accumulation in heart.

Quantitative analysis of ^{18}F-FDG uptake in the lesion was based on the maximum standardized uptake value per focus. This value was calculated as the activity concentration measured at the end of the scan and corrected for individual body weight and dose injected, as follows: tissue activity (counts/pixel/s) multiplied by calibration factor divided by injected ^{18}F-FDG dose (MBq/kg of body weight). The extent of abnormal FDG uptake area within the myocardium was quantified using the automated software (Emory Cardiac Toolbox, Version 3.0, Royal Philips Electronics Co. Tokyo, Japan). After each image volume was automatically reoriented to standard left ventricle short-axis orientation, followed by automated segmentation of the left ventricular myocardium, a constrained spline model was fit to generate the left ventricle polar map regions of interest. On the polar map, the extent (%) of the pixels with standardized uptake value (SUV) over the cut-off level of 50% or 70% for the maximum SUV (SUV max) of the whole pixels was automatically calculated. SUV was defined as the ratio of Bq/mL (decay-corrected) in tissue against Bq of tracer injected/body weight.

2.4. Corticosteroid treatment protocols for patients with active CS

Nineteen of the 20 patients with abnormal accumulation of ^{18}F-FDG in cardiac tissues, indicative of active CS, were administered prednisolone

according to existing Japanese protocols as follows: 30 mg/day for 4 weeks with gradual tapering of the dose to 5–10 mg every other day over 6 months to establish the minimal effective dose [8].

2.5. Endpoints and follow-up

No patients were lost to follow-up (mean follow-up, 48 months). Events were centrally adjudicated using medical records, autopsy reports, and death certificates. The primary endpoint was defined as cardiovascular-related death due to worsening heart failure or sudden cardiac death. Sudden cardiac death, established by the attending physician, was defined as death without definite premonitory signs or symptoms.

2.6. 8-OHdG measurement and other neurohumoral and inflammatory factors

Serum and U-8-OHdG concentrations were measured using enzyme-linked immunosorbent assay (ELISA) kit (Japan Institute for the Control of Aging, Fukuroi, Japan) with anti-8-OHdG antibody (N45.1) as described previously [16–18]. Serum interleukin (IL)-6 concentration was determined using a human IL-6 Quantikine® HS ELISA kit (Quantikine® HS; R&D Systems, Minneapolis, MN, USA) and a human IL-6 chemiluminescence enzyme immunoassay (Fujirebio, Tokyo, Japan) as described previously [16–18]. U-8-isoprostane levels were determined using an 8-isoprostane enzyme immunoassay kit (Cayman Chemical Co., Ann Arbor, MI, USA), as described previously [16–18]. Raw values were normalized to the urinary level of creatinine (Cr). Plasma brain natriuretic peptide (BNP) was measured using the Shionoria BNP Kit (Shionogi Pharmaceutical, Osaka, Japan) as describe previously [16–18].

2.7. Histological assessment of inflammation and fibrosis of hearts obtained from two autopsy cases

Histological examination of the heart tissue was performed at autopsy in two of seven patients with CS after cardiovascular death. Each sample (from the septal, anterior, lateral, posterior, and inferior walls)

Table 1
Clinical information of all included CS patients.

No.	Age	Sex	NYHA class	ECG/Holter findings	Echocardiography		^{18}F-FDG PET	CMR	CAG	Type of diagnosis	Clinical diagnosis		Cardio-vascular death	Autopsy
					LVEF	Basal wall thinning of IVS and PW					Major criteria	Minor criteria		
1	74	M	II	AVB, SVT	15	+	Focal: IVS, Ant wall	Not done	Intact	CD	4	2	+	Not done
2	73	F	II	SVT	30	+	Focal on diffuse: A-S, Lat, Inf wall	Not done	Intact	CD	3	2	+	Not done
3	77	M	II	SVT	28	+	Focal: IVS, Lat wall	Not done	Intact	CD	3	2	+	Done
4	74	M	I	AVB, SVT	40	+	Focal on diffuse: IVS, Lat, Inf wall	Not done	Intact	CD	4	2	+	Done
5	73	M	I	AVB, SVT	48	+	Focal on diffuse: A-S, Lat, Inf wall	Not done	Intact	CD	4	2	+	Not done
6	67	M	II	SVT	27	+	Focal on diffuse: A-S, Pos, Inf wall	Not done	#7 75%	CD	3	2	+	Not done
7	59	F	II	SVT	35	+	Focal: Lat, Pos wall	Not done	Intact	CD	3	2	+	Not done
8	70	F	II	AVB, PVC	45	+	Focal on diffuse: Ant, Lat wall	Not done	Intact	CD	4	2	−	
9	76	F	II	PVC	43	+	Focal: Ant wall	Not done	Intact	CD	3	2	−	
10	59	F	II	AVB	55	+	Focal on diffuse: A-S, Pos, Inf, RV wall	Not done	Intact	CD	2	2	−	
11	68	F	II	SVT	52	+, VA	Focal on diffuse: A-S, Lat, Pos wall	LGE: RV, IVS, Ant wall	Intact	CD	1	3	−	
12	56	M	II	AVB, PVC	25	+	Focal on diffuse: IVS, Lat, Pos, Inf wall	Not done	Intact	CD	3	2	−	
13	67	F	II	SVT	30	−	Focal on diffuse: A-S, Pos, RV wall	LGE: RV, IVS, Lat, Apex wall	Intact	CD	2	3	−	
14	46	F	I	SVT	28	+	Focal on diffuse: A-S, Pos, wall	LGE: IVS, Lat, Apex wall	Intact	CD	3	3	−	
15	78	M	I	AVB, SVT	35	+	Focal: IVS, Ant wall	LGE(−)	Intact	HD	4	2	−	
16	69	M	II	PVC, abnormal Q	30	+	Focal on diffuse: Ant, Lat wall	Not done	Intact	HD	3	2	−	
17	59	F	I	SVT	32	+	Focal on diffuse: Ant, Lat wall	Not done	Intact	CD	3	2	−	
18	56	F	I	SVT	37	+	Focal on diffuse: A-S, Pos, Inf wall	LGE: RV, IVS, Lat, Apex wall	Intact	CD	3	3	−	
19	57	F	I	AVB, PVC	60	+	Focal: A-S, Inf wall	LGE: RV, IVS	Intact	CD	3	3	−	
20	78	F	I	AVB, PVC	50	−	Focal: IVS, Ant wall	Not done	Intact	CD	2	2	−	
21	53	M	II	AVB, SVT	15	+	-	Not done	Intact	CD	3	2	−	
22	74	F	II	AVB, SVT	25	−	-	Not done	Intact	CD	2	2	−	
23	63	M	II	AVB, SVT	45	−	-	Not done	Intact	CD	2	2	−	
24	72	F	II	SVT	30	+	-	Not done	Intact	CD	2	2	+	Not done
25	79	M	II	PVC	25	+	-	Not done	Intact	CD	2	2	−	
26	66	M	II	PVC	18	+	-	Not done	Intact	CD	2	2	−	
27	53	M	II	AVB, SVT	20	−	-	Not done	Intact	CD	2	2	−	
28	27	M	II	AVB, PVC	67	−	-	Not done	Intact	CD	1	2	−	
29	54	M	I	AVB, SVT	25	+	-	Not done	Intact	CD	3	2	−	
30	65	F	II	PVC	25	+	-	Not done	Intact	CD	2	2	−	

NYHA, New York Heart Association; ECG, electrocardiogram; AVB, atrioventricular block; SVT, sustained ventricular tachycardia; LVEF, left ventricular ejection fraction; VA, ventricular aneurysm; IVS, interventricular septum; PW, posterior wall; Ant, anterior; Lat, lateral; A-S, anteroseptal; Pos, posterior; CMR, cardiac magnetic resonance imaging; LGE, late gadolinium enhancement; CAG, coronary angiography; CD, clinical diagnosis; HD, histological diagnosis.

Table 2
Baseline clinical characteristics all patients with cardiac sarcoidosis.

	Total (n = 30)	Non-active CS (n = 10)	Active CS (n = 20)	P value
Sex (M/F)	15/15	7/3	8/12	0.121
Age (years)	65 ± 11	61 ± 14	67 ± 9	0.194
BMI (kg/m^2)	21.1 ± 2.4	21.3 ± 1.5	21.0 ± 2.6	0.451
BSA(m^2)	1.484 ± 0.158	1.523 ± 0.110	1.466 ± 0.172	0.220
NYHA class	1.8 ± 0.6	1.9 ± 0.5	1.8 ± 0.6	0.495
SBP (mm Hg)	108 ± 15	106 ± 16	108 ± 13	0.270
Heart rate (bpm)	66 ± 8	67 ± 6	65 ± 9	0.548
LVDd (mm)	60 ± 10	65 ± 10	58 ± 10	0.085
LVEF (%)	36 ± 14	30 ± 15	38 ± 13	0.026
BNP (pg/mL)	309.5 ± 336.5	308.3 ± 300.5	310.0 ± 349.9	0.660
U-8-OHdG (ng/mg·Cr)	15.8 ± 6.4	10.3 ± 1.6	18.0 ± 6.3	<0.001
U-8-ISO (ng/mg·Cr)	208 ± 84	247 ± 89	197 ± 79	0.308
UA (mg/dL)	6.0 ± 2.1	6.7 ± 2.1	5.9 ± 2.1	0.314
CRP (mg/dL)	0.30 ± 0.72	0.06 ± 0.06	0.37 ± 0.79	0.066
TNFα (pg/mL)	2.1 ± 1.4	1.7 ± 1.1	2.2 ± 1.5	0.560
IL-6 (pg/mL)	4.1 ± 3.6	3.4 ± 3.1	4.3 ± 3.8	1.000
BUN (mg/dL)	21 ± 8	20 ± 5	21 ± 9	0.981
eGFR (mL/min/1.73 m^2)	58.7 ± 18.5	62.2 ± 14.1	58.1 ± 20.7	0.741
TnT (ng/mL)	0.043 ± 0.04	0.013 ± 0.005	0.048 ± 0.37	0.191

Data given as mean ± SD. CS, cardiac sarcoidosis; BMI, body mass index; BSA, body surface area; NYHA, New York Heart Association; SBP, systolic blood pressure; LVDd, left ventricular end-diastolic diameter; LVEF, left ventricular ejection fraction; BNP, brain natriuretic peptide; U-8-OHdG, urinary 8-hydroxy-2′-deoxyguanosine; U-8-ISO, urinary 8-isoprostane; UA, uric acid; CRP, C-reactive protein; TNF-α, tumor necrosis factor-α; IL-6, interleukin-6; BUN, blood urea nitrogen; eGFR, estimated glomerular filtration rate; TnT, troponin T.
The normal range of the U-8-OHdG level is <10 ng/mg·Cr, as taken from a previous study [16].

Table 3
Characteristics of corticosteroid-treated active CS patients in terms of survival outcome.

	Active CS on corticosteroids (n = 19)	Cardiovascular-related death (n = 7)	Survival (n = 12)	P value
Sex (M/F)	8/11	5/2	3/9	0.048
Age (years)	66 ± 9	71 ± 6	64 ± 9	0.082
BSA(m^2)	1.480 ± 0.164	1.558 ± 0.108	1.435 ± 0.173	0.076
NYHA class	1.7 ± 0.6	2.0 ± 0.6	1.5 ± 0.5	0.081
LVDd (mm)	59 ± 9	66 ± 10	55 ± 6	0.022
LVEF (%)	37 ± 11	32 ± 10	39 ± 11	0.189
BNP (pg/mL)	303.2 ± 373.3	532.3 ± 523.0	169.6 ± 112.1	0.063
U-8-OHdG (ng/mg·Cr)	18.0 ± 6.3	24.3 ± 4.6	16.8 ± 5.3	0.009
U-8-ISO (ng/mg·Cr)	203 ± 75	226 ± 72	187 ± 73	0.439
UA (mg/dL)	5.7 ± 1.8	5.6 ± 1.3	5.7 ± 2.0	0.899
CRP (mg/dL)	0.38 ± 0.81	0.38 ± 0.38	0.36 ± 0.98	0.175
TNFα (pg/mL)	1.6 ± 0.6	1.9 ± 0.5	1.4 ± 0.6	0.136
IL-6 (pg/mL)	4.0 ± 3.0	5.8 ± 3.5	2.6 ± 1.6	0.081
BUN (mg/dL)	21 ± 10	29 ± 11	17 ± 5	0.022
eGFR (mL/min/1.73 m^2)	61.3 ± 18.8	44.3 ± 12.3	71.3 ± 14.2	0.004
ACE (U/L)	13.4 ± 7.8	9.5 ± 4.5	15.5 ± 8.4	0.366
TnT (ng/mL)	0.049 ± 0.053	0.059 ± 0.012	0.044 ± 0.052	0.111
SUV max	6.63 ± 2.63	6.49 ± 2.59	6.71 ± 2.65	0.871
SUV over 50%	0.61 ± 0.24	0.73 ± 0.22	0.54 ± 0.22	0.115
SUV over 70%	0.20 ± 0.11	0.18 ± 0.06	0.21 ± 0.13	0.828
Risk factor				
Hypertension	7/19	4/7	3/12	0.161
Diabetes mellitus	3/19	2/7	1/12	0.243
Dyslipidemia	13/19	6/7	7/12	0.216
Symptom				
Sustained VT	12/19	6/7	6/12	0.120
Advanced AVB	3/19	1/7	2/12	0.891
Only HF	4/19	0/7	4/12	0.086
Treatment				
β-Blocker	19/19	7/7	12/12	NS
ACEI/ARB	13/19	4/7	9/12	0.419
Loop diuretics	14/19	6/7	8/12	0.363
Aldosterone antagonist	5/19	2/7	3/12	0.865
Statin	13/19	6/7	7/12	0.216
Steroid	19/19	7/7	12/12	NS
CRT-D/ICD	15/19	7/7	8/12	0.086
DDD PM	3/19	0/7	3/12	0.149

Numerical data are expressed as mean ± SD.
CS, cardiac sarcoidosis; BSA, body surface area; NYHA, New York Heart Association; LVDd, left ventricular end-diastolic diameter; LVEF, left ventricular ejection fraction; BNP, brain natriuretic peptide; U-8-OHdG, urinary 8-hydroxy-2′-deoxyguanosine; U-8-ISO, urinary 8-isoprostane; UA, uric acid; CRP, C-reactive protein; TNF-α, tumor necrosis factor-α; IL-6, interleukin-6; BUN, blood urea nitrogen; eGFR, estimated glomerular filtration rate; ACE, angiotensin-converting enzyme; TnT, troponin T; SUV max, the maximum standardized uptake value; SUV over 50%, the extent of the pixels with SUV over the cut-off level of 50% for SUV max of the whole pixels; SUV over 70%, the extent of the pixels with SUV over the cut-off level of 70% for SUV max of the whole pixels; VT, ventricular tachycardia; AVB, atrioventricular block; HF, heart failure; ACEI, angiotensin-converting enzyme inhibitor; ARB, angiotensin receptor blockers; CRT-D, cardiac resynchronization therapy defibrillator; ICD, implantable cardioverter-defibrillator; DDD PM, DDD pacemaker; NS, not significant among three groups.
The normal range of the U-8-OHdG level is <10 ng/mg·Cr, as taken from a previous study [16].

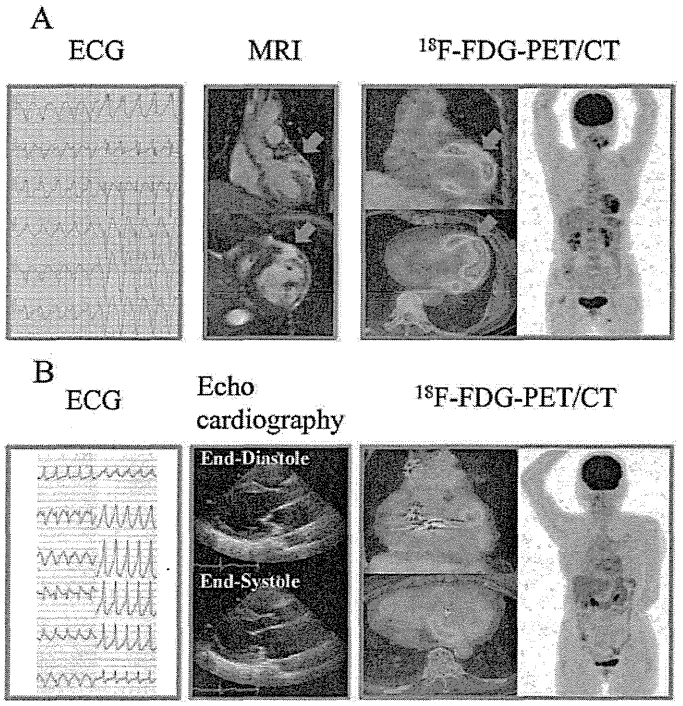

Fig. 1. Representative CS patients with and without cardiovascular death. A. A CS patient without cardiovascular-related death (No.11). Ventricular tachycardia (electrocardiography [ECG]) and left ventricular aneurysm at the left ventricle anterior wall (magnetic resonance imaging [MRI]) that were present on admission are shown. ^{18}F-FDG PET shows strong ^{18}F-FDG accumulation around the aneurysm on the left ventricle anterior and posterior walls (See Supplementary video). B. A CS patient with cardiovascular death (No.7). Ventricular tachycardia (on ECG) and wall thinning of the base of the interventricular septum (echocardiography) on admission are shown. The ^{18}F-FDG PET image shows abnormal accumulation at the left ventricle anterior and posterior walls.

obtained from the left ventricle was fixed in 10% formalin and embedded in paraffin. Sections 4-μm thick were cut and stained with hematoxylin–eosin. Then, the histological findings were assessed by microscopy.

2.8. Statistical analyses

All results are expressed as the mean ± standard deviation (SD). Categorical variables are presented using frequency counts and percentages, and intergroup comparisons were analyzed using the χ^2 test. The differences between the groups were detected using the Mann–Whitney U test, with 2-tailed P values <0.05. The prediction of cardiac events was tested using Cox proportional hazard regression analysis with categorized variables dichotomized at the cut-off levels. Multivariate Cox proportional hazard analysis was performed using stepwise regression. The cut-off level was defined by the maximal point of (sensitivity plus specificity) on receiver operating characteristic (ROC) curve analysis. Kaplan–Meier analysis was performed on the cumulative rates of cardiac event-free status in patients with divided into 2 groups based on the cut-off levels of BNP and U-8-OHdG. Differences between the cardiac event-free curves were analyzed using the log-rank test. All analyses were performed using SPSS version 19 (SPSS, Inc., Chicago, Illinois). P-values <0.05 were considered statistically significant.

2.9. Baseline patient characteristics

Table 1 showed the clinical information of all included CS patients. Twenty-eight out of 30 CS patients belong to clinical diagnosis group, while 2 CS patient belongs to histological diagnosis group due to endomyocardial biopsy, which was performed in 19 CS patients. Patient Nos.1–20 were diagnosed with active CS, based on abnormal accumulation of ^{18}F-FDG in cardiac tissue as well as clinical features and other cardiac imaging findings, while Nos.21–30 were diagnosed with non-active CS. Patient Nos.1–19 underwent corticosteroid therapy, complicated with ventricular arrhythmia and/or advanced atrioventricular block, and progressive heart failure. Patient No.20 rejected corticosteroid therapy. In the present study, coronary angiography showed that coronary artery was intact in all patients with CS, except 1 patient (No.6), and 2 CS patients were histologically diagnosed with CS by endomyocardial biopsy. During the follow-up period for a median 48 months, 7 of 19 patients experienced cardiovascular-related death in the corticosteroid-treated active CS group. By contrast, 1 of 10 patients died from cardiovascular-related causes in the non-active CS group. Table 2 shows the baseline patient characteristics of all the patients with CS. Patients were subdivided into the non-active CS (n = 10) and the active CS (n = 20) groups, depending on the absence or presence of abnormal ^{18}F-FDG accumulation in cardiac tissues, respectively. LVEF and U-8-OHdG levels were significantly higher in the active CS group compared with the non-active CS group.

2.10. Characteristics of corticosteroid-treated patients with active CS in terms of survival outcome

All patients in the active CS group, except 1 patient (No.20) who refused the therapy, were administered with prednisolone. The patient who refused therapy was excluded from subsequent analyses. Seven patients in the CS group experienced cardiovascular-related death during follow-up, whereas only 1 of those in the non-active CS group died due to cardiovascular-related causes. Fifteen out of 19 patients with active sarcoidosis had implanted implantable cardioverter-defibrillator (ICD) or cardiac resynchronization therapy defibrillator (CRTd) before starting corticosteroid therapy (Table 3). Therefore, there was no cardiac sudden death due to lethal arrhythmia, even though 3 out of 15 ICD- or CRTd-implanted patients had appropriate ICD therapy including anti-tachycardia pacing and/or electrical defibrillation during the follow-up. Fig. 1 shows representative imaging data for patients with and without cardiovascular-related death. Table 3 shows the baseline characteristics of active CS patients, who underwent corticosteroid treatment, in terms of survival outcome. Patients who experienced cardiovascular-related death were significantly more likely to be male, with higher blood urea nitrogen (BUN) and U-8-OHdG levels, higher estimated GFRs,

Table 4
Risk factors associated with cardiovascular-related death in corticosteroid-treated patients with active cardiac sarcoidosis.

	Univariate		Multivariate				
	HR	(95% CI)	P value	HR	(95% CI)	P value	Wald coefficient
U-8-OHdG (ng/mg·Cr)	1.130	(1.010–1.265)	0.033	1.203	(1.030–1.407)	0.020	5.408
BNP (pg/mL)	1.002	(1.000–1.004)	0.023	1.003	(1.000–1.006)	0.028	4.817
BUN (mg/dL)	1.114	(1.034–1.201)	0.004				
eGFR (mL/min/1.73 m^2)	0.925	(0.873–0.980)	0.008				
LVDd (mm)	1.141	(1.032–1.260)	0.010				

Abbreviations: HR, hazard ratio; CI, confidence interval; U-8-OHdG, urinary 8-hydroxy-2′-deoxyguanosine; BNP, brain natriuretic peptide; BUN, blood urea nitrogen; eGFR, estimated glomerular filtration rate; LVDd, left ventricular end-diastolic diameter.

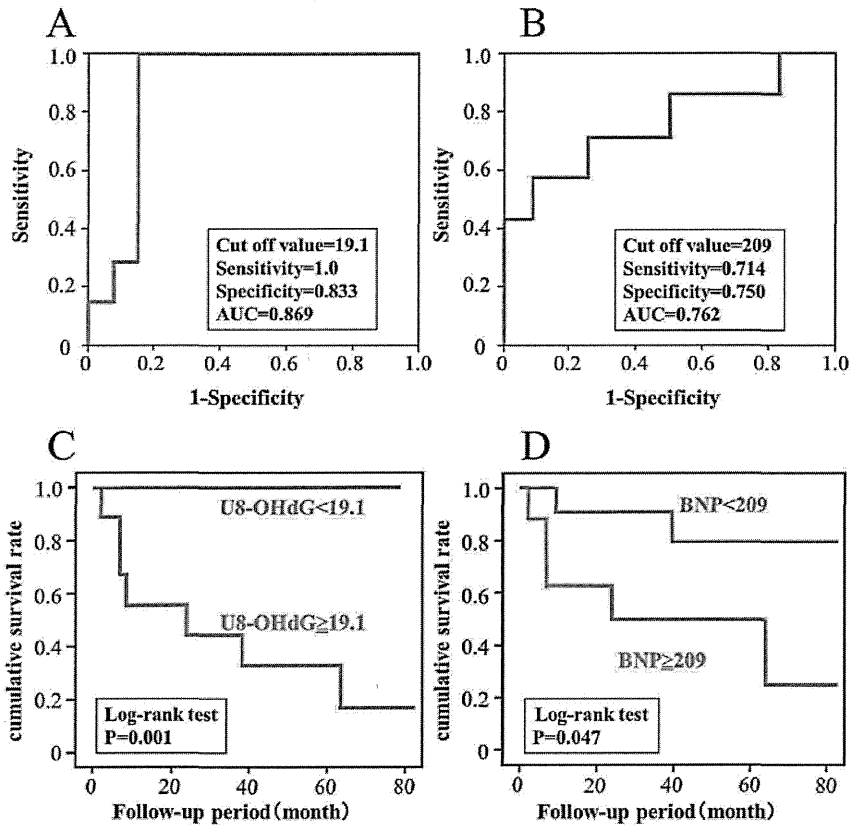

Fig. 2. Urinary 8-hydroxy-2′-deoxyguanosine and plasma brain natriuretic peptide as predictors of cardiovascular-related death. A, Receiver operating characteristic curve analysis of urinary 8-hydroxy-2′-deoxyguanosine (U-8-OHdG). For the diagnosis of active cardiac sarcoidosis, the cut-off level for U-8-OHdG was 19.1 ng/mg·Cr with a sensitivity of 100.0% and a specificity of 83.3%. AUC, area under the curve. B, Receiver operating characteristic curve analysis of brain natriuretic peptide (BNP). For the diagnosis of active cardiac sarcoidosis, the cut-off level for BNP was 209 pg/mL Cr, with a sensitivity of 71.4% and a specificity of 75.0%. AUC, area under the curve. C, Kaplan–Meier curves for patients with active CS divided into 2 groups based on the cut-off value for U-8-OHdG (19.1 ng/mg·Cr). D, Kaplan–Meier curves for patients with CS divided into 2 groups based on the cut-off value for BNP (209 pg/mL).

and an increased incidence of left ventricle diastolic dysfunction (LVDd) compared with patients who survived. There was no difference in SUV max, over 50% and SUV over 70% (the extent (%) of the pixels with standardized uptake value (SUV) over the cut-off level of 50% or 70% for the maximum SUV of the whole pixels) between cardiovascular related death group and survivor group (Table 3). In addition, there was no correlation between SUV max and U-8-OHdG ($p = 0.251$), SUV over 50% and U-8-OHdG ($p = 0.108$), and SUV over 70% and U-8-OHdG ($p = 0.472$) in 19 active CS patients. Similarly, there was no correlation between SUV max and BNP ($p = 0.444$), SUV over 50% and BNP ($p = 0.201$), and SUV over 70% and BNP ($p = 0.833$) in 19 active CS patients.

2.11. Univariate and multivariate analyses of factors associated with cardiovascular-related death in corticosteroid-treated patients with active CS

Table 4 shows the univariate and multivariate analyses of factors associated with cardiovascular-related death in corticosteroid-treated active CS patients. In the univariate analysis, U-8-OHdG, BNP, blood urea nitrogen, estimated GFR, and LVDd were significantly associated with cardiovascular-related death. Age, Sex, BSA, NYHA class, LVDd, LVEF, eGFR, U-8-OHdG, BNP, CRP, IL-6, SUV max, SUV over 50%, and SUV over 70% were entered in the multivariate model, and Cox proportional model was developed using the stepwise downward method. Finally, U-8-OHdG and BNP remained as independent predictor for cardiovascular-related death in the model. Therefore, elevated U-8-OHdG and elevated plasma BNP were significant independent predictors for cardiovascular-related death (U-8-OHdG: hazard ratio [HR], 1.203; 95% confidence interval [CI], 1.030–1.407; BNP: HR, 1.003; 95% CI, 1.000–1.006).

2.12. Determination of the optimal cut-off values of U-8-OHdG and BNP as prognosticators of cardiovascular-related death in corticosteroid-treated patients with active CS

The optimal cut-off values of U-8-OHdG and BNP were determined as those with the largest sum of sensitivity plus specificity on each ROC curve (Fig. 2A and B, respectively). The cut-off level for U-8-OHdG was 19.1 ng/mg·Cr, which gave a sensitivity of 100.0% and a specificity of 83.3%. That for BNP was 209 pg/mL, which yielded a sensitivity of 71.4% and a specificity of 75.0%.

2.13. Kaplan–Meier analysis of cardiac event-free status based on U-8-OHdG and plasma BNP levels

Corticosteroid-treated patients with active CS were subdivided into groups based on cut-off values for U-8-OHdG (19.1 ng/mg·Cr) and plasma BNP (209 pg/mL). Kaplan–Meier cumulative cardiac event-free curves revealed that patients with U-8-OHdG levels ≥19.1 ng/mg·Cr or with BNP levels ≥209 pg/mL had a significantly higher cardiac event rate (Log rank test; $P = 0.001$ for U-8-OHdG and $P = 0.047$ for BNP) (Fig. 2C and D, respectively).

2.14. Histological assessment of the inflammation and fibrosis in hearts obtained from the two autopsy cases

Fig. 3 shows the samples of the heart stained with hematoxylin–eosin from two autopsy cases after corticosteroid therapy. The samples of cases 1 and 2 showed broad and multiple replacement fibrosis with scars as well as stenotic lesions at the intramuscular small arteries due

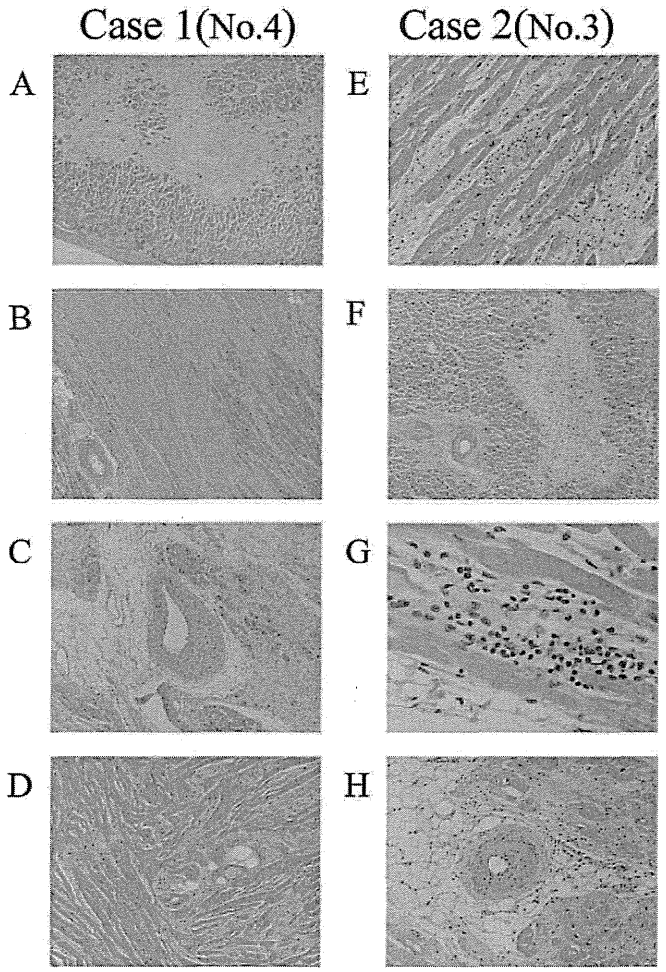

Fig. 3. Case 1 (No.4). A. Multiple and broad replacement fibroses (hematoxylin–eosin [H.E.] staining ×100). B. Scars and replacement fibrosis (H.E. staining ×100). C. Inflammatory intimal thickening at the intramuscular small artery, which is characteristic of cardiac sarcoidosis (H.E. staining ×100). D. The disarray of cardiomyocytes suggests that the myocardial remodeling is accompanied with myocyte death (H.E. staining ×100). Case 2 (No.3). E. Inflammatory cell infiltration and myocytolysis (H.E. staining ×200). F. Multiple and broad replacement fibroses with inflammatory cell infiltration and inflammatory intimal thickening (H.E. staining ×100). G. Acute inflammatory findings with lymphocyte infiltration (H.E. staining ×400). H. Inflammatory intimal thickening at the intramuscular small artery (H.E. staining ×200).

to inflammatory intimal thickening. These histological findings were indeed not specific but characteristic for CS (Fig. 3A–H). In addition, in case 2, multiple and broad replacement fibroses contained inflammatory cell infiltration and myocytolysis was occasionally observed, suggesting residual inflammatory activity (Fig. 3E–H). Interestingly, the U-8-OHdG levels before corticosteroid therapy were markedly elevated (U-8-OHdG: 27.6 ng/mg·Cr in case 1 and 21.3 ng/mg·Cr in case 2).

3. Discussion

The most important finding of the present study was that U-8-OHdG, a biomarker of oxidative stress, was a significant independent factor for predicting cardiovascular-related death in corticosteroid-treated patients with active CS. This notion was supported by the following findings. (1) During the follow-up period, the cardiovascular-related death rate was significantly higher in corticosteroid-treated patients with active CS compared to that in patients with non-active CS (37% vs. 10%, respectively). (2) In corticosteroid-treated patients with active CS, the U-8-OHdG level was higher in those who experienced cardiovascular-related death CS compared with those who did not. However, there was no significant difference between those who died and those who did not in terms of cardiac function (NYHA class, LVEF, and plasma BNP levels) or biomarkers (serum IL-6, TNFα, high-sensitivity C-reactive protein, angiotensin-converting enzyme, troponin T). (3) U-8-OHdG and plasma BNP were identified as significant independent predictors of cardiovascular-related death. (4) Patients with values above the cut-off levels for U-8-OHdG (≥19.1 ng/mg·Cr) and BNP (≥209 pg/mL) had a significantly high risk of cardiovascular-related death compared to those with values below the cut-off. However, according to ROC and Kaplan–Meier analyses, U-8-OHdG was thought to be superior to BNP for the prediction of cardiovascular-related death.

Yazaki et al. reported that steroid therapy improved the prognosis of patients with CS [5]. In addition, Nagai et al. suggested that corticosteroid therapy was associated with fewer long-term adverse events such as cardiac death and heart failure-associated readmission to hospital [7]. Together these studies support a critical role for corticosteroid therapy in inhibiting the progression of cardiac tissue damage caused by inflammation [26]. In corticosteroid-treated patients with CS, Blankstein et al. demonstrated that the presence of abnormal accumulation of ^{18}F-FDG and cardiac perfusion defects predicted cardiac death and VT [19], suggesting that myocardial inflammation and subsequent myocardial damage contributed to the worsening of prognosis in patients with active CS. Our results are consistent with those findings because increases in U-8-OHdG are caused by myocardial oxidative stress induced by abnormal intracellular Ca^{2+} handling and apoptosis, leading to cardiac remodeling, dysfunction, and arrhythmogenicity [27–31]. Because there were no differences in NYHA class, LVEF and BNP between thecardiovascular-related group and the survival group before the start of corticosteroid therapy, our findings suggest that patients with higher U-8-OHdG levels might be resistant to corticosteroid therapy compared to those with lower U-8-OHdG levels.

The histological findings of the heart obtained from two autopsy cases of patients with CS after corticosteroid therapy showed broad and multiple fibroses with scars as well as stenotic lesions at the intramuscular small arteries due to inflammatory intimal thickening (Fig. 3A–H). These findings were consistent with the pathohistological findings of CS, although no non-caseous epithelioid cell granuloma was observed [32–34]. In both cases, the U-8-OHdG levels increased significantly before steroid therapy. This strongly supports the notion that patients with CS with higher myocardial oxidative stress may be resistant to steroid therapy, although it was impossible to clarify whether an increase in myocardial oxidative stress was a cause or a result of myocardial remodeling. Thus, an elevation in U-8-OHdG in CS patients might represent the cardiac remodeling as well as inflammation in cardiac tissue.

Recently, it was reported that extension and intensity of ^{18}F-FDG accumulation in cardiac tissue were associated with occurrence of arrhythmia such as ventricular tachycardia and complete atrioventricular block in patients with CS by ^{18}F-FDG PET/CT [20]. However, because inflammatory activity may be changing according to corticosteroid therapy, it would be difficult to predict prognosis based on the only extent of ^{18}F-FDG accumulation in cardiac tissue before starting corticosteroid therapy. Therefore, combination of ^{18}F-FDG PET/CT and U-8-OHdG may be more useful for predicting cardiovascular related death in corticosteroid-treated CS patients.

The present study has limitations. First, the population evaluated was a small single cohort. A larger study is warranted to confirm whether U-8-OHdG is a useful clinical marker for predicting cardiovascular-related death in corticosteroid-treated patients with active CS. Second, sudden death due to lethal arrhythmia was not included in the present study. Of the 7 patients in the active CS group who died, all experienced progressive heart failure. We believe that this is because 15 of the 19 active CS patients had a defibrillator device implant (ICD or CRTd). Third, there was significant difference in Sex, eGFR between cardiovascular death group and survival group, and level of IL-6 tended to be higher in cardiovascular death group than in survival group.

However, these factors adjusted for U-8-OHdG and BNP by multivariate analysis.

4. Conclusion

Levels of U-8-OHdG before corticosteroid therapy might be a useful biomarker for predicting cardiovascular death after corticosteroid therapy in patients with active CS. Furthermore, the U-8-OHdG level may provide relevant tissue information that can clarify the pathophysiology of CS.

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Conflict of interest

The authors have no conflicts of interest to declare.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.ijcard.2016.03.003.

References

[1] R.H. Swanton, Sarcoidosis of the heart, Eur. Heart J. 9 (Supplement G) (1988) 169–174.
[2] M.C. Iannuzzi, B.A. Rybicki, A.S. Teirstein, Sarcoidosis, N. Engl. J. Med. 357 (2007) 2153–2165.
[3] R.P. Baughman, E.E. Lower, R.M. du Bois, Sarcoidosis, Lancet 361 (2003) 1111–1118.
[4] Y. Yoshida, S. Morimoto, S. Hiramitsu, N. Tsuboi, H. Hirayama, T. Itoh, Incidence of cardiac sarcoidosis in Japanese patients with high-degree atrioventricular block, Am. Heart J. 134 (1997) 382–386.
[5] Y. Yazaki, M. Isobe, M. Hiroe, S. Morimoto, S. Hiramitsu, T. Nakano, et al., Prognostic determinants of long-term survival in Japanese patients with cardiac sarcoidosis treated with prednisone, Am. J. Cardiol. 88 (2001) 1006–1010.
[6] H. Furushima, M. Chinushi, H. Sugiura, H. Kasai, T. Washizuka, Y. Aizawa, Ventricular tachyarrhythmia associated with cardiac sarcoidosis: its mechanisms and outcome, Clin. Cardiol. 27 (2004) 217–222.
[7] T. Nagai, N. Nagano, Y. Sugano, Y. Asaumi, T. Aiba, H. Kanzaki, et al., Effect of corticosteroid therapy on long-term clinical outcome and left ventricular function in patients with cardiac sarcoidosis, Circ. J. 79 (2015) 1593–1600.
[8] R. Kandolin, J. Lehtonen, J. Airaksinen, T. Vihinen, H. Miettinen, K. Ylitalo, et al., Cardiac sarcoidosis: epidemiology, characteristics, and outcome over 25 years in a nationwide study, Circulation 131 (2015) 624–632.
[9] K. Psathakis, G. Papatheodorou, M. Plataki, P. Panagou, S. Loukides, N.M. Siafakas, et al., 8-Isoprostane, a marker of oxidative stress, is increased in the expired breath condensate of patients with pulmonary sarcoidosis, Chest 125 (2004) 1005–1011.
[10] T. Kaneko, S. Tahara, M. Matsuo, Non-linear accumulation of 8-hydroxy-2′-deoxyguanosine, a marker of oxidized DNA damage, during aging, Mutat. Res. 316 (1996) 277–285.
[11] B.N. Ames, Endogenous oxidative DNA damage, aging, and cancer, Free Radic. Res. Commun. 7 (1989) 121–128.
[12] S. Loft, K. Vistisen, M. Ewertz, A. Tjonneland, K. Overvad, H.E. Poulsen, Oxidative DNA damage estimated by 8-hydroxydeoxyguanosine excretion in humans: influence of smoking, gender and body mass index, Carcinogenesis 13 (1992) 2241–2247.
[13] T. Ide, H. Tsutsui, S. Hayashidani, D. Kang, N. Suematsu, K. Nakamura, et al., Mitochondrial DNA damage and dysfunction associated with oxidative stress in failing hearts after myocardial infarction, Circ. Res. 88 (2001) 529–535.
[14] H. Tsutsui, T. Ide, T. Shiomi, D. Kang, S. Hayashidani, N. Suematsu, et al., Oxidative stress mediates tumor necrosis factor-alpha-induced mitochondrial DNA damage and dysfunction in cardiac myocytes, Circulation 104 (2001) 2883–2885.
[15] Y. Kono, K. Nakamura, H. Kimura, N. Nishii, A. Watanabe, K. Banba, et al., Elevated levels of oxidative DNA damage in serum and myocardium of patients with heart failure, Circ. J. 70 (2006) 1001–1005.
[16] S. Kobayashi, T. Susa, T. Tanaka, Y. Wada, S. Okuda, M. Doi, et al., Urinary 8-hydroxy-2′-deoxyguanosine reflects symptomatic status and severity of systolic dysfunction in patients with chronic heart failure, Eur. J. Heart Fail. 13 (2011) 29–36.
[17] T. Susa, S. Kobayashi, T. Tanaka, W. Murakami, S. Akashi, I. Kunitsugu, et al., Urinary 8-hydroxy-2′-deoxyguanosine as a novel biomarker for predicting cardiac events and evaluating the effectiveness of carvedilol treatment in patients with chronic systolic heart failure, Circ. J. 76 (2011) 117–126.
[18] S. Kobayashi, T. Myoren, S. Oda, M. Inari, H. Ishiguchi, W. Murakami, et al., Urinary 8-hydroxy-2′-deoxyguanosine as a novel biomarker of inflammatory activity in patients with cardiac sarcoidosis, Int. J. Cardiol. 190 (2015) 319–328.
[19] R. Blankstein, M. Osborne, M. Naya, A. Waller, C.K. Kim, V.L. Murthy, et al., Cardiac positron emission tomography enhances prognostic assessments of patients with suspected cardiac sarcoidosis, J. Am. Coll. Cardiol. 63 (2014) 329–336.
[20] B.A. Mc Ardle, D.H. Birnie, R. Klein, R.A. de Kemp, E. Leung, J. Renaud, et al., Is there an association between clinical presentation and the location and extent of myocardial involvement of cardiac sarcoidosis as assessed by ^{18}F-fluorodeoxyglucose positron emission tomography? Circ. Cardiovasc. Imaging 6 (2013) 617–626.
[21] K. Soejima, H.D. Yada, The work-up and management of patients with apparent or subclinical cardiac sarcoidosis: with emphasis on the associated heart rhythm abnormalities, J. Cardiovasc. Electrophysiol. 20 (2009) 578–583.
[22] D.H. Birnie, W.H. Sauer, F. Bogun, J.M. Cooper, D.A. Culver, C.S. Duvernoy, et al., HRS expert consensus statement on the diagnosis and management of arrhythmias associated with cardiac sarcoidosis, Heart Rhythm. 11 (2014) 1305–1323.
[23] S. Ishimaru, I. Tsujino, T. Takei, E. Tsukamoto, S. Sakaue, M. Kamigaki, et al., Focal uptake on 18F-fluoro-2-deoxyglucose positron emission tomography images indicates cardiac involvement of sarcoidosis, Eur. Heart J. 26 (2005) 1538–1543.
[24] J.P. Smedema, G. Snoep, M.P. van Kroonenburgh, R.J. van Geuns, W.R. Dassen, A.P. Gorgels, et al., Evaluation of the accuracy of gadolinium-enhanced cardiovascular magnetic resonance in the diagnosis of cardiac sarcoidosis, J. Am. Coll. Cardiol. 45 (2005) 1683–1690.
[25] M.R. Patel, P.J. Cawley, J.F. Heitner, I. Klem, M.A. Parker, W.A. Jaroudi, et al., Detection of myocardial damage in patients with sarcoidosis, Circulation 120 (2009) 1969–1977.
[26] N. Hamzeh, D.A. Steckman, W.H. Sauer, M.A. Judson, Pathophysiology and clinical management of cardiac sarcoidosis, Nat. Rev. Cardiol. 12 (2015) 278–288.
[27] M. Maytin, W.S. Colucci, Cardioprotection: a new paradigm in the management of acute heart failure syndromes, Am. J. Cardiol. 96 (2005) 26G–31G.
[28] M. Yano, S. Okuda, T. Oda, T. Tokuhisa, H. Tateishi, M. Mochizuki, et al., Correction of defective interdomain interaction within ryanodine receptor by antioxidant is a new therapeutic strategy against heart failure, Circulation 112 (2005) 3633–3643.
[29] A.R. Marks, Calcium cycling proteins and heart failure: mechanisms and therapeutics, J. Clin. Invest. 123 (2013) 46–52.
[30] K. Tziomalos, J.M. Hare, Role of xanthine oxidoreductase in cardiac nitroso-redox imbalance, Front. Biosci. (Landmark Ed.) 14 (2009) 237–262.
[31] T.A1. McKinsey, D.A. Kass, Small-molecule therapies for cardiac hypertrophy: moving beneath the cell surface, Nat. Rev. Drug Discov. 6 (2007) 617–635.
[32] Y. Matsui, K. Iwai, T. Tachibana, T. Fruie, N. Shigematsu, T. Izumi, et al., Clinicopathological study of fatal myocardial sarcoidosis, Ann. N. Y. Acad. Sci. 278 (1976) 455–469.
[33] M. Sekiguchi, Y. Yazaki, M. Isobe, M. Hiroe, Cardiac sarcoidosis: diagnostic, prognostic, and therapeutic considerations, Cardiovasc. Drugs Ther. 10 (1996) 495–510.
[34] T.P. Darcy, R. Virmani, Coronary Vasculitis. Nonatherosclerotic Ischemic Heart Disease, Raven Press, New York, 1989 237–275.