AperTO - Archivio Istituzionale Open Access dell'Università di Torino

Sex differences in anthracycline-induced cardiotoxicity: the benefits of estrogens

This is the author's manuscript

Original Citation:

Availability:

This version is available <http://hdl.handle.net/2318/1736812> since 2020-04-20T17:26:24Z

Published version:

DOI:10.1007/s10741-019-09820-2

Terms of use:

Open Access

Anyone can freely access the full text of works made available as "Open Access". Works made available under a Creative Commons license can be used according to the terms and conditions of said license. Use of all other works requires consent of the right holder (author or publisher) if not exempted from copyright protection by the applicable law.

(Article begins on next page)

Sex differences in anthracycline-induced cardiotoxicity: the benefits of estrogens

Christian Cadeddu Dessalvi¹, Alessia Pepe², Claudia Penna³, Alessia Gimelli⁴, Rosalinda Madonna⁵, Donato Mele⁶, Ines Monte⁷, Giuseppina Novo⁸, Cinzia Nugara⁸, Concetta Zito⁹, Javid J Moslehi¹⁰, Rudolf A de Boer¹¹, Alexander R Lyon¹², Carlo G Tocchetti^{13,14}, Giuseppe Mercuro¹

Anthracyclines are the cornerstone for many oncologic treatments, but their cardiotoxicity has been recognized for several decades. Female subjects, especially before puberty and adolescence, or after menopause, seem to be more at increased risk, with the prognostic impact of this sex issue being less consistent compared to other cardiovascular risk factors. Several studies imply that sex differences could depend on the lack of the protective effect of sex hormones against the anthracycline-initiated damage in cardiac cells, or on differential mitochondria-related oxidative gene expression. This is also reflected by the results obtained with different diagnostic methods, such as cardiovascular biomarkers and imaging techniques (echocardiography, magnetic resonance, and nuclear medicine) in the diagnosis and monitoring of cardiotoxicity, confirming that sex differences exist. The same is true about protective strategies from anthracycline cardiotoxicity. Indeed, first studied to withstand oxidative damage in response to ischemia/reperfusion (I/R) injury, cardioprotection has different outcomes in men and women. A number of studies assessed the differences in I/R response between male and female hearts, with oxidative stress and apoptosis being shared mechanisms between the I/R and anthracyclines heart damage. Sex hormones can modulate these mechanisms, thus confirming their importance in the pathophysiology in cardioprotection not only from the ischemia/reperfusion damage, but also from anthracyclines, fueling further cardio-oncologic research on the topic.

Keywords Anthracycline cardiotoxicity . Gender differences . Pathophysiology, monitoring, and protection from anthracycline cardiotoxicity

CC, AP, and CP share first authorship

GM and CGT share senior authorship

¹ Department of Medical Sciences and Public Health, University of Cagliari, Cagliari, Italy.

² Magnetic Resonance Imaging Unit, Fondazione G. Monasterio C.N.R.- Regione Toscana, Pisa, Italy

³ Department of Clinical and Biological Sciences, University of Turin, Turin, Italy

⁴ Nuclear Medicine Unit, Fondazione G. Monasterio C.N.R.- Regione Toscana, Pisa, Italy

⁵ Center of Aging Sciences and Translational Medicine - CESI-MeT, “G. d’Annunzio” University, Chieti, Italy.

⁶ Cardiology Unit, Emergency Department, University Hospital of Ferrara, Ferrara, Italy.

⁷ Department of General Surgery and Medical-Surgery Specialties- Cardiology-University of Catania, Catania, Italy

⁸ Department of Cardiology, University of Palermo, Palermo, Italy

⁹ Department of Clinical and Experimental Medicine – Cardiology, University of Messina, Messina, Italy

10 Vanderbilt Ingram Cancer Center, Department of Medicine, Vanderbilt University Medical Center, Nashville, TN, USA

11 University of Groningen, University Medical Center Groningen, Department of Cardiology, Hanzeplein 1, 9713 GZ, Groningen, the Netherlands

12 Royal Brompton Hospital and Imperial College London, UK

13 Department of Translational Medical Sciences, Federico II University, Naples, Italy

14 Interdepartmental Center for Clinical and Translational Research (CIRCET), Federico II University, Naples, Italy

Keywords: anthracyclines cardiotoxicity; gender differences; pathophysiology, monitoring and protection from anthracyclines cardiotoxicity

Correspondence to

Carlo Gabriele Tocchetti, MD, PhD, FHFA

Dipartimento di Scienze Mediche Traslazionali

Centro Interdipartimentale di Ricerca Clinica e Traslazionale (CIRCET)

Universita' degli Studi di Napoli Federico II

Via Pansini 5, Edificio 2

80131 Napoli

ITALY

Phone: +39-081-746-2242

Fax: +39-081-746-2246

carlogabriele.tocchetti@unina.it

ORCID [0000-0001-5983-688X](https://orcid.org/0000-0001-5983-688X)

INTRODUCTION

Anthracyclines are first-line therapies for the treatment of many malignancies; unfortunately, their efficacy is limited by cardiotoxicity (CTX), which is dose-dependent. Recent clinical guidelines on cardiotoxicity from anticancer drugs highlight that identification of subjects at a higher risk is a fundamental step in successful screening and pre-emptive treatment [1-3]. Many factors including cumulative dose, body mass index $>30\text{kg/m}^2$ (especially for anti-HER2 compounds) [4], age (elderly and pediatric population treated with anthracyclines) [5-8], concomitant or previous radiation therapy, previous cardiotoxic anticancer therapies, pre-existing cardiovascular disease (CVD) [9], demographic and other CV risk factors, as well as common lifestyle and genetic risk factors may predispose patients to CTX, already at mild to intermediate doses of anthracyclines [1,2,5, 10-12].

In particular, the association between obesity ($\text{BMI} \geq 30 \text{ kg/m}^2$) and high risk of heart failure has been well established. In addition, previous studies demonstrated that obesity was an important prognostic factor that affected both overall survival and disease-free survival in patients with breast cancer [13]. The mechanisms by which obesity could negatively influence cardiotoxicity are affected by numerous confounding factors. Obesity may increase the expression of pro-inflammatory adipokines and downregulate the anti-inflammatory adipokines, which could result in an adipokine imbalance and maintain a chronic inflammatory state to promote the development of cardiovascular diseases. Second, patients with obesity are more sensitive to the cardiotoxic effects of anthracyclines; an animal model of rats with a high lipid diet showed similar results. Finally, obesity is significantly associated with activation of neurohormones, increased oxidative stress, increased hemodynamic load, and remodeling of the left ventricle[14] .

In addition, ErbB2 (or HER2) seems to be a modulator of oxidative stress, with anti-ErbB2 drugs being able to block protective mechanisms triggered by ErbB2, thus enhancing the oxidative damage induced by anthracyclines. The interactions between anthracyclines and trastuzumab have been extensively studied. Importantly, in breast cancer, coadministration of trastuzumab with ANTs in the first trials increased the latter's toxicity and is now avoided [15–18].

Also, protocols with elevated anthracyclines dosages (cumulative doses higher than 350 mg/m²) are being seldom used, due to the increased risk of cardiotoxicity.

It is important to assess the best protocols for early identification of these high-risk patients and for follow-up during cancer therapies, in order to be able to obtain optimal protection from cardiac damage [19]. This paper describes the importance of sex differences in pathophysiology of anthracycline cardiotoxicity, in the assessment and monitoring imaging techniques of this cardiomyopathy, and in the protection from cardiac damage induced by this chemotherapeutic. In terms of protection, here we focus on the role of estrogens, acknowledging that in the CECCY trial, no difference was seen between carvedilol and placebo in both subgroups pre- and post-menopause [20], while in the MANTICORE [21], there is no mention of menopausal status.

SEX DIFFERENCES OF ANTHRACYCLINE-INDUCED CARDIOTOXICITY

Anthracyclines can lead to cell dysfunction and death by interfering with mitochondrial function, bioenergetics, signaling pathways and redox balance [5,22]. Most of these targets are known to exhibit sexual dimorphism, like “redox features” of cells (i.e., different aspects of redox-associated molecules and enzymes in regard to gender differences in terms of the intracellular production and biochemical activity of reactive species) and expression of mitochondria-related genes at different ages [23,24]. In addition to pharmacodynamics, sex-specific differences in pharmacokinetics (absorption, distribution, excretion) may have important clinical consequences, influencing drug side effects. Regarding doxorubicin and its main metabolite doxorubicinol, important intra- and inter-patient variations in pharmacokinetic parameters have been demonstrated [25]. Several authors showed that men have a significantly higher doxorubicin clearance than women [26]. This was supported by the finding of a higher proportion of doxorubicinol detected in the men, which could be related to a greater aldo-ketoreductase activity. [27]. Moreover, doxorubicin and doxorubicinol might accumulate following a reduced expression of p-glycoprotein in females, leading to higher rate of cardiotoxicity [28].

A comparable sex-based difference has been reported for the pharmacokinetics of epirubicin [27]. Therefore, sex-specific characteristics in anthracycline-induced cardiotoxicity can be potentially expected based on both the pharmacodynamic and pharmacokinetic profile.

Experimental data point toward better resistance of females regarding CTX with involvement of mitochondria and less oxidative stress, in fact very few studies have been conducted in humans, with female patients in clinical studies rather appearing to be more susceptible to doxorubicin-induced CTX [1,29,30]. This apparent paradox may be explained because both age and the menopausal state of female patients seem to be the two most important determinants of the sex-specific differences observed in the clinical setting, with higher susceptibility of prepuberal girls and post-menopausal women (Figure 1). Studies in young children receiving anticancer drugs for hematological malignancies [29,31] suggest that prepuberal girls are more susceptible to develop early or late cardiac toxicity than boys of the same age. These data are consistent with absence of female hormones at this age. Unfortunately, no survey has been conducted to specifically assess sex differences in the occurrence of anthracycline cardiotoxicity in adults. Studying the cardiac status of long-term survivors (at least 5 years after therapy) and estimating the features of subclinical cardiotoxicity induced after conventional treatment of lymphoma with doxorubicin, some authors found that male sex was related, together with other factors, to the development of subclinical cardiotoxicity [32]. In cohort studies of cancer survivors, the predicted 10-year cumulative incidence of congestive heart failure for males without or with preexisting cardiac disease is higher than in females [33,34].

In addition, the majority of breast cancer patients are post-menopausal [35], supporting the hypothesis of protection conferred by female sex hormones against doxorubicin cardiotoxicity. Interestingly, doxorubicin itself seems to be likely to cause premature ovarian failure, with a sharp decline in female sex hormones [36].

In studies concerning hematological malignancies, both sexes were treated risk factor for adverse cardiac events and the authors attributed this to a higher baseline cardiac disease in males [33]. In addition, data with hematological malignancies in postmenopausal women suggest that

similar baseline cardiovascular health in elderly men and women leads to similar susceptibility to anthracycline-induced cardiotoxicity [37]. Oxidative stress is well known to play a major role in anthracycline-induced cardiotoxicity. Reactive Oxygen Species (ROS) include several oxygen radicals such as Superoxide ($O_2^{\bullet-}$) and hydroxyl radicals ($OH^{\bullet}$), and non-radicals molecules, e.g. hydrogen peroxide (H_2O_2) and singlet oxygen (1O_2). Also the Reactive Nitrogen Species (RNS) comprise radicals such as Nitric Oxide ($NO^{\bullet}$) and Nitric Dioxide ($NO_2^{\bullet}$), and non-radicals, e.g. nitrous acid (HNO_2) and dinitrogen tetroxide (N_2O_4)

Overproduction of ROS and RNS induced by anthracyclines determines redox stress, which induces cardiac injury [5], including DNA damage and lipid peroxidation, leading to membrane injury and/or apoptosis and alterations of the enzymatic activity of the mitochondrial redox system. Among altered enzymes are those of the respiratory complexes, the enzymes of Krebs cycle, oxidative phosphorylation, and β -oxidation and nitric oxide synthases (NOSs) [18]. While cross-talk in breast cancer cell lines between estrogen receptors and ROS/Notch/Wnt pathways may play an important role in regulating cell death, differentiation, and angiogenesis [38, 39], a complex inter-relationship between estrogen receptors and enzyme activity involved in redox mechanisms may sustain different oxidative stress that mediates cardiotoxicity in a gender-specific manner. In support of this hypothesis, the work of Gonzalez and colleagues [40] demonstrates that adult tumor-bearing male SH rats are more cardiosensitive to doxorubicin treatment than female or hormone-deficient animals. These results suggest that reproductive hormones regulate doxorubicin-induced cardiotoxicity and the selective cytotoxic mechanism likely functions through the greater activation of oxidative stress and apoptosis in male SH rats [40].

SEX DIFFERENCES IN ASSESSMENT AND MONITORING OF ANTHRACYCLINES TOXICITY

Beside the pathophysiology of anthracycline-cardiotoxicity, sex differences may also prompt differential assessment and monitoring of patients who are treated with these drugs.

Transthoracic echocardiography (TTE) is the most useful tool to monitor cardiac function in patients undergoing chemotherapy because of its safety, availability and low cost [41]; nonetheless, there are some sex-related differences that should be considered during a TTE evaluation of patients with suspected cardiotoxicity. Indeed, pathophysiological and epidemiological features, specific of female sex, often imply higher heart rate, smaller ventricles with seemingly higher contractility, supraventricular arrhythmias, and higher prevalence of HFpEF [42,43.]. Although LVEF is a gross and poorly sensitive parameter to evaluate cardiotoxicity, it is still the most used parameter for follow-up of oncologic patients [1,44]. The last TTE recommendations indicate as normal a LVEF (by 2D-modified Simpson's rule) >52% for men and 54% for women [45]. Cardiotoxicity has been defined as a decrease in LVEF of >10% to a value <53% [44] (or <50% independent from sex) [1]. However, LVEF is poorly sensitive for the detection of small changes in LV contractility, particularly if these are limited to few segments and in hypertrophic, small ventricles, as often occurs in women. [46,47]. Thus, it should be combined with wall motion score index (WMSI) calculation which enables the identification of early regional damage very common in women treated with trastuzumab [44](Figure 2). A 5-unit fall of contractile reserve at low-dose dobutamine stress echocardiography in women with breast cancer seems to be able to predict subsequent LVEF reduction <50% [48].

Importantly, WMSI has been applied in previous clinical studies. A work from the early son hematological malignancies suggested that the 16-segment evaluation of LV function at rest by WMSI might be superior to global 2D measurement of EF. Moreover, gradual worsening of WMSI during anthracyclines paralleled the decline of radionuclide EF [49]. Also, autopsy studies have shown that the cardiac injury caused by anthracyclines is patchy, and at times is limited to one or more ventricle walls [50]. Therefore, segmental abnormalities can be detected before any global systolic LV dysfunction is apparent. More recently, in childhood cancer survivors (CCS) treated with anthracyclines, platinum, and/or radiotherapy between 1976 and 1999, at 18 years post-treatment, there was an increased prevalence of abnormal WMSI compared to controls, and NT-proBNP was associated to increased WMSI [51]. In addition, CCS with persistent LV regional wall motion

abnormalities (WMA) show reduced LV myocardial performance, evaluated through 3D speckle tracking echocardiography, compared with those without WMA, despite a preserved LVEF. Multiple linear regression analysis identified global radial strain as a significant determinant of the existence of WMA in these patients [52]. 9Indeed, global systolic longitudinal strain (GLS) was recognized to accurately predict a subsequent decrease in LVEF in several studies on women treated with anthracyclines with or without additional agents [53]. A relative percentage reduction of GLS of >15% from baseline is considered abnormal and a marker of early LV subclinical dysfunction in the last position statement [1], based on the results of a study in women with breast cancer treated with trastuzumab, with or without anthracyclines. In this context, it is important to refer to 248 mean values for GLS according to sex and age, because values of GLS are slightly higher in women than in men [54]. Kocabay and colleagues reported a mean normal GLS of 20.7 ± 2 for men and 22.1 ± 1.8 for women [55]. These values are almost identical to the ones reported by the Japanese Ultrasound Speckle Tracking of the Left Ventricle (JUSTICE) study for the same vendor, showing an effect of gender on GLS values [56]. Interestingly, in early 2019, Barros and colleagues analyzed prospectively the role of LV WMSI among echocardiographic parameters in the prediction of development of cardiotoxicity in breast cancer patients undergoing chemotherapy. In the multivariate analysis using the logistic regression model, LVWMA, LV systolic dimension increase and GLS reduction were strongly associated with cardiotoxicity and could be use full in risk stratification of these patients [57]. In summary, during cardiac toxicity monitoring it is very helpful to refer to current recommendations on the cardiac chamber normal values [45]. It is also strongly recommended not only to use TTE parameters of global function such as EF, volumes, and GLS, but also to refine the analysis through the regional function assessment (WMSI and bull's eye map of longitudinal strain), in both baseline patient's examination and during follow-up [58]. As for the other cardiac evaluations (valves, atria, right ventricle, pericardium), no particular gender-related difference was found in TTE studies on cardiotoxicity diagnosis and monitoring.

Cardiovascular magnetic Resonance (CMR) is an ionizing radiation-free imaging method accepted as the gold standard for quantifying biventricular parameters. It is well known that biventricular function parameters are strongly correlated to gender and age [59]. Thus, in clinical practice it is strongly recommended to apply cut off values according to gender and normalized to body surface area (BSA) for the volumes and the mass in order to accurately detect cardiotoxicity related to anthracyclines. Many studies showed a significant decrease in LVEF due to chemotherapy treatment. In adult patients LVEF changes seem to be not affected by sex [60]. One study involving 62 long-term survivors of childhood cancer showed a trend toward a male predominance among those with an abnormal LVEF (29% vs 9%; $P=0.053$) and RVEF (39% vs 18%; $P=0.057$) [61]. Post-treatment biventricular volumes and LV mass were significantly higher in males, but these parameters were not normalized to BSA [62]. Moreover, these results can be a consequence of the different morphology of male and female hearts, due to different adrenergic and hormonal states [63], body weight, height, different muscular mass, and body conformation. Importantly, sex differences can be observed in hemodynamics only in patients with normal LV function, but not with HF: Mitoff and colleagues speculated that the intrinsic or extrinsic factors responsible for sex differences in patients with normal cardiac function may be masked by the HF disease state or its treatment [64].

CMR provides an accurate and reliable evaluation of myocardial deformation by tagging techniques and few studies have demonstrated a significant decrease in longitudinal and circumferential deformation in chemotherapy-treated patients compared with controls [65]. In 46 asymptomatic post chemotherapy pediatric patients average circumferential and longitudinal strain magnitude was lower among male subjects, but this finding was due to the fact that gender affects normal values [62] and this issue was not taken in account in the study.

In the evaluation of anthracycline-induced CTX, CMR has the unique feature of providing information on tissue characterization. In fact, chemotherapeutic agents can cause oedema and hyperaemia, and even cellular necrosis and subsequent fibrosis. No study has evaluated if the prevalence of oedema was different between males and females. Macroscopic fibrosis can be detected

by means of the late gadolinium enhancement (LGE) technique and its presence is a strong prognosticator in all cardiomyopathies [44]. Diffuse myocardial fibrosis can be detected by T1 mapping with the evaluation of the extracellular volume (ECV). Toro-Salazar et al. found that T1 values after contrast administration were significantly lower in 46 long-term survivors in comparison with volunteers, revealing mild diffuse fibrosis [62]. In this study, as well as in a study by Tham et al [66], treated females showed higher mean ECV and lower postcontrast myocardial T1 values compared to males. Similar trends were detected in studies involving healthy subjects, and thus may reflect general sex differences.

In recent years, several biomarkers have been tested in the context of anticancer anthracycline-induced cardiotoxicity [67]. However, their use is severely hampered by important sex-specific differences [68]. Specifically, high sensitivity-troponin levels are about 50% lower in women than in men, while NT-proBNP levels are 50% higher [69]. Cardinale et al observed a greater prevalence of women in a group of patients with L-TnI levels >0.08 ng/mL with a further increase; they also noted that in breast cancer TnI was most commonly positive [70]. However, another study showed that gender had no influence on the occurrence of cTnT positivity [71].

Use of BNP or N-terminal pro-B-type natriuretic peptide (NT-proBNP) to detect subclinical CTX is under investigation and results of published studies are controversial [72-74]. Also, increases of gal3 were found to be not predictive of anthracycline-induced CTX as defined by echo-derived LVEF reduction, and no data are available about differences between the sexes [75].

SEX DIFFERENCES IN CARDIOPROTECTION

Acknowledgment of sex differences in the mechanisms and clinical manifestations of anthracyclines cardiotoxicity may help elucidate cardioprotective approaches and develop therapeutic strategies. The concept of cardioprotection was introduced in the 1960s with pharmacological and reperfusion interventions, following an ischemic damage, and has evolved to include several

strategies that may save the functionality and vitality of cardiac cells jeopardized by ischemia/reperfusion (I/R) or toxic insult [76].

Despite the fact that experimental and pre-clinical studies clearly demonstrate cardioprotective benefits with conditioning strategies, their translation into clinical therapy has been disappointing [77]. This is due to several reasons, including the complexity of the cellular mechanism of cardioprotection. Furthermore, additional confounding factors such as age, comorbidities, comedications and gender may affect the injury, as well as the endogenous cardioprotective aspects triggered by conditioning procedures [78,79]. Cardioprotective strategies that have been first adopted against the oxidative damage that characterizes the ischemia/reperfusion injury, have different outcomes according to the sex analyzed. Estrogens may play a role in modulating protection from anthracyclines cardiotoxicity, too, but their roles in cardioprotection deserve further studies.

A number of studies analyzed the differences in I/R response between male and female hearts, and there are numerous epidemiological data reporting that premenopausal females have a reduced risk for cardiovascular disease [80–86]. In the majority of studies, estrogens are considered responsible for the better tolerance to I/R by female hearts [87], as we have already described in a previous section about estrogens protecting from cardiac injury from anthracyclines. Endogenous estrogens may affect with tonic and phasic effects cardiovascular homeostasis in premenopausal females and may interfere with the development of several cardiovascular diseases, including coronary artery disease (Fig. 3). Acting on three receptors (ERs), namely ER α , ER β , and GPER, estrogens exert transcriptional regulation. Moreover, GPER is implicated in rapid, phasic signaling via RISK pathway. Indeed, estrogen receptor activation induces the activation of PI3-kinase (PI3K)/Akt signaling converging on nitric oxide (NO) synthases (NOS) [87]. Nitric oxide plays a central role in mechanisms of cardioprotection and may be a key factor in gender differences. Estrogen-induced NO production favors S-nitrosylation (SNO) and closure of L-type Ca^{2+} channels so that cytosolic and mitochondrial calcium overload during ischemia and early reperfusion is reduced [88]. Mechanisms related to the control of Ca^{2+} homeostasis and mitochondrial function

may be important in determining gender differences not only in response to I/R, but also to anthracycline challenge. The importance and gender differences of NO pathway in regulating the expression of genes related directly and indirectly to cardiomyocyte Ca^{2+} handling have been recently confirmed by Bienvenu and coworkers [89].

Recently, Murphy and colleagues have demonstrated a characterization of the sex-dependent cardiac SNO proteome [90]. Interestingly, higher level of SNO in membrane fractions derived from female hearts with respect the level found in male hearts have been described, [91]. The same group also showed that female hearts display higher cardiac eNOS expression and that the estrogen-dependent NOS activation/phosphorylation induces NO production, which in turn enhances SNO protein levels and consequently a cardioprotective phenotype occurs. Conversely, in male hearts, the expression of eNOS is reduced, with minor production of nitric oxide derivative and SNO protein levels [90]. The cardioprotective effects of SNO proteins are demonstrated in several reports [92]. In female hearts, it has been also demonstrated that enhanced S-nitrosogluthathione reductase (GSNOR) activity may play a role in the cardioprotective pathway by limiting protein SNO from accumulating to levels that are at risk to trigger deleterious nitrosative stress [90].

As we have already described in a previous section, estrogens are very important not only in modulating the ischemia/reperfusion damage, but also in protecting from cardiac injury from anthracyclines. Indeed, oxidative stress and apoptosis are shared mechanisms between the I/R and anthracyclines heart damage. Hence, anthracyclines are better tolerated in female adult cardiomyocytes [93] in experimental models [40]. Intriguingly, regardless of any therapy, cancer itself can induce cardiac atrophy and autophagy in a sexually dimorphic way, with estrogens conferring protection against cancer-induced cardiac atrophy and body weight loss by signaling through its receptor [94].

Other mechanisms of doxorubicin cardiotoxicity are related to mitochondrial dysfunction and down-regulation of energy metabolism signaling pathways [5,22]. In male rats, doxorubicin treatment resulted in important alterations in mitochondrial function, which appeared unaffected or remarkably

preserved in treated females [95]. Moreover, mitochondrial dysfunction and energy metabolism signaling pathways seem associated with early cardiotoxicity in males but not in females [95], with doxorubicin altering mitochondria more severely in males, as evidenced by a downregulation of gene expression of mitochondrial biogenesis, mitochondrial function and mitochondrial dynamic, decreased mitochondrial respiration, and mitochondrial DNA content. Also, a sex-specific impact of doxorubicin on the heart phospholipidome, especially on cardiolipin, an essential mitochondrial lipid, has also been shown [96].

We have shown recently that estrogens may influence the cardiotoxicity of antineoplastic drugs, with the activation of GPER mitigating the cardiotoxic effects of Doxorubicin (Dox), thus suggesting GPER as an interesting pharmacological target to limit the detrimental myocardial effects of Dox treatment [97]. Some animal studies show that females develop less cardiomyopathy and nephropathy than males after chronic administration of anthracyclines [98,99], but such protection is abrogated after ovariectomy [99]. On the other hand, 17- β -estradiol confers protection against cardiac injury in ovariectomized rats treated with Dox [100]. These observations seem to indicate a protective role of female hormones. Intriguingly, it has been also observed that testosterone is able to protect from ANT-induced damage in cardiac myocytes in vitro [101].

CONCLUSIONS

The studies we reviewed suggest there are sex differences in the triggering of specific cardioprotective signaling pathways in response to cardiac injury. Tonic and phasic estrogen actions on specific receptors may explain many of gender differences in I/R injury, in cardiotoxicity from anthracyclines, in post-ischemic systolic recovery and in conditioning protection. The intricate inter-relationship between estrogen receptors, NOS activity, and other related or not related mechanisms, acute and chronic conditions, and their roles in cardioprotection deserve further studies, with large clinical studies and more complex preclinical models that take into account sex differences, in order to get more insight into clinical applicability of novel approaches for diagnosis and protection from

anthracycline cardiotoxicity. Considering the impact that confounding factors including gender differences have on the triggering cardiac injury as well as on the effects of cardioprotective therapies, extrapolation from animal findings to clinical relevance in humans should be made cautiously [102], since estrogens may play a role of increased risk of malignancy, with the majority of post-menopausal women having ER + breast cancer.

FUNDING: Carlo Gabriele Tocchetti is funded by a Federico II University/Ricerca di Ateneo grant. Rudolf A de Boer is supported by the Netherlands Heart Foundation (CVON DOSIS, grant 2014-40, CVON SHE-PREDICTS-HF, grant 2017-21, and CVON RED-CVD, grant 2017-11); and the Netherlands Organization for Scientific Research (NWO VIDI, grant 917.13.350).

DISCLOSURES

Carlo Gabriele Tocchetti received speaking fees from Alere; Rudolf A de Boer: the UMCG, which employs RAdB has received research grants and/or fees from AstraZeneca, Abbott, Bristol-Myers Squibb, Novartis, Roche, Trevena, and ThermoFisher GmbH. RAdB is a minority shareholder of scPharmaceuticals, Inc. RAdB received personal fees from MandalMed Inc, AstraZeneca, Novartis, Servier, and Vifor. Alexander R Lyon has received speaker, advisory board or consultancy fees and/or research grants from Pfizer, Novartis, Servier, Amgen, Clinigen Group, Takeda, Roche, Eisai Ltd, Eli Lilly and Boehringer Ingelheim. Javid Moslehi: has served as a consultant/advisor for Novartis, Pfizer, Bristol-Myers Squibb, Takeda/Millennium, Ariad, Acceleron, Vertex, Incyte, Rgenix, Verastem, Pharmacyclics, StemCentRx, Heat Biologics, Daiichi-Sankyo, and Regeneron. All other authors have no conflicts of interest or financial ties to disclose.

FIGURE LEGENDS

Figure 1

Sexual dimorphism of anthracycline cardiotoxicity

Figure 2:

Bull's eye map of LV longitudinal strain showing early reduction of myocardial deformation at the basal LV segments (especially of the septum and anterior wall) in a woman treated with anticancer therapies.

Figure 3:

Estrogens and cardioprotection

REFERENCES

1. Zamorano JL, Lancellotti P, Rodriguez Munoz Detal (2016) 2016 ESC position paper on cancer treatments and cardiovascular toxicity developed under the auspices of the ESC Committee for Practice Guidelines: The Task Force for cancer treatments and cardiovascular toxicity of the European Society of Cardiology (ESC). *Eur Heart J* 37:2768–2801
2. Armenian SH, Lacchetti C, Barac A, Carver J, Constine LS, Denduluri N, Dent S, Douglas PS, Durand JB, Ewer M, Fabian C, Hudson M, Jessup M, Jones LW, Ky B, Mayer EL, Moslehi J, Oeffinger K, Ray K, Ruddy K, Lenihan D (2017) Prevention and monitoring of cardiac dysfunction in survivors of adult cancers: American Society of Clinical Oncology clinical practice guideline. *J Clin Oncol* 35:893–911
3. Denlinger CS, Sanft T, Baker KS, Broderick G, Demark Wahnefried W, Friedman DL, Goldman M, Hudson M, Khakpour N, King A, Koura D, Lally RM, Langbaum TS, McDonough AL, Melisko M, Montoya JG, Mooney K, Moslehi JJ, O'Connor T, Overholser L, Paskett ED, Peppercorn J, Pirl W, Rodriguez MA, Ruddy KJ, Silverman P, Smith S, Syrjala KL, van der Weide A, Urbaniak SG, Wakabayashi MT, Zee P, McMillian NR, Freedman-Cass DA (2018) Survivorship, Version 2.2018, NCCN clinical practice guidelines in oncology. *J Natl Compr Cancer Netw* 16:1216–1247.
4. Wang H-Y, Yin B-B, Jia D-Y, Hou Y-L (2017) Association between obesity and trastuzumab-related cardiac toxicity in elderly patients with breast cancer. *Oncotarget* 8(45):79289–79297.
5. Varricchi G, Ameri P, Cadeddu C, Ghigo A, Madonna R, Marone G, Mercurio V, Monte I, Novo G, Parrella P, Pirozzi F, Pecoraro A, Spallarossa P, Zito C, Mercurio G, Pagliaro P, Tocchetti CG (2018) Antineoplastic drug-induced cardiotoxicity: a redox perspective. *Front Physiol* 9:167.

6. Kremer LC, van Dalen EC, Offringa M, Voute PA (2002) Frequency and risk factors of anthracycline-induced clinical heart failure in children: a systematic review. *Ann Oncol* 13:503–512
7. Suter TM, Ewer MS (2013) Cancer drugs and the heart: importance and management. *Eur Heart J* 34:1102–1111
8. Swain SM, Whaley FS, Ewer MS (2003) Congestive heart failure in patients treated with doxorubicin: a retrospective analysis of 551three trials. *Cancer* 97:2869–2879
9. Ameri P, Canepa AMS et al (2018) Cancer diagnosis in patients 553with heart failure: epidemiology, clinical implications and gaps in knowledge. *Eur J Heart Fail* 20:879–887.
10. Mele D, Nardoza M, Spallarossa P, Frassoldati A, Tocchetti CG, Cadeddu C, Madonna R, Malagù M, Ferrari R, Mercuro G (2016) Current views on anthracycline cardiotoxicity. *Heart Fail Rev* 21: 621–634
11. Mehta LS, Watson KE, Barac A, Beckie TM, Bittner V, Cruz561 Flores S, Dent S, Kondapalli L, Ky B, Okwuosa T, Piña IL, Volgman AS, American Heart Association Cardiovascular Disease in Women and Special Populations Committee of the Council on Clinical Cardiology; Council on Cardiovascular and Stroke Nursing; and Council on Quality of Care and Outcomes Research (2018) Cardiovascular disease and breast cancer: where these entities intersect: a scientific statement from the American Heart Association. *Circulation* 137:e30–e66.
12. Kenigsberg B, Wellstein A, Barac A (2018) Left ventricular dysfunction in cancer treatment: is it relevant? *JACC Heart Fail* 6:87–95.
13. Chan DSM, Vieira AR, Aune D, Bandera EV, Greenwood DC, McTiernan A, Navarro Rosenblatt D, Thune I, Vieira R, Norat T (2014) Body mass index and survival in women with breast cancer-systematic literature review and meta-analysis of 82 follow-up studies. *Ann Oncol* 25:1901–1914.
14. Mitra MS, Donthamsetty S, White B, Mehendale HM (2008) High fat diet-fed obese rats are highly sensitive to doxorubicin-induced cardiotoxicity. *Toxicol Appl Pharmacol* 231:413–422

15. Seidman A, Hudis C, Pierri MK, Shak S, Paton V, Ashby M, Murphy M, Stewart SJ, Keefe D (2002) Cardiac dysfunction in the trastuzumab clinical trials experience. *J Clin Oncol* 20(5):1215–1221.
16. Ewer MS, Ewer SM (2010) Cardiotoxicity of anticancer treatments: what the cardiologist needs to know. *Nat Rev Cardiol* 7(10):564–575.
17. Suter TM, Procter M, van Veldhuisen DJ, Muscholl M, Bergh J, Carlomagno C, Perren T, Passalacqua R, Bighin C, Klijn JGM, Ageev FT, Hitre E, Groetz J, Iwata H, Knap M, Gnant M, Muehlbauer S, Spence A, Gelber RD, Piccart-Gebhart MJ (2007) Trastuzumab-associated cardiac adverse effects in the herceptin adjuvant trial. *J Clin Oncol* 25(25):3859–3865. <https://doi.org/10.1200/JCO.2006.09.1611>
18. Tocchetti CG, Cadeddu C, Di Lisi D, Femminò S, Madonna R, Mele D, Monte I, Novo G, Penna C, Pepe A, Spallarossa P, Varricchi G, Zito C, Pagliaro P, Mercurio G (2019 Jun 20) From molecular mechanisms to clinical management of antineoplastic drug-induced cardiovascular toxicity: a translational overview. *Antioxid Redox Signal* 30(18):2110–2153. <https://doi.org/10.1089/ars.2016.6930>
19. Spallarossa P, Maurea N, Cadeddu C et al (2016) A recommended practical approach to the management of anthracycline-based chemotherapy cardiotoxicity: an opinion paper of the working group on drug cardiotoxicity and cardioprotection, Italian Society of Cardiology. *J Cardiovasc Med (Hagerstown)* 17(Suppl 1):S84–608 S92
20. Avila MS, Ayub-Ferreira SM, de Barros Wanderley MR Jr, das Dores Cruz F, Gonçalves Brandão SM, VOC R, Higuchi-DosSantos MH, Hajjar LA, Kalil Filho R, Hoff PM, Sahade M, MSM F, de Paula Costa RL, Mano MS, Bittencourt Viana Cruz CB, Abduch MC, Lofrano Alves MS, Guimaraes GV, Issa VS, Bittencourt MS, Bocchi EA (2018) Carvedilol for prevention of chemotherapy-related cardiotoxicity: the CECCY trial. *J Am Coll Cardiol* 71(20):2281–2290.

21. Pituskin E, Mackey JR, Koshman S, Jassal D, Pitz M, Haykowsky MJ, Pagano JJ, Chow K, Thompson RB, Vos LJ, Ghosh S, Oudit GY, Ezekowitz JA, Paterson DI (2017 Mar 10) Multidisciplinary approach to novel therapies in cardio-oncology research (MANTICORE 101-breast): a randomized trial for the prevention of trastuzumab-associated cardiotoxicity. *J Clin Oncol* 35(8):870 – 877.
22. Štěrba M, Popelová O, Vávrová A, Jirkovský E, Kovaříková P, Geršl V, Šimůnek T (2013) Oxidative stress, redox signaling, and metal chelation in anthracycline cardiotoxicity and pharmacological cardioprotection. *Antioxid Redox Signal* 18:899–929. <https://doi.org/10.1089/ars.2012.4795>
23. Malorni W, Campesi I, Straface E, Vella S, Franconi F (2007) Redox features of the cell: a gender perspective. *Antioxid Redox Signal* 9:1779 –1801.
24. Vijay V, Han T, Moland CL, Kwekel JC, Fuscoe JC, Desai VG (2015) Sexual dimorphism in the expression of mitochondria related genes in rat heart at different ages. *PLoS One* 10:e0117047.
25. Jacquet JM, Bressolle F, Galtier M et al (1990) Doxorubicin and doxorubicinol: intra- and inter-individual variations of pharmacokinetic parameters. *Cancer Chemother Pharmacol* 27:219 –225
26. Dobbs NA, Twelves CJ, Gillies H, James CA, Harper PG, Rubens RD (1995) Gender affects the doxorubicin pharmacokinetics in 641 patients with normal liver bio chemistry. *Cancer Chemother Pharmacol* 36:473 –476.
27. Wade JR, Kelman AW, Kerr DJ, Robert J, Whiting B (1992) Variability in the pharmacokinetics of epirubicin: a population analysis. *Cancer Chemother Pharmacol* 29:391 –395
28. van Asperen J, van Tellingen O, Tijssen F, Schinkel AH, Beijnen JH (1999) Increased accumulation of doxorubicin and doxorubicinol in cardiac tissue of mice lacking mdrlaP-glycoprotein. *Br J Cancer* 79:108 –113.

29. Lipshultz SE, Lipsitz SR, Mone SM, Goorin AM, Sallan SE, Sanders SP, Orav EJ, Gelber RD, Colan SD (1995) Female sex and drug dose as risk factors for late cardiotoxic effects of doxorubicin therapy for childhood cancer. *N Engl J Med* 332:1738 – 1743.
30. Lipshultz SE, Scully RE, Lipsitz SR, Sallan SE, Silverman LB, Miller TL, Barry EV, Asselin BL, Athale U, Clavell LA, Larsen E, Moghrabi A, Samson Y, Michon B, Schorin MA, Cohen HJ, Neuberg DS, Orav EJ, Colan SD (2010) Assessment of dexrazoxane as a cardioprotectant in doxorubicin-treated children with high-risk acute lymphoblastic leukaemia: long-term follow up of a prospective, randomised, multicentre trial. *Lancet Oncol* 11:950 –961.
31. Krischer JP, Epstein S, Cuthbertson DD, Goorin AM, Epstein ML, Lipshultz SE (1997) Clinical cardiotoxicity following anthracycline treatment for childhood cancer: the pediatric oncology group experience. *J Clin Oncol* 15:1544 –1552.
32. Hequet O, Le QH, Moullet I et al (2004) Subclinical late cardiomyopathy after doxorubicin therapy for lymphoma in adults. *J Clin Oncol* 22:1864 –1871.
33. Myrehaug S, Pintilie M, Tsang R, Mackenzie R, Crump M, Chen Z, Sun A, Hodgson DC (2008) Cardiac morbidity following modern treatment for Hodgkin lymphoma: supra-additive cardiotoxicity of doxorubicin and radiation therapy. *Leuk Lymphoma* 49:1486 –1493.
34. Myrehaug S, Pintilie M, Yun L, Crump M, Tsang RW, Meyer RM, Sussman J, Yu E, Hodgson DC (2010) A population-based study of cardiac morbidity among Hodgkin lymphoma patients with preexisting heart disease. *Blood* 116:2237 –2240
35. Caram MEV, Guo C, Leja M, Smerage J, Henry NL, Giacherio D, Rubenfire M, Schott A, Davis M, Hayes DF, van Poznak C, Cooney KA, Hertz DL, Banerjee M, Griggs JJ (2015) Doxorubicin-induced cardiac dysfunction in unselected patients with a history of early-stage breast cancer. *Breast Cancer Res Treat* 152:163 –172
36. Schmidt KT, Andersen CY (2012) ISFP Practice Committee. Recommendations for fertility preservation in patients with lymphomas. *J Assist Reprod Genet* 29:473–477.

37. Hershman DL, McBride RB, Eisenberger A, Tsai WY, Grann VR, Jacobson JS (2008) Doxorubicin, cardiac risk factors, and cardiac toxicity in elderly patients with diffuse B-cell non-Hodgkin's lymphoma. *J Clin Oncol* 26(19):3159–3165
38. Pupo M, Pisano A, Abonante S, Maggiolini M, Musti AM (2014) GPER activates Notch signaling in breast cancer cells and cancer associated fibroblasts (CAFs). *Int J Biochem Cell Biol* 46:56–67
39. Lubecka K, Kurzava L, Flower K, Buvala H, Zhang H, Teegarden D, Camarillo I, Suderman M, Kuang S, Andrisani O, Flanagan JM, Stefanska B (2016) Stil benoids remodel the DNA methylation patterns in breast cancer cells and inhibit oncogenic NOTCH signaling through epigenetic regulation of MAML2 transcriptional activity. *Carcinogenesis*. 37(7):656–668.
40. Gonzalez Y, Pokrzywinski KL, Rosen ET, Mog S, Aryal B, Chehab LM, Vijay V, Moland CL, Desai VG, Dickey JS, Rao VA (2015) Reproductive hormone levels and differential 707 mitochondria-related oxidative gene expression as potential mechanisms for gender differences in cardiosensitivity to doxorubicin in tumor-bearing spontaneously hypertensive rats. *Cancer Chemother Pharmacol* 76:447–459
41. Cadeddu Dessalvi C, Deidda M, Mele D et al (2018) Chemotherapy-induced cardiotoxicity: new insights into mechanisms, monitoring, and prevention. *J Cardiovasc Med (Hagerstown)* 19:315–323.
42. Masoudi FA, Havranek EP, Smith G, Fish RH, Steiner JF, Ordin DL, Krumholz HM (2003) Gender, age, and heart failure with preserved left ventricular systolic function. *J Am Coll Cardiol* 41(2):217–223
43. Gori M, Lam CS, Gupta DK, Santos AB, Cheng S, Shah AM, Claggett B, Zile MR, Kraigher-Krainer E, Pieske B, Voors AA, Packer M, Bransford T, Lefkowitz M, McMurray JJ (2014) Solomon SD; PARAMOUNT Investigators. Sex-specific cardiovascular structure and function in heart failure with preserved ejection fraction. *Eur J Heart Fail* 16(5):535–542

44. Plana JC, Galderisi M, Barac A, Ewer MS, Ky B, Scherrer Crosbie M, Ganame J, Sebag IA, Agler DA, Badano LP, Banchs J, Cardinale D, Carver J, Cerqueira M, DeCara JM, Edvardsen T, Flamm SD, Force T, Griffin BP, Jerusalem G, Liu JE, Magalhães A, MarwickT, Sanchez LY, Sicari R, VillarragaHR, Lancellotti P (2014) Expert consensus for multimodality imaging evaluation of adult patients during and after cancer therapy: a report from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Soc Echocardiogr* 27:911–939
45. Lang RM, Badano LP, Mor-Avi Vet al (2015) Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Soc Echocardiogr* 28(1–39):e14
46. Gerds E, Okin PM, de Simone G, Cramariuc D, Wachtell K, Boman K, Devereux RB (2008) Gender differences in left ventricular structure and function during antihypertensive treatment: the Losartan Intervention for Endpoint Reduction in Hypertension Study. *Hypertension*. 51(4):1109–1114
47. Stokke TM, Hasselberg NE, Smedsrud MK, Sarvari SI, Haugaa KH, Smiseth OA, Edvardsen T, Remme EW (2017) Geometry as a confounder when assessing ventricular systolic function: comparison between ejection fraction and strain. *J Am Coll Cardiol* 70(8):942–954
48. Civelli M, Cardinale D, Martinoni A, Lamantia G, Colombo N, Colombo A, Gandini S, Martinelli G, Fiorentini C, Cipolla CM (2006) Early reduction in left ventricular contractile reserve detected by dobutamine stress echo predicts high-dose chemotherapy-induced cardiac toxicity. *Int J Cardiol* 111:120 –126
49. Bountiukos M, Doorduijn JK, Roelandt JR, Vourvouri EC, Bax JJ, Schinkel AF, Kertai MD, Sonneveld P, Poldermans D (2003) Repetitive dobutamine stress echocardiography for the prediction of anthracycline cardiotoxicity. *Eur J Echocardiogr* 4(4):300 –305

50. Isner JM, Ferrans VJ, Cohen SR, Witkind BG, Virmani R, Gottdiener JS, Beck JR, Roberts WC (1983) Clinical and morphologic cardiac findings after anthracycline chemotherapy. Analysis of 64 patients at necropsy. *Am J Cardiol* 51:1167 –1174
51. Brouwer CA, Postma A, Vonk JM, Zwart N, van den Berg MP, Bink-Boelkens MT, Dolsma WV, Smit AJ, de Vries EG, Tissing WJ, Gietema JA (2011) Systolic and diastolic dysfunction in longterm adult survivors of childhood cancer. *Eur J Cancer* 47(16): 2453 –2462
52. Okuma H, Noto N, Tanikawa S, Kanezawa K, Hirai M, Shimozawa K, Yagasaki H, Shichino H, Takahashi S (2017) Impact of persistent left ventricular regional wall motion abnormalities in childhood cancer survivors after anthracycline therapy: assessment of global left ventricular myocardial performance by 3D speckle-tracking echocardiography. *J Cardiol* 70(4):396 –401
53. Zito C, Longobardo L, Citro R, Galderisi M, Oreto L, Carerj ML, Manganaro R, Cusmà-Piccione M, Todaro MC, Di Bella G, Imbalzano E, Khandheria BK, Carerj S (2018) Ten years of 2D longitudinal strain for early myocardial dysfunction detection: a clinical overview. *Biomed Res Int* 2018:8979407
54. Sugimoto T, Dulgheru R, Bernard A, Ilardi F, Contu L, Addetia K, Caballero L, Akhaladze N, Athanassopoulos GD, Barone D, Baroni M, Cardim N, Hagendorff A, Hristova K, Lopez T, de la Morena G, Popescu BA, Moonen M, Penicka M, Ozyigit T, Rodrigo Carbonero JD, van de Veire N, von Bardeleben RS, Vinereanu D, Zamorano JL, Go YY, Rosca M, Calin A, Magne J, Cosyns B, Marchetta S, Donal E, Habib G, Galderisi M, Badano LP, Lang RM, Lancellotti P (2017) Echocardiographic reference ranges for normal left ventricular 2D strain: results from the EACVI NORRE study. *Eur Heart J Cardiovasc Imaging* 18(8): 833 –840
55. Kocabay G, Muraru D, Peluso D, Cucchini U, Mihaila S, Padayattil-Jose S, Gentian D, Iliceto S, Vinereanu D, Badano LP (2014) Normal left ventricular mechanics by two-dimensional speckle-tracking echocardiography. Reference values in healthy adults. *Rev Esp Cardiol (Engl Ed)* 67:651 –658

56. Takigiku K, Takeuchi M, Izumi C, Yuda S, Sakata K, Ohte N, Tanabe K, Nakatani S, on behalf of the JUSTICE investigators (2012) Normal range of left ventricular 2-dimensional strain: Japanese Ultrasound Speckle Tracking of the Left Ventricle (JUSTICE) study. *Circ J* 76:2623 –2632
57. Barros MVL, Macedo AVS, Sarvari SI, Faleiros MH, Felipe PT, Silva JLP, Edvardsen T (2019) Left ventricular regional wall motion abnormality is a strong predictor of cardiotoxicity in breast cancer patients undergoing chemotherapy. *Arq Bras Cardiol* 112(1):50 –56
58. Zito C, Longobardo L, Cadeddu C, Monte I, Novo G, Dell'Oglio S, Pepe A, Madonna R, Tocchetti CG, Mele D (2016) Cardiovascular imaging in the diagnosis and monitoring of cardiotoxicity: role of echocardiography. *J Cardiovasc Med (Hagerstown)* 17(Suppl 1 Special issue on Cardiotoxicity from Antiblastic Drugs and Cardioprotection):e35 –e44
59. Aquaro GD, Camastra G, Monti L et al (2016) Reference values of cardiac volumes, dimensions, and new functional parameters by MR: a multicentre, multivendor study. *J Magn Reson Imaging* 45: 1055 –1067
60. Jordan JH, D'Agostino RB Jr, Hamilton CA et al (2014) Longitudinal assessment of concurrent changes in left ventricular ejection fraction and left ventricular myocardial tissue characteristics after administration of cardiotoxic chemotherapies using T1-weighted and T2-weighted cardiovascular magnetic resonance. *Circ Cardiovasc Imaging* 7:872–879
61. Ylanen K, Poutanen T, Savikurki-Heikkila P, Rinta-Kiikka I, Eerola A, Vettenranta K (2013) Cardiac magnetic resonance imaging in the evaluation of the late effects of anthracyclines among long-term survivors of childhood cancer. *J Am Coll Cardiol* 61: 1539–1547
62. Toro-Salazar OH, Gillan E, O'Loughlin MT et al (2013) Occult cardiotoxicity in childhood cancer survivors exposed to anthracycline therapy. *Circ Cardiovasc Imaging* 6:873–880

63. Mitoff PR, Gam D, Ivanov J, Al-hesayen A, Azevedo ER, Newton GE, Parker JD, Mak S (2011) Cardiac-specific sympathetic activation in men and women with and without heart failure. *Heart*. 97(5):382–387.
64. Mitoff PR, Al-Hesayen A, Azevedo E, Newton GE, Mak S (2007) Sex differences in basal hemodynamics and left ventricular function in humans with and without heart failure. *Am Heart J* 154(3): 575–580
65. Armstrong GT, Plana JC, Zhang N, Srivastava D, Green DM, Ness KK, Daniel Donovan F, Metzger ML, Arevalo A, Durand JB, Joshi V, Hudson MM, Robison LL, Flamm SD (2012) Screening adult survivors of childhood cancer for cardiomyopathy: comparison of echocardiography and cardiac magnetic resonance imaging. *J Clin Oncol* 30:2876–2884
66. Tham EB, Haykowsky MJ, Chow K, Spavor M, Kaneko S, Khoo NS, Pagano JJ, Mackie AS, Thompson RB (2013) Diffuse myocardial fibrosis by T1-mapping in children with subclinical anthracycline cardiotoxicity: relationship to exercise capacity, cumulative dose and remodeling. *J Cardiovasc Magn Reson* 15:48
67. Yu AF, Ky B (2016) Roadmap for biomarkers of cancer therapy cardiotoxicity. *Heart*. 102:425–430
68. Daniels LB, Maisel AS (2015) Cardiovascular biomarkers and sex: the case for women. *Nat Rev Cardiol* 12:588–596
69. Suthahar N, Meijers WC, Ho JE, Gansevoort RT, Voors AA, van der Meer P, Bakker SJL, Heymans S, van Empel V, Schroen B, van der Harst P, van Veldhuisen DJ, de Boer RA (2018) Sex specific associations of obesity and N-terminal pro-B-type natriuretic peptide levels in the general population. *Eur J Heart Fail* 20:1205–1214.
70. Cardinale D, Sandri MT, Colombo A, Colombo N, Boeri M, Lamantia G, Civelli M, Peccatori F, Martinelli G, Fiorentini C, Cipolla CM (2004) Prognostic value of troponin I in cardiac risk stratification of cancer patients undergoing high-dose chemotherapy. *Circulation* 109:2749–2754

71. Auner HW, Tinchon C, Linkesch W, Tiran A, Quehenberger F, Link H, Sill H (2003) Prolonged monitoring of troponin T for the detection of anthracycline cardiotoxicity in adults with hematological malignancies. *Ann Hematol* 82:218–222
72. Levis BE, Binkley PF, Shapiro CL (2017) Cardiotoxic effects of anthracycline-based therapy: what is the evidence and what are the potential harms? *Lancet Oncol* 18:e445–e456
73. Feola M, Garrone O, Occelli M, Francini A, Biggi A, Visconti G, Albrile F, Bobbio M, Merlano M (2011) Cardiotoxicity after anthracycline chemotherapy in breast carcinoma: effects on left ventricular ejection fraction, troponin I and brain natriuretic peptide. *Int J Cardiol* 148:194–198
74. Pistillucci G, Ciorra AA, Sciacca V, Raponi M, Rossi R, Veltri E (2015) Troponin I and B-type natriuretic peptide (BNP) as biomarkers for the prediction of cardiotoxicity in patients with breast cancer treated with adjuvant anthracyclines and trastuzumab. *Clin Ther* 166:e67–e71
75. Ky B, Putt M, Sawaya H, French B, Januzzi JL Jr, Sebag IA, Plana JC, Cohen V, Banchs J, Carver JR, Wiegers SE, Martin RP, Picard MH, Gerszten RE, Halpern EF, Passeri J, Kuter I, Scherrer Crosbie M (2014) Early increases in multiple biomarkers predict subsequent cardiotoxicity in patients with breast cancer treated with doxorubicin, taxanes, and trastuzumab. *J Am Coll Cardiol* 63:809–816
76. Lecour S, Bøtker HE, Condorelli G et al (2014) ESC working group cellular biology of the heart: position paper: improving the preclinical assessment of novel cardioprotective therapies. *Cardiovasc Res* 104:399–411
77. Hausenloy DJ, Lecour S, Yellon DM (2011) Reperfusion injury salvage kinase and survivor activating factor enhancement prosurvival signaling pathways in ischemic postconditioning: two sides of the same coin. *Antioxid Redox Signal* 14:893–907.

78. Hausenloy DJ, Botker HE, Condorelli G et al (2013) Translating cardioprotection for patient benefit: position paper from the Working Group of Cellular Biology of the heart of the European Society of Cardiology. *Cardiovasc Res* 98:7 –27.
79. Madonna R, Cadeddu C, Deidda M, Giricz Z, Madeddu C, Mele D, Monte I, Novo G, Pagliaro P, Pepe A, Spallarossa P, Tocchetti CG, Varga ZV, Zito C, Geng YJ, Mercurio G, Ferdinandy P (2015) Cardioprotection by gene therapy: a review paper on behalf of the Working Group on Drug Cardiotoxicity and Cardioprotection of the Italian Society of Cardiology. *Int J Cardiol* 191:203 –210.
80. Deschamps AM, Murphy E, Sun J (2010) Estrogen receptor activation and cardioprotection in ischemia reperfusion injury. *Trends Cardiovasc Med* 20(3):73 –78.
81. Fels JA, Manfredi G (2019) Sex differences in ischemia/reperfusion injury: the role of mitochondrial permeability transition. *Neurochem Res*. <https://doi.org/10.1007/s11064-019-02769-6>
82. Murphy E, Steenbergen C (2007) Gender-based differences in mechanisms of protection in myocardial ischemia-reperfusion injury. *Cardiovasc Res* 75(3):478 –486
83. Ostadal B, Drahota Z, Houstek J, Milerova M, Ostadalova I, Hlavackova M, Kolar F (2019) Developmental and sex differences in cardiac tolerance to ischemia/reperfusion injury: the role of mitochondria. *Can J Physiol Pharmacol*:1 –7.
84. Ostadal B, Ostadal P (2014) Sex-based differences in cardiac ischaemic injury and protection: therapeutic implications. *Br J Pharmacol* 171(3):541 –554.
85. Penna C, Tullio F, Merlino A, Moro F, Raimondo S, Rastaldo R, Perrelli MG, Mancardi D, Pagliaro P (2009) Postconditioning cardioprotection against infarct size and post-ischemic systolic dysfunction is influenced by gender. *Basic Res Cardiol* 104(4): 390 –402.
86. Rocca C, Femminò S, Aquila G, Granieri MC, DeFrancesco EM, Pasqua T, Rigracciolo DC, Fortini F, Cerra MC, Maggiolini M, Pagliaro P, Rizzo P, Angelone T, Penna C (2018) Notch1

- mediates preconditioning protection induced by GPER in normotensive and hypertensive female rat hearts. *Front Physiol* 9:521.
87. Mendelsohn ME, Karas RH (1999) The protective effects of estrogen on the cardiovascular system. *N Engl J Med* 340:1801 –1811
88. Murphy E, Kohr M, Sun J, Nguyen T, Steenbergen C (2012) S-nitrosylation: a radical way to protect the heart. *J Mol Cell Cardiol* 52:568 –577.
89. Bienvenu LA, Morgan J, Reichelt ME, Delbridge LMD, Young MJ (2017) Chronic in vivo nitric oxide deficiency impairs cardiac functional recovery after ischemia in female (but not male) mice. *J Mol Cell Cardiol* 112:8 –15.
90. Shao Q, Fallica J, Casin KM, Murphy E, Steenbergen C, Kohr MJ (2016) Characterization of the sex-dependent myocardial S- nitrosothiol proteome. *Am J Physiol Heart Circ Physiol* 310: H505–H515.
91. Tong G, Aponte AM, Kohr MJ, Steenbergen C, Murphy E, Sun J (2014) Postconditioning leads to an increase in protein S-nitrosylation. *Am J Physiol Heart Circ Physiol* 306:H825–H832
92. Penna C, Angotti C, Pagliaro P (2014) Protein S-nitrosylation in preconditioning and postconditioning. *ExpBiol Med* (Maywood) 239:647–662
93. Wang F, He Q, Sun Y, Dai X, Yang XP (2010) Female adult mouse cardiomyocytes are protected against oxidative stress. *Hypertension* 55:1172–1178
94. Cospers and Leinwand (2011) Cancer causes cardiac atrophy and autophagy in a sexually dimorphic manner. *Cancer Res* 71:1710– 720
95. Moulin M, Piquereau J, Mateo P, Fortin D, Rucker-Martin C, Gressette M, Lefebvre F, Gresikova M, Solgadi A, Veksler V, Garnier A, Ventura-Clapier R (2015) Sexual dimorphism of doxorubicin-mediated cardiotoxicity: potential role of energy metabolism remodeling. *Circ Heart Fail* 8:98–108

96. Moulin M, Solgadi A, Veksler V, Garnier A, Ventura-Clapier R, Chaminade P (2015) Sex-specific cardiac cardiolipin remodelling after doxorubicin treatment. *Biol Sex Differ* 6:20
97. De Francesco EM, Rocca C, Scavello F et al (2017) Protective role of GPER agonist G-1 on cardiotoxicity induced by doxorubicin *J Cell Physiol* 232:1640–1649.
98. Julicher RH, Sterrenberg L, Haenen GR, Bast A, Noordhoek J (1988) The effect of chronic Adriamycin treatment on heart kidney and liver tissue of male and female rat. *Arch Toxicol* 61:275 –281
99. Zhang J, Knapton A, Lipshultz SE, Cochran TR, Hilaragi H, Herman EH (2014) Sex-related differences in mast cell activity and doxorubicin toxicity: a study in spontaneously hypertensive rats. *Toxicol Pathol* 42:361 –375
100. Munoz-Castaneda JR, Montilla P, Munoz MC, Bujalance I, Muntane J, Tunez I (2005) Effect of 17-beta-estradiol administration during adriamycin-induced cardiomyopathy in ovariectomized rat. *Eur J Pharmacol* 523:86 –92
101. Altieri P, Barisione C, Lazzarini E et al (2016) Testosterone antagonizes doxorubicin-induced senescence of cardiomyocytes. *J Am Heart Assoc* 5(1):pii: e002383
102. Madonna R, Cadeddu C, Deidda M, Mele D, Monte I, Novo G, Pagliaro P, Pepe A, Spallarossa P, Tocchetti CG, Zito C, Mercurio G (2015) Improving the preclinical models for the study of chemotherapy-induced cardiotoxicity: a position paper of the Italian Working Group on Drug Cardiotoxicity and Cardioprotection. *Heart Fail Rev* 20:621 –631.

Figure 1

Sexual dimorphism of anthracycline cardiotoxicity

Figure 2.

Figure 3