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ORIGINAL RESEARCH

Characterization of Immune Checkpoint Inhibitor-Related Cardiotoxicity in Lung Cancer Patients From a Rural Setting

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ABSTRACT

BACKGROUND Immune checkpoint inhibitor (ICI)-related cardiotoxicity (iRC) is uncommon but can be fatal. There have been few reports of iRC from a rural cancer population and few data for iRC and inflammatory biomarkers.

OBJECTIVES The purpose of this study was to characterize major adverse cardiac events (MACE) in ICI-treated lung cancer patients based in a rural setting and to assess the utility of C-reactive protein (CRP) and neutrophil-lymphocyte ratio (NLR) in the diagnosis of iRC.

METHODS Patients with lung cancer treated with ICIs at Vidant Medical Center/East Carolina University (VMC/ECU) between 2015 and 2018 were retrospectively identified. MACE included myocarditis, non-ST-segment elevated myocardial infarction (NSTEMI), supraventricular tachycardia (SVT), and pericardial disorders. Medical history, laboratory values, pre-ICI electrocardiography (ECG), and echocardiography results were compared in patients with and without MACE.

RESULTS Among 196 ICI-treated patients, 23 patients (11%) developed MACE at a median of 46 days from the first ICI infusion (interquartile range [IQR]: 17 to 83 days). Patients who developed MACE experienced myocarditis (n = 9), NSTEMI (n = 3), SVT (n = 7), and pericardial disorders (n = 4). Ejection fraction was not significantly different at the time of MACE compared to that at baseline (p = 0.495). Compared to baseline values, NLR (10.9 ± 8.3 vs. 20.7 ± 4.2, respectively; p = 0.032) and CRP (42.1 ± 10.1 mg/l vs. 109.9 ± 15.6 mg/l, respectively; p = 0.010) were significantly elevated at the time of MACE.

CONCLUSIONS NLR and CRP were significantly elevated at the time of MACE compared to baseline values in ICI-treated patients. Larger datasets are needed to validate these findings and identify predictors of MACE that can be used in the diagnosis and management of ICI-related iRC. (J Am Coll Cardiol CardioOnc 2020;2:491-502) © 2020 The Authors. Published by Elsevier on behalf of the American College of Cardiology Foundation. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

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The authors attest they are in compliance with human studies committees and animal welfare regulations of the authors' institutions and Food and Drug Administration guidelines, including patient consent where appropriate. For more information, visit the *JACC: CardioOncology* author instructions page.

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ABBREVIATIONS AND ACRONYMS

CAR T = chimeric antigen receptor T cell

CTCAE = common terminology for clinical adverse events

CTLA-4 = cytotoxicity T-cell lymphocyte antigen

ICI = immune checkpoint inhibitor

irAE = immune-related adverse events

iRC = immune checkpoint inhibitor-related cardiotoxicity

MACE = major adverse cardiac events

NLR = neutrophil-to-lymphocyte ratio

NSCLC = non-small cell lung cancer

PD = programmed cell death

PD-L1 = programmed cell death-ligand 1

The introduction of immune checkpoint inhibitors (ICI) has significantly improved clinical outcomes in multiple cancer types, including melanoma, lung, kidney, and colorectal cancers (1). ICIs are monoclonal antibodies against programmed cell death-1 (PD-1), programmed cell death-ligand 1 (PD-L1), and cytotoxicity T-cell lymphocyte antigen-4 (CTLA-4) (2). These agents function by blocking an immune checkpoint at the coreceptor and ligand interface of the T-cell and antigen-presenting cell (anti-CTLA-4) or by inhibiting the interaction between the T cell and the tumor cell (anti-PD-1 and anti-PD-L1), enabling the increased destruction of cancer cells (1,2).

A consequence of blocking the physiological immune checkpoint is overactivation of the immune system and regulatory proteins, resulting in an enhanced inflammatory response that can affect multiple organ systems, specifically termed immune-related adverse effects (irAEs) (3). The most common irAEs encountered in clinical trials and practice are colitis, pneumonitis, dermatitis, and thyroiditis (3). Immune checkpoint inhibitor-related cardiotoxicities (iRCs) are less common. Most datasets have observed iRCs in 0.1% to 0.3% of patients. Importantly, however, cases of iRC may be associated with high morbidity and mortality (4). In a recent retrospective analysis from Vigibase (World Health Organization pharmacovigilance database, Basel, Switzerland) that records drug-related adverse events, there were several cases of ICI-related myocarditis and pericarditis, with some overlap with vasculitis (5).

Studies from several groups have estimated the timing and incidence of iRCs (5-8), but potential indicators of iRC development using markers of inflammation such as the neutrophil-to-lymphocyte ratio (NLR) and C-reactive protein (CRP) have not been fully evaluated in patients with iRCs. In other studies, these markers have been used for identifying irAEs (9–11) and assessing cardiovascular disease risk. We hypothesized that patients in rural eastern North Carolina (NC), a community where cardiovascular risk factors and disease are prevalent (12,13), had a high incidence of iRCs. Furthermore, we hypothesized that the inflammatory markers NLR and CRP were associated with the development of iRCs.

METHODS

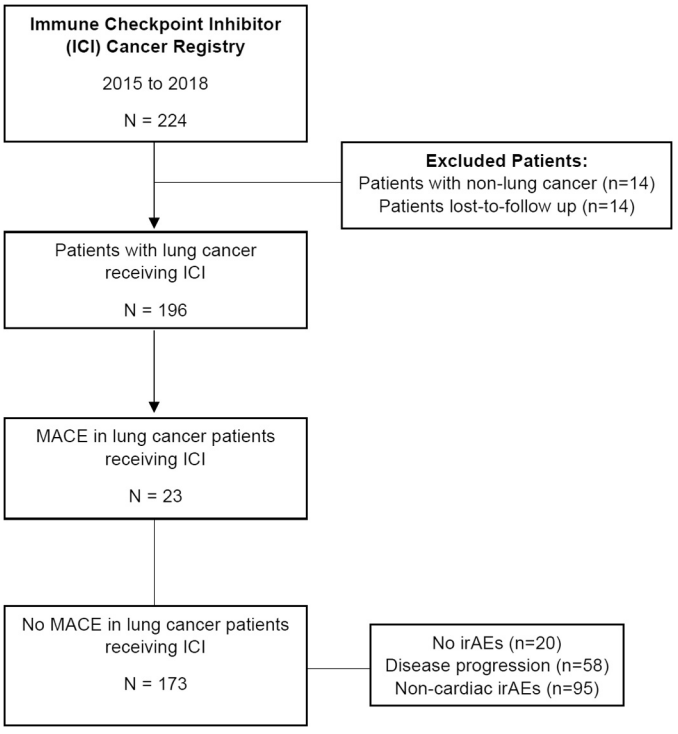
Patients with a diagnosis of lung cancer who received ICI treatment between February 2015 and February 2018 were identified from the tumor registry at Vidant Medical Center/East Carolina University (VMC/ECU). The study was approved through the ECU Institutional Review Board (15-001400). Patients ≥18 years of age who completed at least 1 infusion of ICI therapy at VMC were included in the study (**Figure 1**). Inpatient and outpatient clinic office visits were reviewed. Demographic data including age, sex, body mass index, and ethnicity were collected. Retrospective data collected also included patient lung cancer type and stage; prior radiation therapy; prior and concomitant chemotherapy; ICI therapy and dosage; medical history; cardiac medications; baseline electrocardiograms (ECGs) and echocardiograms; cardiac biomarkers including troponin I (TnI) and brain natriuretic protein (BNP); NLR, calculated from the complete blood count profile; and CRP.

The following major adverse cardiac events (MACE) were observed after ICI: 1) non-ST-segment elevation myocardial infarction (NSTEMI) in the setting of acute coronary syndrome symptoms; 2) new onset supraventricular tachycardias (SVT); 3) myocarditis; or 4) pericardial disorders (acute pericarditis and/or nonmalignant pericardial effusion), each in the absence of sepsis, electrolyte disorders, or other confounding medical conditions. MACE in ICI-treated patients were graded based on the Common Terminology for Clinical Adverse Events version 5.0 (CTCAE v5.0, National Institutes of Health, National Cancer Institute, Bethesda, Maryland) (14). A clinical diagnosis of myocarditis was based on the European Society of Cardiology consensus statement (15) which included 2 or more of the following clinical presentations: 1) new onset (0 days up to 3 months) or subacute or chronic (>3 months of worsening dyspnea at rest or exertion and/or fatigue with left and/or right heart failure signs and/or imaging findings of new right and/or left ventricular dysfunction or elevations of BNP or TnI; 2) sudden cardiac death or aborted cardiac death; 3) new atrioventricular block or bundle branch block, sinus arrest, ventricular tachycardia, or fibrillation and asystole; or 4) tissue characterization by cardiac magnetic resonance (CMR) imaging showing edema or late gadolinium enhancement. Post hoc review of patients based on definitions suggested by Bonaca et al. (16) as definite, probable, or possible myocarditis were further used to verify adjudication of suspected myocarditis cases. ICI-treated patients without MACE were further categorized as patients who did not experience any irAEs, or experienced noncardiac irAEs, or who had disease progression (did not complete 4 cycles of ICI due to tumor progression).

At the time of MACE, patients' symptoms were obtained based on initial history of presenting illness documentation. Time to onset of MACE was defined as the number of days from the first ICI infusion to presentation of MACE. Presence of new pericardial effusion, new wall motion abnormalities, diastolic dysfunction, and ejection fraction (EF) at the time of MACE were compared to prior echocardiograms. Patients with evidence of malignant pericardial effusion from pathology were not considered part of the outcomes analysis. Cardiac biomarkers, obtained according to the treating clinicians' discretion, including TnI, BNP, and inflammatory markers including NLR and CRP at the time of an irAE or MACE, were compared to respective baseline values. In addition, baseline NLR and CRP in patients with MACE were compared to baseline values of patients without MACE. TnI and BNP values of up to 6 months prior to ICI treatment were considered baseline values. In patients who did not experience any irAEs, available NLR and CRP data were also obtained at ICI cycle 6 (C6) or ICI cycle 8 (C8). Institutional normal values are as follows: TnI, <0.03 ng/ml; BNP, <200 pg/ml; and CRP, <20 mg/l. Although there is no standardized NLR range that defines normal or abnormal, a recent study suggested that, in an adult, healthy, nongeriatric population, a normal NLR is between 0.78 and 3.53 (17).

STATISTICS. Numerical data are presented as mean ± SD or median (interquartile range [IQR]). Categorical variables are presented as total number and percentage. A Fine and Gray competing risk univariable analysis was used to evaluate the associations among baseline demographics, cancer history, other irAEs, baseline laboratory values, comorbidities, and medications with MACE. To assess the cardiac presentation and diagnostic workup in patients with MACE, baseline echocardiographic and ECG parameters were compared to respective values at the time of MACE. Continuous variables (EF and PR and QTc intervals) were compared using a paired Student's *t*-test, whereas categorical variables (pericardial effusion, right ventricular systolic pressure >35 mm Hg, wall motion abnormalities, diastolic dysfunction, and ECG rhythms) were compared using a McNemar test. Furthermore, baseline NLR and CRP were compared to respective values at the time of MACE, irAE, or cycle 6 by using a paired Student's *t*-test. A Fine and Gray competing risk analysis was used to determine the relationship between baseline NLR or CRP and risk of MACE. A p value <0.05 was considered statistically significant. Statistical analyses were performed using SAS version 9.4 (SAS Institute, Cary, North Carolina).

FIGURE 1 Study Design for Retrospective Evaluation of MACE in Lung Cancer Patients Receiving ICI



Retrospective study of MACE in lung cancer patients receiving ICIs. Patients receiving ICIs for lung cancer were retrospectively identified (n = 196) for MACE. Patients without MACE included those without any irAEs (n = 20), other noncardiac irAEs (n = 58), or disease progression (n = 95). ICI = immune checkpoint inhibitors; irAEs = immune-related adverse events (irAEs); MACE = major adverse cardiac events.

RESULTS

DEMOGRAPHICS AND BASELINE CHARACTERISTICS OF ICI-TREATED PATIENTS WITH AND WITHOUT MACE. There were 196 patients who received ICIs for lung cancer at the time of this retrospective review. Patients were predominantly white and male and received ICI for non-small cell lung cancer (NSCLC) (Table 1). Patients with MACE tended to be older than patients without MACE (68.7 ± 1.8 vs. 64.3 ± 0.8 years of age, respectively; p = 0.064). Nivolumab was more frequently administered than other agents in both patients without MACE (68%) and those with MACE (83%), at a dose of 3 or 240 mg/kg. Only 25% of patients without MACE and 17% of patients with MACE received pembrolizumab. There were 11 patients without MACE who received atezolizumab (1,200 mg) and 1 patient who received a combination of nivolumab (3 mg/kg) plus ipilimumab (3 mg/kg). Patients

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TABLE 1 Univariable Associations Among Baseline Demographics, Cancer History, and Other irAEs and MACE in ICI-Treated Lung Cancer Patients Based on Fine and Gray Competing Risk Analysis

	No MACE (n = 173, 88%)	MACE (n = 23, 11%)	Estimated HR (95% CI)	p Value
Age, yrs	64.3 ± 0.8	68.7 ± 1.8	1.04 (0.99-1.08)	0.064
BMI, kg/m^2	26.4 ± 7.2	25.5 ± 4.5	0.96 (0.90-1.03)	0.264
Females	74 (43)	8 (35)	1.38 (0.59-3.24)	0.456
Males	99 (57)	15 (65)	–	–
Ethnicity				
Blacks	66 (38)	5 (21)	2.21 (0.84-5.86)	0.108
Whites	107 (62)	18 (78)	–	–
Type of lung cancer				
NSCLC	158 (91)	21 (87)	1.49 (0.45-4.91)	0.509
SCLC	15 (9)	3 (13)	–	–
Stage of lung cancer				
III	56 (32)	12 (52)	1.21 (0.28-5.18)	0.798
IV	117 (68)	11 (48)	–	–
ICI				
Anti-PD-1				
Nivolumab	118 (68)	19 (83)	1.69 (0.59-4.89)	0.329
3 mg/kg	79 (67)	10 (53)	–	–
240 mg	39 (34)	9 (47)	–	–
Pembrolizumab (200 mg)	43 (25)	4 (17)	–	–
Anti-PD-L1				
Atezolizumab (1,200 mg)	11 (6)	0 (0)	–	–
Anti-PD-1 + Anti-CTLA-4				
Nivolumab, (3 mg/kg) + ipilimumab, (3 mg/kg)	1	0 (0)	–	–
Other cancer therapy				
Anti-topoisomerase	66 (38)	9 (39)	0.76 (0.32-1.79)	0.526
Anti-VEGF	13 (7)	0 (0)	–	–
Alkylating agents	139 (80)	21 (91)	1.10 (0.51-2.37)	0.804
Antimetabolites	68 (40)	11 (48)	0.87 (0.39-1.95)	0.743
Taxanes	11 (6)	6 (26)	4.78 (1.88-12.1)	0.001
History of thoracic radiation	75 (61)	10 (43)	0.79 (0.33-1.89)	0.593
Concomitant irAEs				
None	21 (12)	14 (56)	0.31 (0.07-1.34)	0.117
Adrenal insufficiency	3 (2)	0 (0)	–	–
Cerebritis	15 (9)	0 (0)	–	–
Colitis	6 (3)	2 (8)	3.94 (1.05-14.75)	0.042
Dermatitis	5 (1)	0 (0)	–	–
Encephalitis	6 (5)	0 (0)	–	–
Hepatitis	4 (2)	0 (0)	–	–
Hypophysitis	2 (1)	0 (0)	–	–
Myositis	2 (0.5)	0 (0)	–	–
Pancreatitis	3 (2)	0 (0)	–	–
Pneumonitis	54 (31)	2 (8)	0.88 (0.05-16.9)	0.931
Thyroiditis	3 (3)	0 (0)	–	–

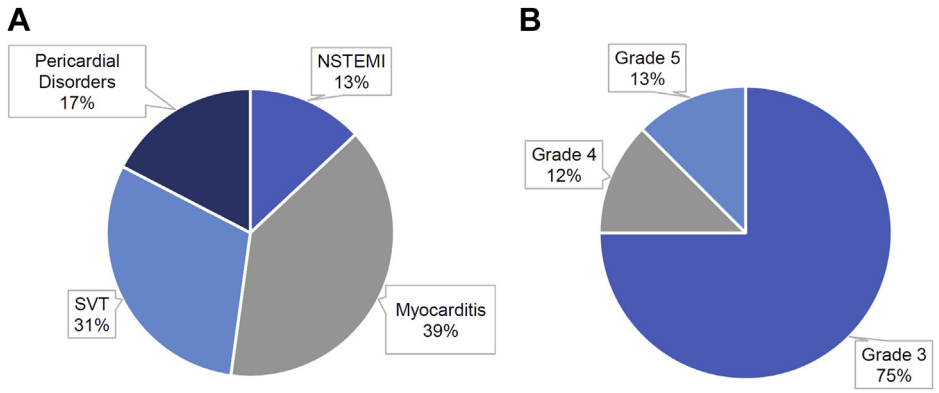
Values are mean ± SD or n (%).

BMI = body mass index; CI = confidence interval; CTLA-4 = cytotoxic T-lymphocyte-associated protein-4; HR = hazard ratio; ICI = immune checkpoint inhibitor; irAEs = immune-related adverse events; MACE = major adverse cardiac events; ICIs = immune checkpoint inhibitors; NSCLC = non-small cell lung cancer; PD-1 = programmed cell death-1; PD-L1 = programmed death ligand-1; SCLC = small cell lung cancer; VEGF = vascular endothelial growth factor.

also received various nonimmunotherapy cancer therapy regimens, most of which were alkylating agents.

Among 196 patients, there were 23 patients (11%) with MACE that included possible myocarditis (n = 9 of 23; 39%), NSTEMI (n = 3 of 23; 13%), SVT (n = 7 of 23; 31%) and pericardial disorders (n = 4 of 23; 17%) (**Figure 2A**). Cardiotoxicity was graded based on CTCAE v5.0 criteria (14) (Supplemental Table 1). Fulfilled criteria for myocarditis was based on European

FIGURE 2 Incidence of MACE and Percentage of MACE Toxicity in Lung Cancer Patients Receiving ICI



(A) Incidence of MACE in lung cancer patients receiving ICI. **(B)** Percentage of MACE toxicity severity based on CTCAE grading. **(A)** Among 196 patients, 23 (11%) patients receiving ICI developed MACE, including NSTEMI (n = 3); myocarditis (n = 9); SVT (n = 7); and pericardial disorders (n = 4). **(B)** A majority of MACE were grade 3 toxicity (75%), with 3 patients (13%) experiencing grade 5 toxicity (death). CTCAE = common terminology for clinical adverse event; NSTEMI: non-ST-segment elevation myocardial infarction; SVT = supraventricular tachycardia.

Society of Cardiology consensus statement (15) and definitions suggested by Bonaca et al. (16) in 9 patients are detailed in Supplemental Table 2. A total of 75% had grade 3 toxicity, defined as a severe or medically significant but not immediately life-threatening adverse event that is disabling, limiting self-care of activities of daily living, and requiring hospitalization or prolonged hospitalization. Three patients (14%) experienced grade 5 toxicity (death) related to the adverse event (**Figure 2B**) caused by NSTEMI, possible myocarditis, and pericardial tamponade. More than 50% of the patients with MACE did not experience concomitant irAEs; however, 4 patients developed colitis (n = 2) and pneumonitis (n = 2). There were 54 patients (31%) without MACE who developed pneumonitis, 15 patients (9%) with cerebritis or encephalitis, 6 patients (3%) with colitis, 5 patients (3%) with dermatitis and other irAEs (**Table 1**).

In patients with and without MACE, diabetes mellitus, hypertension, hyperlipidemia, atrial fibrillation, chronic kidney disease, chronic obstructive pulmonary disease, and cerebrovascular disease were similarly present. There was a higher proportion of patients with MACE who had a history of coronary artery disease compared than patients without MACE (estimated hazard ratio [HR]: 2.79; 95% confidence interval [CI]: 1.18 to 6.59; p = 0.019). There was also a higher proportion of patients with MACE who were taking steroids than patients without MACE (n = 18 of 23; 78%; vs. n = 61 of 173; 35%, respectively; p < 0.001) (**Table 2**). Among the patients without MACE who were receiving steroids, 79% (n = 48 of 61) were receiving dexamethasone as palliative treatment for metastatic disease (Supplemental Table 3).

CLINICAL PRESENTATION AND CARDIAC DIAGNOSTIC WORKUP OF ICI-TREATED LUNG CANCER PATIENTS WITH MACE. More than 50% of patients without MACE presented with dyspnea and palpitations, whereas <25% of patients presented with chest pain (**Figure 3C**). The median time to onset of development of iRC from the first day of ICI infusion was 46 days (IQR: 17 to 83 days) (**Figure 3A**). The median number of doses until the onset of MACE was 3 (IQR: 2 to 4 doses) (**Figure 3B**).

There were only 4 patients with MACE who had a measured baseline TnI prior to ICI treatment with a mean of 0.03 ± 0.01 ng/ml. In patients without MACE with baseline measured TnI (n = 42 of 173), mean TnI was 0.02 ± 0.01 ng/ml. At the time of MACE, TnI was available in only 12 patients and was elevated to a mean of 0.98 ± 0.36 ng/ml with a peak value across 12 patients of 1.35 ± 0.49 ng/ml. There were no baseline BNP data available in patients with MACE. BNP value was available in only 18 patients at the time of MACE and was 384 ± 339 pg/ml (**Table 2**). Baseline BNP in 11 of the 173 patients without MACE was 158 ± 148 pg/ml. EFs were not significantly different at the time of MACE (n = 22 of 23) in comparison to baseline EFs (n = 17 of 23; 46.2 ± 16.8% vs. 50.5 ± 16.2%, respectively; p = 0.495) (**Table 3**). Patients were predominantly in normal sinus rhythm (63%). PR interval was

TABLE 2 Univariable Associations Among Baseline Laboratory Values, Comorbidities, and Medications and MACE in ICI-Treated Lung Cancer Patients Based on Fine and Gray Competing Risk Analysis

	No MACE	MACE	Estimated HR (95% CI)	p Value
Baseline laboratory values				
WBC, K/μl	8.86 ± 5.1	8.92 ± 5.4	1.07 (1.02-1.11)	0.003
Hemoglobin, g/dl	11.9 ± 7.4	10.6 ± 1.6	1.01 (1.01-1.02)	<0.001
Hematocrit, %	35.9 ± 15	32.5 ± 5.1	0.91 (0.89-0.93)	<0.001
Platelets, K/μl	282 ± 117	239 ± 117	0.91 (0.98-0.99)	0.005
Creatinine, mg/dl	1.06 ± 0.9	0.91 ± 0.3	1.29 (1.15-1.45)	<0.001
eGFR, ml/min/1.73 m^2	73.5 ± 18.1	67.9 ± 15.3	0.96 (0.94-0.98)	<0.001
NLR	8.1 ± 9.0	10.9 ± 8.3	1.05 (1.01-1.09)	0.022
CRP, mg/l	37.5 ± 48.1	42.1 ± 46.0	1.01 (0.99-1.01)	0.546
Troponin, ng/ml*	0.02 ± 0.01	0.03 ± 0.01	–*	–*
BNP, pg/ml	158 ± 148	–*	–*	–*
Comorbidities				
CAD	27 (16)	8 (35)	2.79 (1.18-6.59)	0.019
CKD	9 (5)	1 (4)	0.82 (0.11-5.94)	0.845
Atrial fibrillation/flutter	14 (9)	4 (13)	1.54 (0.45-5.15)	0.485
COPD	61 (35)	11 (48)	1.61 (0.71-3.63)	0.252
CVA	20 (12)	3 (13)	1.14 (0.34-3.82)	0.838
DM	44 (25)	7 (30)	1.26 (0.52-3.03)	0.609
HLD	56 (32)	4 (17)	0.45 (0.16-1.29)	0.135
HTN	111 (64)	12 (52)	0.63 (0.29-1.42)	0.267
Medications				
RAAS inhibitors	5 (22)	41 (24)	0.91 (0.34-2.42)	0.846
BB	51 (30)	16 (65)	3.87 (1.65-9.09)	0.001
CCB	34 (20)	4 (17)	0.87 (0.29-2.56)	0.804
Thiazide diuretic	20 (11)	3 (13)	1.13 (0.34-3.79)	0.846
Loop diuretics	21 (12)	8 (35)	3.81 (1.69-8.59)	0.001
Statins	58 (33)	12 (52)	2.01 (0.89-4.55)	0.093
Steroids	61 (35)	18 (78)	4.10 (1.89-8.88)	<0.001

Values are mean ± SD or n (%). *Analysis was not performed secondary to limited sample size.

BB = beta-blocker; BNP = brain natriuretic peptide; CAD = coronary artery disease; CCB = calcium channel blocker; CKD = chronic kidney disease; COPD = chronic obstructive pulmonary disease; CRP = C-reactive protein; CVA = cerebrovascular disease; DM = diabetes mellitus; eGFR = estimated glomerular filtration rate; HLD = hyperlipidemia; HTN = hypertension; NLR = neutrophil-lymphocyte ratio; RAAS = renin-angiotensin-aldosterone system; WBC = white blood cell count; other abbreviations as in Table 1.

shorter (−19.6 ± 8.2 ms from baseline; p = 0.031) and QTc was more prolonged (26.8 ± 12.0 from baseline; p = 0.036) at the time of MACE (n = 17 of 23) in comparison to values at baseline (n = 12 of 173), though QTc was <500 ms.

INFLAMMATORY MARKERS OF ICI-TREATED LUNG CANCER PATIENTS WITH AND WITHOUT MACE. Baseline NLR and CRP values were available in all 196 patients. Baseline NLR in patients with MACE were significantly higher than in patients without MACE (10.9 ± 8.3 vs. 8.1 ± 9.0, respectively; p = 0.022) (Table 2) and compared to patients without any irAEs (10.9 ± 8.3 vs. 7.4 ± 3.4, respectively; p = 0.029) (Central Illustration). NLR in patients who did not experience any irAEs (n = 20 of 173) at baseline and between C6 and C8 remained ≤10 at all-time points (p = 0.380). There was a significant increase in NLR observed at the time of MACE from baseline (20.7 ± 4.2 vs. 10.9 ± 8.3, respectively; p = 0.032) and in patients with other noncardiac irAEs compared to baseline values (n = 95 of 173; 7.45 ± 1.1 vs. 17.3 ± 2.4, respectively; p = 0.032) (Central Illustration). There were no statistically significant differences between baseline NLR in patients with disease progression (n = 58 of 173) and NLR values in patients with no irAEs.

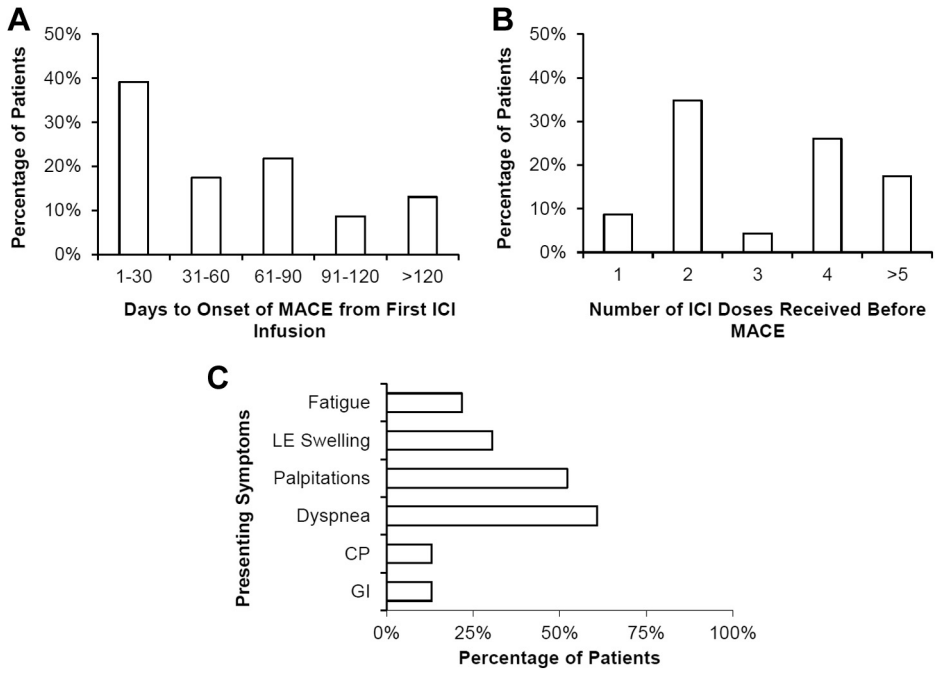
CRP at the time of MACE and in patients with noncardiac irAEs was significantly elevated compared with respective baseline values (109.9 ± 15.6 vs. 42.1 ± 10.1, respectively; p = 0.010; and 107.1 ± 9.8 mg/l vs. 37.7 ± 5.4 mg/l, respectively; p = 0.031) (Central Illustration). However, baseline CRP did not differ according to MACE (HR: 1.01; 95% CI: 0.99 to 1.01; p = 0.546). Baseline CRP in patients with disease progression was similar to that in patients with MACE and with other noncardiac irAEs. CRP in patients with no irAEs was <20 mg/l at baseline and between C6 and C8 also was not significantly different (p = 0.168).

DISCUSSION

In this retrospective study of lung cancer patients receiving ICI, there was an 11% incidence of MACE consisting of various cardiotoxicities that included possible myocarditis, NSTEMI, new onset SVT, and pericardial disorders. Importantly, a significant increase was observed in NLR and CRP at the time of MACE in comparison to patients who did not experience any irAEs (Central Illustration), highlighting its potential utility in the screening and diagnosis of MACE in ICI-treated patients.

Possible myocarditis (4.5%) and SVT (3.6%) were the predominant adverse cardiac events observed in this study. The high incidence observed for MACE may have been the result of the definition of MACE, which captured a spectrum of cardiotoxicities not limited to myocarditis, inclusion of only lung cancer patients who could have higher risks to development of MACE such as chest radiation therapy (18) and the potential role of host-environmental factors unique to the rural population of eastern NC (12,13). Indeed, higher inflammatory signals and incidence of pneumonitis were also observed in this lung cancer population in comparison to those reported previously (13,19). Furthermore, in the lung cancer patients who received radiation, it has been suggested that the synergistic effect of radiotherapy and immunotherapy for priming of an endogenous antigen-specific immune response may contribute to a higher incidence of MACE by T-cell recognition of shared antigens (20).

FIGURE 3 Clinical Presentation of MACE in Lung Cancer Patients Receiving ICI



(A) Time to MACE onset from first ICI infusion. Median days to onset of MACE from the first ICI infusion was 46 days (IQR: 17 to 83 days). **(B)** Number of ICI doses received before MACE. Approximately 35% of patients developed MACE after 2 ICI infusion cycles with a median dose of 3 doses (IQR: 2 to 4 doses) at the time of iRC. **(C)** Presenting symptoms at the time of MACE. More than 50% of patients presented with palpitations and dyspnea. Less than 25% of patients presented with CP. CP = chest pain; GI = gastrointestinal (symptoms: nausea, vomiting, abdominal pain); IQR = interquartile range; iRC = immune-related cardiotoxicity; other abbreviations as in Figure 1.

Timing of the development of MACE with a median delay of 46 days and a median of 3 doses from the first ICI administration in this study was similar to that previously reported (5,21,22). In view of this timing of onset from earlier studies, there is a general recommendation that baseline and surveillance cardiac testing (echocardiogram, ECG, TnI and BNP) be considered during this potential MACE window period, noted as after the second and after the fourth ICI cycle administration (21). Most of the present MACE cases were not associated with a decrease in EF from baseline, which is consistent with previous studies (18,21), suggesting that relying solely on EF in ICI-treated patients with MACE may be limited for the detection of iRCs. TnI was observed to be mildly elevated at the time of MACE. However, elevations in TnI have also been observed in cancer patients receiving cancer therapy, including ICIs without any cardiotoxicities (23,24), thus suggesting that its utility may also be limited for the detection of iRC.

As demonstrated by this study, symptoms of iRC may be variable, including nonspecific symptoms of shortness of breath and palpitations that may overlap with other common cancer-related complications, which could result in underdiagnosis of iRC if unsuspected. Inflammatory markers such as CRP and NLR as diagnostic tools for identifying and monitoring irAEs have been previously documented (9-11); however, they have not been specifically investigated in iRCs. In this study, there was an elevation of CRP and NLR in patients at the time of MACE in comparison to baseline values, and this may possibly reflect similar inflammatory downstream effects such as those seen with cytokine release storm. In the setting of ICI-related pneumonitis, an upward trend of CRP has been previously documented (19) similar to that seen with cytokine release storm in chimeric antigen receptor T (CAR T) cells and tumor-infiltrating (TIL) therapy (25). In patients who did not experience any irAEs throughout the ICI treatment course, CRP remained <20 mg/l. In comparison, a significant >2-fold elevation in CRP, compared to baseline, was observed during the time of MACE and irAE. These findings are similar to a previous study conducted by

TABLE 3 Cardiac Imaging and Biomarker Values at Time of MACE Compared to Baseline (N = 23)

	Baseline	MACE	95% CI	p Value
Echocardiogram	n = 17	n = 22		
Ejection fraction, %	50.5 ± 16.2	46.2 ± 16.8 Δ4.22 ± 6.00	−17.3 to 8.89	0.495
+Pericardial effusion	1 (6)	8 (36)	–	0.016
+RVSP >35 mm Hg	0 (0)	5 (22)	–	–
+WMA	2 (12)	2 (9)	–	1.000
+Diastolic dysfunction	1 (6)	4 (19)	–	1.000
Cardiac biomarkers	n = 4	n = 12		
Troponin I ng/ml	0.03 ± 0.01	0.98 ± 0.36	–	–
BNP, pg/ml	–	384 ± 339	–	–
Electrocardiogram	n = 12	n = 17		
PR interval, ms	171.1 ± 29.9	155.9 ± 30.6 Δ−19.6 ± 8.22	−37.3 to −2.08	0.031
QTc interval, ms	442.4 ± 37.9	466.1 ± 34.8 Δ26.8 ± 12.0	1.9 to 51.7	0.036
Rhythm				
Normal sinus rhythm	10 (63)	7 (29)	–	0.388
Sinus bradycardia	0 (0)	1 (5)	–	–
Sinus tachycardia	1 (5)	6 (29)	–	0.727
Atrial fibrillation/flutter	3 (19)	7 (33)	–	0.125
Bundle branch block	2 (13)	2 (9)	–	1.000
ST-segment depression/elevation	0 (0)	4 (19)	–	–
Nonspecific T wave abnormality	5 (31)	3 (14)	–	1.000

Values are mean ± SD or n (%). PR and QTc intervals at the time of MACE (n = 17) were compared to baseline (n = 12). Ejection fraction and the presence of pericardial effusion at time of MACE (n = 23) were compared to available baseline echocardiograms (n = 17).

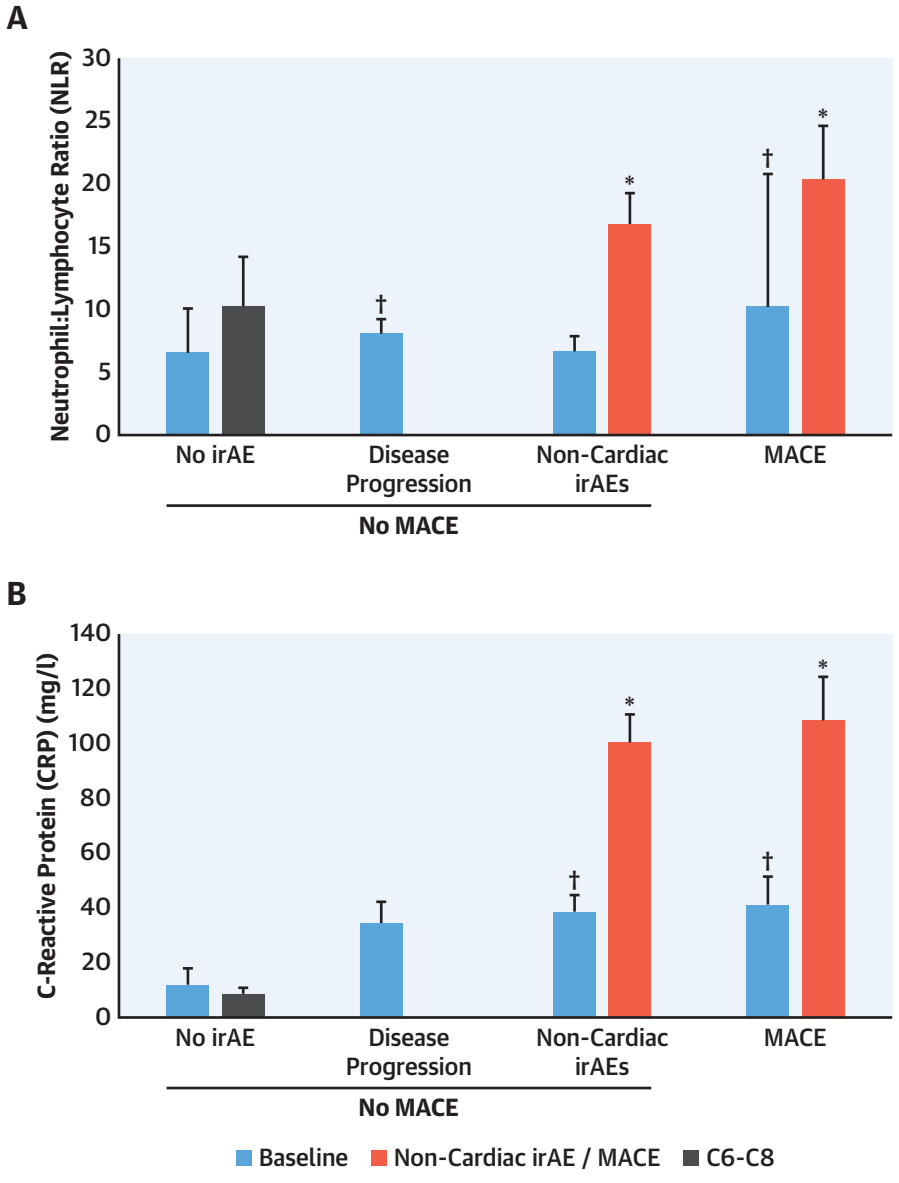
Δ = change from baseline; BNP = brain natriuretic peptide; OR = odds ratio; RVSP = right ventricular systolic pressure; WMA = wall motion abnormalities; other abbreviations as in Tables 1 and 2.

our institution that observed a significant increase in CRP with nivolumab-related pneumonitis that was mitigated with use of tocilizumab (19). Similar inflammatory-mediated mechanisms such as interleukin (IL)-6 may be observed in CAR T cell-related cardiotoxicities (26) further highlighting and extending the utility of CRP in the detection of iRCs.

Another inflammatory marker that has been previously studied in noncardiac irAEs and observed to be elevated at the time of irAE is NLR (8,11). NLR is easily obtained, inexpensive, and a routine test, and is calculated from the total white blood cell count that reflects the ratio of the innate (neutrophils) and the adaptive (lymphocyte) immune pathways, whereby an elevation results in an imbalanced toxic inflammatory response with release of cytokines (27). We observed an increase in NLR in patients who experienced noncardiac irAEs, but, importantly, a significant elevation of NLR was found in patients at the time of MACE, which has not been previously reported in ICI-related cardiotoxic events.

In coronary disease, an elevation in NLR has been observed in animal models where neutrophils are recruited early after myocardial injury along with proinflammatory monocytes and lymphocytes which activate the inflammatory cascade through the neutrophil-activated CD11B/CD18, reviewed in detail elsewhere (28). The protective roles of PD-L1, PD-1, and CTLA-4 in the intricacy of the immunity-inflammatory environment against development of atherosclerotic plaque and myocardial infarction have also been previously described in animal models. Hypercholesteremic PD-L1 and PD-1 knockout mice have an exaggerated T-cell-mediated immune response and cytokine secretion that results in a larger atherosclerotic lesion size composed of numerous $CD8^+$ T cells and apoptotic core (29,30). Similarly, CTLA-4 inhibition results in accelerated atherosclerosis that can be mitigated by CTLA-4-Ig (31). Thus, in the setting of ICI use, this may result in a proatherosclerotic inflammatory signal leading to an increased risk for the development of iRC in patients with underlying coronary heart disease. In cases of ICI-related myocarditis, an increase in inflammatory cells, particularly abundant T cells and macrophages were observed in the postmortem assessment of the myocardium (4). Although the mechanisms of ICI-related myocarditis remain to be fully elucidated, mistaken recognition of cardiac self-antigen for foreign antigen is one plausible pathway.

CENTRAL ILLUSTRATION **NLR and CRP at Baseline Versus at the Time of irAE, MACE, or C6-C8 During ICI Treatment**



Moey, M.Y.Y. et al. J Am Coll Cardiol CardioOnc. 2020;2(3):491–502.

(A) NLR in ICI-treated patients with and without MACE. In patients who did not experience any irAEs, NLR at baseline and C6 to C8 were <10. NLR at baseline was >10 in patients with MACE and patients who had disease progression. NLR increased significantly at the time of MACE and noncardiac irAEs **(red bars)**. **(B)** CRP in ICI-treated patients with and without MACEs. In patients who did not experience any irAEs, CRP was <20 mg/l at baseline and C6 to C8. CRP was elevated in patients with MACE and in patients with disease progression and who had noncardiac irAEs. At the time of irAE or MACE, CRP was significantly elevated in comparison to baseline CRP. *$p < 0.05$ in comparison to respective baseline values; †$p < 0.05$ in comparison to baseline values in patients with no irAEs. C6-C8 = cycle 6 to cycle 8; CRP = C-reactive protein; ICI = immune checkpoint inhibitor; irAE = immune-related adverse events; MACE = major adverse cardiac events; NLR = neutrophil-lymphocyte ratio.

Interestingly, a significant proportion of patients with MACE were receiving concomitant corticosteroids predominantly for palliative treatment. The effect of baseline steroids concurrent with ICI treatment on outcomes is complex, whereby chronic prednisone usage of >10 mg per day in NSCLC has been shown to be associated with decreased clinical benefit and shorter overall survival (32,33) in comparison to transient use (33); however, other retrospective studies have debated this (34,35). ICI therapy efficacy may not be adversely affected by steroids due to the dose-dependent, cell cycle-dependent, and time-dependent effects of steroids on T-cell survival (36). Therefore, it is possible that, after ICI-mediated T cell activation or upregulation, T cells may be protected from chronic corticosteroid-induced T-cell cytolysis, resulting in increased risk for irAEs or iRCs.

NLR in cancer patients receiving ICI may also predict the response and development of irAEs. In solid cancers and melanoma, an elevated NLR prior to and during ICI treatment has demonstrated poor response to chemoradiation therapy (37,38) as well as worse outcomes in overall and progression free survival (39,40). This may be explained on the basis of ICI therapy, which recruits lymphocytes for tumor destruction such that an elevated NLR may represent an impaired immune tumor response. Although there is no standardized NLR range that defines normal, in an adult, healthy, nongeriatric population, a normal NLR is between 0.78 and 3.53 (17). NLR at baseline in patients with MACE and patients who experienced noncardiac irAEs was also significantly elevated in comparison to baseline NLR values of patients who did not develop any irAEs. Baseline NLR in patients who had significant tumor disease progression was almost equal to the baseline NLR of patients with MACE, which may be explained by activation of a proinflammatory status with rapid release of poorly differentiated neutrophils that has been seen in patients with significant tumor progression (40). Although the current findings may seem to conflict with some prior studies which observed an association with low baseline NLR and development of irAEs (38,39), increased NLR and a higher proportion of steroid use suggests an inflammatory state quite consistent with the cardiotoxic events experienced by the present patients who were receiving ICIs.

STUDY LIMITATIONS. There are several limitations to this study stemming from the bias inherent in any single-institution retrospective analysis. The present study population was also small with the potential for uncontrolled confounding and missing data. The definitions of MACE encompassed various cardiotoxicities that were from International Classification of Diseased codes and electronic health record documentation. At the time of data acquisition, iRCs were not always well recognized, and CMR imaging was not routinely performed at our institution, thus resulting in myocarditis diagnosis based on clinical and diagnostic criteria without myocardial biopsy and cardiac CMR. Baseline values of cardiac biomarkers were missing in a number of patients, as it was not considered standard of practice to obtain in the outpatient setting or in asymptomatic patients. As such, trends and utility of these cardiac biomarkers could not be fully evaluated. As such, these results are hypothesis-generating. Earlier studies have demonstrated that combination ICI is associated with higher mortality (5) in comparison to either anti-PD-1 or anti-PD-L1 monotherapy. The different effects of monotherapy versus ICI combination therapy on MACE and changes in inflammatory markers could not be assessed because most patients received nivolumab at our institution.

CONCLUSIONS

The introduction of ICIs has significantly changed treatment strategies for many cancer patients. Although ICI therapy is highly effective for some cancer patients, these agents may adversely affect other organs, including the heart. iRCs are uncommon but, as shown, myocarditis and SVT were the major MACE noted in our patient population. The most striking finding from this study was the rise in the inflammatory biomarkers CRP and NLR at the time of MACE. Studies are needed to understand whether CRP and NLR can be used as data to inform multidisciplinary decisions among the patient, oncologist, and cardiologist regarding diagnosis and optimal therapeutic management of ICI therapy. Further prospective studies are needed to identify risk factors of iRC, chronic effects of ICIs, and the utility of NLR and CRP in the prognostication of MACE.

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PERSPECTIVES

COMPETENCY IN MEDICAL KNOWLEDGE: Incidence of MACE in ICI-treated patients was 11%, and events consisted of myocarditis, NSTEMI, supraventricular tachycardia, and pericardial disorders. EF was preserved in patients who experienced MACE, suggesting its role as a sole marker for cardiotoxicity may be limited. With further study, NLR and CRP may be useful surveillance inflammatory biomarkers for the detection of MACE following ICI treatment.

TRANSLATIONAL OUTLOOK: Prospective studies assessing cardiac and inflammatory surveillance markers, including NLR and CRP, during the course of ICI treatment are needed to inform algorithms for the early detection and management of MACE in patients receiving ICI.

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KEY WORDS **immune checkpoint inhibitors, inflammatory markers, myocarditis, neutrophil-to-lymphocyte ratio**

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