State of the Art: Precision Cardio-Oncology

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Running Title: Precision Cardio-Oncology
NOTEWORTHY

The recent gains in cancer survivorship have produced a growing cohort of patients with increased cardiovascular complications and led to the field of cardio-oncology. (pg.4)

Within cardio-oncology, current risk stratification schemes primarily include only basic clinical characteristics. (pg.6)

Refined imaging measures and sensitive biomarkers of disease represent tools with the potential to tailor treatments to underlying pathophysiologic differences in patients with otherwise similar clinical presentations. (pg.6)
ABSTRACT

Modern oncologic therapies and care have resulted in a growing population of cancer survivors with comorbid, chronic health conditions. As an example, many survivors suffer from an increased risk of cardiovascular complications secondary to cardiotoxic systemic and radiation therapies. In response, the field of cardio-oncology has emerged as an integral component of oncologic patient care, committed to the early diagnosis and treatment of adverse cardiac events. However, as current clinical management of cancer therapy-related cardiovascular disease (CVD) remains limited by a lack of phenotypic data, implementation of precision medicine approaches has become a focal point for deep phenotyping strategies. In particular, -omics approaches have shown enormous potential in identifying sensitive biomarkers of CVD, applying sophisticated, pattern-revealing technologies to growing databases of biological molecules. Moreover, the use of -omics to inform radiologic strategies may add an additional dimension to future clinical practices. In this review we present a paradigm for a precision medicine approach to the care of cardiotoxin-exposed cancer patients. We discuss the role of current imaging techniques; demonstrate how -omics can advance our understanding of disease phenotypes; and describe how molecular imaging can be integrated to personalize surveillance and therapeutics, ultimately reducing cardiovascular morbidity and mortality in cancer patients and survivors.

Key Words: cardio-oncology; precision medicine; precision cardio-oncology; -omics; cardiotoxicity
INTRODUCTION

The last decade has witnessed a revolution in cancer therapeutics, producing a population of cancer survivors now exceeding 15.5 million in the United States alone (1). While encouraging, this gain in cancer survivorship has also produced a growing cohort of patients with increased cardiovascular complications. Cardiovascular disease (CVD) currently accounts for approximately 32% of deaths globally, with a significant percentage in those with previous cancer treatment (2). To address this growing public health burden, a new discipline of cardio-oncology dedicated to the cardiovascular care of cancer patients has emerged. However, despite substantial progress made in elucidating mechanisms of therapy-related cardiotoxicity, personalized diagnostic and therapeutic strategies remain limited, with few distinctions made between underlying mechanisms of CVD.

One strategy that has gained momentum in cardiology and oncology is the advancement of precision medicine, which relies on computational integration of clinical data with advanced genetic, biomolecular, and demographic profiling to better risk stratify patients. This process, known as deep phenotyping, directly undermines the reductionist assumption that similar phenotypes arise from similar underlying pathophysiology. In the case of cancer therapy-related CVD, this reductionist assumption remains the current therapeutic standard, largely due to a lack of phenotyping data in risk stratification and disease development. As the link between cancer therapeutics and CVD continues to heterogeneously manifest in the clinic, our understanding of the potential applications of precision medicine in cardio-oncology is central to actualizing successful preventative, diagnostic, and therapeutic interventions.

In this review we present a paradigm for how to advance our understanding of the disease phenotype in cancer patients exposed to potential cardiotoxins. We start with a background on the state of cardio-oncology and follow with a discussion of -omics approaches to enhance disease characterization and elucidate novel biomarkers. We then illustrate how a variety of current and emerging imaging
techniques are differentially suited for phenomapping disease. We conclude by using radiotherapy-induced microvascular injury as a case study for how -omics identification of molecular imaging targets could shape the future of personalized cardio-oncology.

BACKGROUND

Clinical manifestations of cancer therapy cardiotoxicity are heterogeneous. The most recent European Society of Cardiology guidelines broadly divide cardiovascular complications of cancer therapy into nine categories, including myocardial dysfunction, coronary artery disease, valvular heart disease, arrhythmias, and pericardial diseases (3). The heterogeneity in cardiovascular complications of cancer therapies can, in part, be attributed to the increasing number of cancer therapeutics used in clinical practice, each with unique pathogeneses. These include traditional chemotherapeutics such as anthracyclines, targeted therapies with trastuzumab or anti-angiogenic tyrosine kinase inhibitors, and immunotherapies. The importance of early detection of cardiotoxicity has become increasingly apparent; left ventricular ejection fraction (LVEF) recovery and cardiac event reduction have been shown to occur more frequently with early diagnosis and treatment of cardiotoxicity in patients with cancer therapy-induced cardiomyopathy (4). Accordingly, recommendations for managing potential cardiac dysfunction in patients receiving cardiotoxic therapies include LVEF monitoring with serial echocardiograms, population stratification by treatments received and pre-existing cardiovascular risk factors (e.g. age, hypertension), and surveillance with serum cardiac biomarkers (troponin) (3). Additionally, a staging system (stages A-D) for heart failure exists, spanning from any cardiotoxin-exposed patient (Stage A) to those with observed, asymptomatic LV dysfunction (Stage B) and symptomatic heart failure (Stage C and D) (5).

While such guidelines represent a step forward, recommendations are generally vague, and a deficiency in evidence-based data for clinical decision-making still remains. Interestingly, the limitations
of current approaches highlight the multiple features that make the field of cardio-oncology uniquely suited for precision medicine. For example, current risk stratification schemes only include basic clinical characteristics, and do not incorporate the wealth of information potentially offered by deep phenotyping. Even with the potential, distinct categorizations that may emerge from current risk stratification models, differences in clinical management remain largely limited to adjustments in surveillance frequency and medication dosing. Moreover, more precise characterization of disease with refined imaging measures and sensitive biomarkers represents relatively untapped tools with which to tailor treatments to underlying pathophysiologic differences in otherwise similar clinical presentations. Going forward, a multidisciplinary commitment to precision medicine approaches is needed, with molecular imaging and -omics sciences at the forefront of promising archetypes.

**PANOMICS: BIOMARKER IDENTIFICATION**

The value of deep phenotyping has been demonstrated in studies of common cardiovascular diseases (6). A natural extension of such studies has been to embrace principles of more precise clinical phenotyping in cardio-oncology. Consequently, recent research has focused on identifying circulating biomarkers of disease as a complementary approach to the pursuit of sensitive imaging metrics.

To date, most investigations have focused on troponin and brain natriuretic peptide (BNP), two well-established biomarkers of cardiac injury and stress whose elevated levels have been associated with cardiotoxicity (7,8). However, it still remains to be determined if systematic evaluation of these biomarkers improves outcomes, and there are currently no guidelines for cut-off values, timing of assessments, or therapy-specific considerations. These limitations compounded by a lack of biomarker specificity have motivated a search for more evidence-based biomarkers. For example, myeloperoxidase, growth differentiation factor-15, and asymmetric dimethylarginine have all been postulated as potentially useful biomarkers, as all play critical roles in oxidative stress or nitric oxide metabolism - potential mechanisms of anthracycline-induced cardiotoxicity (9,10). Additionally, a multi-marker approach aimed
at increasing sensitivity and specificity through the simultaneous evaluation of multiple contributory pathways has also been proposed, with elevations in troponin and myeloperoxidase demonstrating significant additive predictive value (9).

More recently, advancements in -omics technologies have amplified the clinical relevance of phenotyping in both magnitude and spectrum. -Oomics sciences seek to computationally identify patterns from sizeable networks of data to elucidate causative pathophysiology. In the case of therapy-induced cardiotoxicity, these sciences can elucidate novel disease metrics that can inform the personalization of medicine in cardio-oncology (Fig. 1). However, it is important to note the inherent limitation of a systems-based analysis which is not specific to the heart, for predicting cardiac effects. While -omic technologies applied in broad discovery studies may reveal measurable changes in select candidate markers, the lack of specificity of these markers for cardiac tissue must be considered; abnormalities in variables may be due to processes unrelated to the cardiovascular system. Thus, recognition of potential confounding variables has an important function in interpreting results of panomics for biomarker identification.

**Genomics**

Much of our understanding of the genetics of anthracycline-induced cardiotoxicity has come from genome wide association studies (GWAS) of childhood survivors, which have implicated single nucleotide polymorphisms (SNP) in carbonyl reductase (CBR) and hyaluronan synthase 3 (HAS3) as independent modifiers of anthracycline-related cardiomyopathy risk (11). CBRs catalyze the reduction of anthracyclines to cardiotoxic alcohol metabolites, while the HAS3 gene encodes for hyaluronan, a ubiquitous component of the extracellular matrix that plays a role in tissue response to injury. Additional studies in childhood cancer survivors have identified polymorphisms in genes that regulate intra-cellular transport (SLC28A3, SLC28A1) of anthracyclines as independent predictors of cardiomyopathy risk.
A study of adult hematopoietic cell transplantation patients treated with anthracyclines identified an association between a polymorphism in the doxorubicin efflux transporter (ABCC2) and cardiotoxicity (14). Another GWAS study of anthracycline cardiotoxicity suggested susceptibility locus at chromosome 1p36.21 near PRDM2 (p=6.5x10^{-7}) (15). Additionally, genetic polymorphisms in the HER2 gene (Ile 655 Val and Pro 1170 Ala) have also been identified as conferring increased risk for trastuzumab cardiotoxicity (16).

Circulating microRNA, a direct product of genomic profiles, also has significant potential for identifying subclinical cardiac damage in patients receiving certain therapies. This large class of noncoding small RNAs which circulate in the bloodstream, enter distant recipient cells, and regulate gene expression has already demonstrated potential as a biomarker of CVD. In a case-control study of 12 children, 84 microRNAs were profiled 24-48 hours and ~1 year following initiation of anthracycline chemotherapy, and showed an association between decreased LVEF and three specific microRNAs (17). In another study of 33 children receiving anthracycline chemotherapy and noncardiotoxic chemotherapy, profiling of 24 microRNAs at pre- and post-cycle timepoints revealed greater chemotherapy-induced dysregulation in patients receiving anthracyclines compared with those receiving noncardiotoxic chemotherapy (24-hour MANOVA; p=0.024) (18).

Proteomics

A similar and parallel approach has been implemented in biomarker identification using proteomics. In experiments on heart tissue samples from control rats and rats exposed to docetaxel and doxorubicin, nine proteins involved in energy production were found to be differentially expressed in control versus treatment groups, with higher levels of glyceraldehyde-3-phosphate-dehydrogenase associated with lower mortality (19). The importance of energy metabolism was also confirmed in daunorubicin-induced cardiotoxicity, with alterations in mitochondrial proteins involved in oxidative
phosphorylation and energy channeling along with increased proteins involved in autophagy, membrane repair, and apoptosis (20). Small human studies have also suggested the importance of the immune system in the pathophysiology of cardiotoxicity (21). Nevertheless, findings from proteomic studies are still largely preliminary.

**Metabolomics**

Variation in metabolite profiles uniquely reflects a spectrum of molecular influences, intricately linking changes in DNA sequences, cellular physiology, and environmental factors. Despite a lack of studies investigating metabolic changes specific to cancer therapy-related CVD, metabolomics has emerged as a potential tool for cardio-oncologists to use in characterizing chemical intermediates in a variety of biosamples, especially as abnormal cardiac metabolism has been increasingly linked to CVD. For cardio-oncology patients, metabolic studies of general CVD offer two main insights: an understanding of the pathophysiologic, metabolomic alterations that occur in specific disease states (i.e. heart failure and myocardial ischemia), and a potential approach to CVD risk prediction with novel biomarkers. For example, the sequential use of progressively macroscopic -omics, i.e. from genomics to clinical phenotyping, as an approach to modeling and tracking the course of a disease may be promising. This approach directly contrasts current paradigms in which changing clinical phenotypes are what motivate investigation into progressively microscopic pathophysiology, i.e. from clinical chemistry and imaging to genomics.

With respect to risk prediction, the ability to one day perform real-time monitoring of blood or urine metabolites could allow clinicians to detect novel molecular biomarkers associated with differential clinical trajectories. Similarly, changes in metabolite profiles over time, for example after administration of a drug, could be used to define an individual’s response to therapy and guide subsequent management.
PHENOMAPPING WITH IMAGING

The varying clinical manifestations and unpredictable timing of treatment-related CVD have made surveillance and diagnosis challenging. One attractive approach has been to use sensitive imaging measures for phenomapping the risk of developing subclinical and overt disease. Current strategies rely almost exclusively on measurements of LVEF, however, a lack of consensus criteria for meaningful cut-off values, inherent variability in LVEF values and assessment, and an inability to detect subclinical myocardial damage limit LVEF as a gold standard. The ensuing discussion focuses on prominent imaging indices used clinically and evaluates how emerging strategies may offer more comprehensive phenotypic data.

Left Ventricular Ejection Fraction

LVEF is the most commonly used measure of cardiac function in cancer patients. 2D echocardiography (2DE) is typically the modality of choice in measuring LVEF, as it is inexpensive, easily available, and avoids radiation exposure. However, concern over its reproducibility and inability to detect small decreases in LVEF have motivated many to explore advanced techniques including strain analyses, ultrasound-based contrast agents (recommended with suboptimal 2DE views), and 3D echocardiography (3DE). A recent prospective study comparing 2DE and 3DE in breast cancer patients found that temporal variability in LVEF decreased from 10-13% with 2DE to 5-6% with 3DE (22). Variability in LVEF measurement with 3DE is therefore more comparable to that with cardiac magnetic resonance imaging (CMR) (2%), and was proposed as the preferred modality by several organizations (23). Still, due to its operator dependence, decreased availability and increased cost, routine clinical use of 3DE has been challenging.

Multi-gated acquisition has also been used for LVEF assessment. In one of the largest studies using serial multi-gated acquisition, decreases in LVEF to less than 50% or by 10% from baseline
predicted heart failure in 19% of patients (24). Additionally, in a clinical trial involving 944 breast cancer patients treated with chemotherapy, LVEF decreases by multi-gated acquisition independently predicted cardiac events over a 7-year follow-up period (25). However, the lack of information beyond LVEF and radiation exposure have limited the use of multi-gated acquisition.

Myocardial Deformation and Mechanics

Myocardial deformation, as measured by echocardiographic tissue doppler imaging or speckle-tracking echocardiography (STE), which makes use of a computerized algorithm to track natural echocardiographic signals, has recently emerged as a novel imaging measure. Deformation can be characterized by strain and/or strain rate (a measure of longitudinal, radial, and circumferential dimensions in peak systole), twist, or torsion. A major appeal of this approach comes from evidence that changes in strain can be observed prior to changes in LVEF and can predict cardiotoxicity (26).

Most recently, 3D STE has been proposed as a new modality to track linear myocardial deformation in multiple dimensions simultaneously, torsion, and mechanical desynchrony. Initial results have demonstrated advantages over 2D STE, including faster image acquisition, improved accuracy, and more complete analysis (27). Additionally, myocardial mechanics, including ventricular-arterial coupling and vascular stiffness, have been shown to predict declines in LVEF (28). Pending determination of optimal test timing and cut-off values along with further validation in large cohort studies, myocardial deformation and mechanics parameters may become key components of phenotyping in cancer-associated CVD.

Cardiac Magnetic Resonance (CMR) Imaging Metrics

CMR is an effective method for identifying CVD in cancer patients. The high spatial and temporal resolution and reproducibility of CMR enable the identification of inflammatory or infiltrative
processes, abnormal myocardial masses, and pericardial or valvular abnormalities, elucidating potential etiologies for reduced LVEF. One approach to characterizing cardiac tissue is to use CMR T1-weighted images and T1 mapping to detect myocardial inflammation, extent and topography of fibrosis, and pericardial tumor invasion with late gadolinium enhancement (29). Studies have demonstrated abnormal T1 in patients who received cardiotoxic chemotherapy, postulated to be due to either an increase in extracellular distribution volume or enhanced water exchange elicited by myocardial injury (30,31). Increases in extracellular volume (ECV) fraction and total ECV have also been observed in small studies of patients receiving anthracycline chemotherapy, potentially suggestive of edema and fibrosis. However, the mechanistic basis for this increase in ECV fraction was recently challenged in a study of patients who received anthracycline chemotherapy, with results suggesting that increases in myocardial ECV fraction may not necessarily be due to expansion of interstitial space, but may actually be related to decreases in myocellular volume and mass (32). It is also worth noting that T2 mapping has similarly demonstrated utility in detecting subclinical cardiotoxicity, as it enables qualitative and quantitative assessment of water content that may increase following myocellular or microvascular injury.

CMR also has a role in assessing cardiac function and blood flow. For example, cine white-blood steady-state free precession imaging allows for evaluation of wall motion abnormalities and calculation of volumes, LVEF, and mass (29). CMR can also be used to quantify myocardial strain by either spatial modulation of magnetization or displacement encoding with stimulated echoes techniques. In a prospective study of 53 cancer patients, CMR-measured global LV circumferential strain declined in parallel to declines in LVEF following treatment with anthracyclines (33). Further studies corroborating prognostic and diagnostic value along with considerations of cost and availability are warranted, and will likely be necessary before widespread implementation in clinical practice can occur.
Myocardial Perfusion Imaging

A common cardiovascular complication of cancer therapies is impairment of the coronary circulation either through direct vascular damage and/or accelerated atherosclerosis. As such, non-invasive methods for evaluating myocardial perfusion with such parameters as myocardial blood flow (MBF) and coronary flow reserve (CFR) quantification, are desirable in cardio-oncology care. For years single photon emission computerized tomography (SPECT) imaging has been one of the principle methods for evaluation of flow-limiting coronary stenosis in cardio-oncology patients, with the most commonly used radiotracers being 99mTc-labeled sestamibi and tetrofosmin. A number of SPECT-based studies have observed a high incidence of new resting myocardial perfusion deficits (up to 60% in some series) as early as six months post-radiation therapy (RT) in patients receiving left breast/chest wall RT compared to pre-RT SPECT scans (34,35). Importantly, these changes in perfusion 1) are typically limited to the anterior wall and apex, and/or follow the typical distribution of the left anterior descending (LAD) coronary artery (Fig. 2) (35), 2) correlate with the volume of heart irradiated (36), 3) remain relatively unchanged at 12- and 18-month follow-up compared to 6 months post-RT (35), and 4) correlate with cardiovascular symptoms in those with new perfusion defects (37).

More recently, cardiac PET has witnessed more widespread clinical implementation and is the current gold standard technique to assess myocardial perfusion due to its higher spatiotemporal resolution, count sensitivity, and accuracy when compared to SPECT. Additionally, PET tracers including N-13 ammonia (13NH3) and Rubidium-82 (82Rb) have superior pharmacokinetic properties due to a greater myocardial net uptake rate at higher coronary flows compared to their SPECT counterpart, (38) and combined with dynamic imaging allow for noninvasive quantification of MBF at baseline and during pharmacologic hyperemia. Stress-MBF is typically impaired in the presence of flow-limiting coronary stenosis, however, in its absence, impaired stress-MBF, and thereby CFR, is considered a robust marker of coronary microvascular dysfunction. Yet, limited clinical studies have applied cardiac PET to monitor
for cancer therapy-related cardiotoxicity. In one of the few human studies applying PET in cardio-oncology, a cross-sectional analysis of 35 patients who underwent 82Rb PET/CT at a median interval of 3.6 years post-RT found significant inverse correlations between CFR and specific RT dose-volume metrics, including a significant inverse correlation between global LV CFR and increasing mean heart RT dose (R= -0.4, p=0.03), and regionally between CFR of and radiation dose to the left anterior descending artery (R= -0.5, p=0.002) (39).

Despite these advancements, several challenges have limited the use of these PET imaging techniques in cardio-oncology patients. For one, limited data are available with regard to optimal thresholds used to distinguish pathological from normal hyperemic MBF and CFR. Additionally, optimal thresholds may vary depending on radiotracer and software. Lastly, human clinical trials are needed to assess the diagnostic and prognostic value of myocardial perfusion imaging parameters before clinical implementation can occur.

**Cardiovascular Molecular Imaging Techniques**

Molecular imaging exploits radio-labeled imaging probes to elucidate the biomolecular events that underlie clinical phenotypes. In the field of cardiology, molecular imaging has demonstrated years of clinical utility and value, with both SPECT and PET at the center of many diagnostic approaches. Recently, such imaging techniques have demonstrated the potential to play a similar role in the field of cardio-oncology. The radiotracers, mechanisms of uptake, and targets for imaging cancer-related cardiotoxicity can be found in Table 1.

Metabolic imaging with fluorodeoxygluocse (FDG) PET/CT remains the backbone of molecular imaging. FDG is a sensitive molecular probe for the investigation of cancer-related cardiotoxicity since tissue injury (40), inflammation (41), and hypoxia/ischemia (42), are potent stimuli for glucose
transporter expression. In a canine model of radiation-induced cardiotoxicity, focal myocardial FDG uptake (typically in the anterior wall) corresponding to the irradiated field was observed 3 months post-RT (Fig. 3) (40). On histopathology, all dogs displayed areas of myocardial damage in the irradiated field consisting of perivascular fibrosis and mild myocyte degeneration and mitochondria injury, but interestingly, no inflammatory cell infiltration was detected, implying that FDG accumulation within the irradiated field was mediated through mechanisms other than inflammation, including tissue hypoxia/ischemia due to microvascular damage and/or changes in metabolism caused by mitochondrial injury (40). Similar observations have been made in a retrospective study of 39 lung cancer patients treated with RT (Figs. 4 and 5), in which 47% of patients receiving 20 Gy to ≥5 cm³ of the heart developed increased FDG uptake versus 0% of the patients who received 20 Gy to <5 cm³ (p=0.02), again supporting the role of FDG as a marker of RT-induced myocardial injury (43).

On the other hand, with the development of a number of new molecular imaging agents, many of which target early markers of cardiotoxicity (sympathetic nerve terminals, angiogenesis, reactive oxygen species, and apoptosis), the potential impact of molecular imaging on patient outcomes in cardio-oncology has become even greater. In one study of rats treated with anthracycline, PET imaging with ¹⁸F-DHMT, a marker of reactive oxygen species, revealed an elevation in cardiac superoxide production at 4 weeks post-treatment, compared to a decrease in LVEF on echocardiography detectable only at 6 weeks post-treatment (44). In one human study of 20 breast cancer patients treated with either anthracycline and trastuzumab or anthracycline alone, the ratio of ¹²³I-mIBG uptake between the heart and mediastinum on scintigraphic images decreased in 25% of patients, and the washout rate increased in 82% of patients compared to matched controls, demonstrating the potential utility of ¹²³I-mIBG as a marker of cardiac sympathetic activity and cardiotoxicity (45). However, in a more recent study of 89 asymptomatic patients previously treated with anthracyclines, neither the heart to mediastinum ratio nor the washout rate were able to discriminate anthracycline-exposed patients from controls (46).
As molecular imaging techniques with newer probes are still under study, the role for these approaches remains to be defined. Still, molecular imaging innovations occurring concurrently with those in -omics technologies have the potential to reveal radiologically-targetable, novel molecular biomarkers. Thus, a next step in cardio-oncology care could involve the coupling of biomarker discovery with radiographic technologies to capture real-time molecular events, ultimately creating a key role for nuclear medicine in the future of cardio-oncology.

**RADIATION-INDUCED CARDIOTOXICITY**

Radiation-induced CVD represents a nuanced clinical challenge. A recent retrospective study reported that compared with non-irradiated patients, chest radiation therapy (RT) patients have a 2% and 23% higher absolute risk of cardiac morbidity and death at 5 and 20 years post-treatment, respectively (47). In contrast to chemotherapy-induced toxicity, which is largely dependent on metabolic rates and/or genetic differences in drug metabolism, RT toxicity can be directly linked to received dose to the heart or nearby organs. However, in line with novel precision medicine strategies for drug-induced CVD, radiation oncologists have proposed therapeutic approaches such as the use of gene-expression assays to identify potential SNPs that may predict adverse reactions to radiotherapy and guide radiation dosing guidelines - a concept known as the “genomic-adjusted radiation dose” (48). Furthermore, as cardiovascular radiation damage is postulated to be caused in part by monocyte activation and the formation of inflammatory plaque and intimal fatty streaks - major components of atherosclerosis – leading to vessel occlusion, perfusion defects, and myocardial necrosis and fibrosis, RT patients are uniquely suited for surveillance and risk stratification with radionuclide imaging techniques.

**Radiation-Induced Atherosclerosis**

A potential approach to early diagnosis and surveillance of radiation-induced atherosclerosis is to use molecular imaging to predict likely outcomes based on molecular evaluation of plaque rupture
propensity. For example, PET imaging of plaque macrophages with $^{18}$F-FDG could enable the differentiation of stable and rupture-prone plaques by quantifying macrophage density and $^{18}$F-FDG uptake, an approach which capitalizes on the role of activated macrophages in digesting atheroma fibrous caps and triggering plaque rupture (49). An alternative approach could make use of ultra-small superparamagnetic iron oxide (USPIO) which tends to selectively accumulate in rupture-prone plaques; in a prospective human study, USPIO-MRI was used to serially monitor the effect of atorvastatin on plaque inflammation (50).

Apoptosis of smooth muscle cells or macrophages within a plaque, causally linked to plaque rupture, is another promising imaging target. Specifically, the translocation of phosphatidylserine to the external surface of the cell membrane is a targetable, early event occurring in apoptotic cells. For this reason, $^{99m}$Tc-labeled annexin V, a plasma protein with high affinity for phosphatidylserine, has been tested in a pilot study to detect cellular apoptosis associated with unstable human carotid plaques (51). Applying this technology to doxorubicin-treated patients, imaging with $^{99m}$Tc-HYNIC-annexin V demonstrated the ability to detect dose-dependent cell death before echocardiography (52). Future prospective studies in larger cohorts will help determine its true sensitivity and specificity.

While these imaging procedures could potentially revolutionize cardiovascular risk stratification algorithms for cardio-oncology patients, they have not yet been studied specifically in RT populations, so translation of their findings remains to be determined. Nevertheless, as our understanding of the key molecular determinants of RT-induced CVD grows, the use of molecular imaging specifically tailored to the biomolecular targets identified by -omics approaches could ultimately create a platform for personalized, multidisciplinary cardio-oncologic care.
CONCLUSION

Cardiotoxic cancer therapies continue to contribute to the global burden of CVD. Further resources should be invested in the pursuit of a more personalized approach to clinical management, centered on deep phenotyping with -omics. Molecular imaging represents a unique component to this potential strategy for diagnostics and surveillance, enabling precise exploitation of underlying pathophysiology. Trends towards individualized medicine will be a critical feature of future paradigms of evidence-based, risk-specific precision medicine in cardio-oncology, ultimately reducing cardiovascular morbidity and mortality in a growing population of cancer patients and survivors.

DISCLOSURES

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REFERENCES


(32) Hundley W, Jordan J. When left ventricular extracellular volume fraction changes after anthracyclines: Is it due to a change in the numerator, denominator, or both? *JACC Cardiovasc Imaging.* 2018;11:1056-1058.


Deep Phenotyping in Cardio-Oncology Patients

A

Demographics
Exposures
Clinical Evaluation

All Cardiotoxic Therapy-Exposed Patients

• • • • •

LVEF < 50%

• • •

B

Radiomics
Cell Biomics
Metabolomics
Proteomics
Transcriptomics
Genomics

Precision Phenotyping Based on:

Genomic Biomarker
Molecular Imaging
Metabolome Profile

Improved Risk Stratification
Personalized Preventative Strategies
Tailored Surveillance Strategies
Targeted Cardio-protection
Better Clinical Outcomes
Figure 1. (A) Current approaches to cardio-oncology patients involve phenotyping with demographics, exposure history, and clinical evaluation (including basic labs and imaging). This method identifies disease phenotypes such as decreased LVEF to broadly guide therapeutic management. (B) Individuals with the same phenotype (e.g., LVEF<50%) can be further stratified through precision phenotyping and applying –omics to elucidate defining biological data (e.g. – genomic biomarker (purple), molecular imaging (gold), metabolome profile (green)), resulting in more targeted management and better outcomes.
Figure 2. Myocardial perfusion SPECT images pre- and 6 months post-radiation therapy (RT) for left-sided breast cancer demonstrate the development of a new perfusion deficit in the anterior wall and apex consistent with radiation-induced myocardial damage. Short axis = SA; vertical long axis = VLA
Figure 3. Radiation-induced myocardial injury in dogs. A. Dose distribution contrast-enhanced cardiac CT. B. FDG PET/CT showing no myocardial uptake pre-RT. C. Focally intense FDG uptake along the LV apex matching the irradiated field 3 months post-RT. Adapted and reprinted with permission from Ref. 40.
Figure 4. Radiation pneumonitis, pericarditis, and myocarditis. A patient with left upper lung cancer in close proximity to the LV lateral wall (panel A arrows) was treated with surgical resection and RT. Please notice the development of a medium-sized pericardial effusion (panel C arrows), pneumonitis changes (panel D arrows), and possibly focal FDG uptake along the LV lateral wall on FDG PET/CT imaging and fibrotic changes on cardiac MRI (figure 5) approximately 6 months post-RT.
Figure 5. Radiation-induced myocardial injury/myocarditis, continuation of figure 4. Patchy late gadolinium enhancement and focally increased FDG are seen along the anterolateral and inferolateral wall segments on cardiac MRI and PET/CT, respectively. Pericardial effusion is also visualized on MRI and CT.
### Table 1. Radiotracers, mechanisms of uptake, and targets for imaging cancer-related cardiotoxicity

<table>
<thead>
<tr>
<th>Molecular Target / Mechanism of Uptake</th>
<th>Cardiovascular Application</th>
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<tr>
<td>82Rb* Na/K-ATPase</td>
<td>Perfusion/flow quantification</td>
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<tr>
<td>13NH3* Glutamine synthetase activity</td>
<td>Perfusion/flow quantification</td>
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<td>99mTc-sestamibi* Mitochondrial function</td>
<td>Perfusion</td>
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<td>18F-FDG* Glucose transporters</td>
<td>Glucose metabolism</td>
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<td>123I-MIBG* Norepinephrine transporter</td>
<td>Sympathetic nerve integrity</td>
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<td>11C-hydroxyephedrine Norepinephrine transporter</td>
<td>Sympathetic nerve integrity</td>
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<td>11C-acetate Krebs cycle flux</td>
<td>Oxidative metabolism</td>
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<tr>
<td>123I-BMIPP α-oxidation and β-oxidation</td>
<td>Fatty acid metabolism</td>
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<td>111In-Trastuzumab HER2</td>
<td>Apoptosis</td>
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<td>111In-Antimyosin Exposed myosin</td>
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<td>99mTc-RP805† Activated MMPs</td>
<td>Remodeling</td>
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<td>18F-DHMT† Reactive oxygen species</td>
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<td>99mTc-Annexin V† Exposed phosphatidylerine</td>
<td>Apoptosis</td>
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<td>18F-CP18† Caspase 3 activity</td>
<td>Apoptosis</td>
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* FDA-approved radiotracer for clinical use
† Pre-clinical phase. No published human data in cardiotoxicity
FDG: fluorodeoxyglucose; MIBG: metaiodobenzylguanidine; BMIPP: beta-methyl-p-iodophenylpentadecanoic acid; HER2: human epidermal growth factor receptor 2; MMP: matrix metalloproteinases