UNIVERSIDADE DE LISBOA

FACULDADE DE MEDICINA LISBOA

TRABALHO FINAL
MESTRADO INTEGRADO EM MEDICINA

Instituto de Fisiologia

CHEMOTHERAPY-INDUCED CARDIOTOXICITY IN BREAST CANCER

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MAIO'2022

Resumo

A neoplasia da mama é o segundo cancro mais comum em mulheres com cerca de 2.3 milhões de novos casos em 2020. A melhoria da taxa de sobrevivência nas últimas duas décadas devido a progressos no tratamento acompanhou-se por aumento dos efeitos adversos, particularmente cardíacos, ameaçando comprometer a eficácia das terapias disponíveis. A cardiotoxicidade é um dos efeitos adversos mais associados aos agentes antineoplásicos das terapias da neoplasia mamária. A sua prevenção, monitorização, diagnóstico e tratamento são cruciais para melhorar o prognóstico.

Esta tese procura efetuar uma revisão da literatura existente acerca da toxicidade cardiovascular associada à quimioterapia do cancro da mama, com enfoque nos efeitos adversos mais comuns. Os agentes citotóxicos mais frequentemente usados, seus potenciais mecanismos cardiotóxicos e os métodos de avaliação e monitorização da função cardíaca serão revistos. Destacaremos, também, potenciais estratégias preventivas e/ou de mitigação de doença cardiovascular nestas pacientes e discutiremos perspetivas futuras em Cardio-Oncologia.

Em resumo, a abordagem da cardiotoxicidade permanece indefinida. Apesar da monitorização da FEVE, são necessárias diretrizes baseadas em evidências para otimização da prevenção e diagnóstico, melhorando a gestão da cardiotoxicidade pré-clínica e clínica, desenvolvimento de biomarcadores que permitam melhor estratificação de risco em estadio pré-clínico da cardiotoxicidade e uma definição universalmente aceite. Identificámos limitações e lacunas no conhecimento atual nomeadamente na aplicação dos biomarcadores, na monitorização da saúde cardiovascular durante a terapêutica do cancro, papel da profilaxia com fármacos cardioprotetores e na deteção de danos cardiovasculares latentes nos sobreviventes.

Assim, a investigação deve procurar definir e uniformizar estratégias de abordagem, identificar biomarcadores mais precoces e sensíveis e esclarecer os mecanismos de cardiotoxicidade envolvidos. É igualmente importante a procura de evidência robusta na área da Cardio-Oncologia para apoio de decisões informadas acerca da terapia e diagnóstico precoce da doença cardiovascular.

Palavras-Chave: “Cancro da mama”, “Cardiotoxicidade”, “Quimioterapia”, “Cardio-oncologia”, “Insuficiência Cardíaca”.

Summary

Breast cancer is the second most common cancer in women with an estimated 2.3 million new cases in 2020. Improvements in cancer survival rates in the last two decades due to treatment progress were accompanied by an increase of mainly cardiac adverse side effects, threatening to compromise the effectiveness of available therapies. Cardiotoxicity is one of the most significant adverse effects associated with anti-cancer agents used in breast cancer therapies. Its prevention, monitoring, diagnosis and treatment are crucial to improve prognosis.

This dissertation aims to review the existing literature on cardiovascular toxicity associated with chemotherapy in breast cancer patients, addressing its most frequent adverse effects. The most frequently used cytotoxic agents, their potential cardiotoxic mechanisms and the methods for assessing and monitoring cardiac function will be reviewed. We will also highlight potential preventive and/or mitigation strategies of cardiovascular disease in these patients and future perspectives in Cardio-Oncology will be discussed.

In summary, the assessment of cardiotoxicity remains indefinite. Despite LVEF monitoring, evidence-based guidelines are needed to optimize prevention and diagnosis improving preclinical and clinical cardiotoxicity management, the development of biomarkers that allow for better risk stratification in preclinical stages of cardiotoxicity and its universally accepted definition. We identified limitations and gaps on current knowledge concerning the applicability of biomarkers, on cardiovascular health monitoring during cancer therapy, the prophylactic role of cardioprotective drugs and in detecting latent cardiovascular damage in cancer survivors.

Thus, investigation should seek to define and standardize strategies for assessing, earlier identification and more sensitive biomarkers and enlighten the mechanisms involved in cardiotoxicity. Equally important is the search for robust data in the field of Cardio-Oncology to support informed decisions concerning therapy and early diagnosis of cardiovascular disease.

Keywords: “Breast Cancer”, “Cardiotoxicity”, “Chemotherapy”, “Cardio-oncology”, “Heart failure”.

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Acknowledgments

Throughout the writing of the present work, I have received a great deal of support and, for that, I am grateful to all involved.

I would firstly like to thank my mother for being the person I admire the most in the world as well as an amazing role-model always inspiring me to be a better person each passing day and the one from whom I inherited the will to help others and pursue a carrier in healthcare. Thank you for teaching me to always fight for what I believe and never give up on my dreams.

I would also like to thank to my father and brother for their endless patience and for always having a friendly word to cheer me up when I needed.

To my friends, thank you for your always great advice, motivation but also for those sporadic breaks that provided fun and rest between chapters.

Finally, I would like to express my deepest gratitude to my supervisor, Dr ª Vera Geraldes, for all her support and advice throughout the writing of this work.

List of Abbreviations

2D – 2 Dimensional

3D – 3 Dimensional

5-FU – 5-Fluorouracil

ACC – American College of Cardiology

ACEi – Angiotensin-converting enzyme inhibitors

AHA – American Heart Association

ANT – Anthracycline

ARBs – Angiotensin Receptor Blockers

ASCO – American Society of Clinical Oncology

ASE/EACVI – American Society of Echocardiography / European Association of Cardiovascular Imaging

ATP – Adenosine triphosphate

BNP – B-type Natriuretic Peptide

CAD – Coronary Artery Disease

CV – Cardiovascular

CMRI – Cardiac Magnetic Resonance Imaging

DNA – Deoxyribonucleic Acid

DPD – Dihydropyrimidine Dehydrogenase

ECG – Electrocardiogram

EMA – European Medicines Agency

ESC – European Society of Cardiology

ESMO – European Society of Medical Oncology

FDA – United States Food and Drug Administration

GLP-1 – Glucagon-like peptide-1

GLS – Global Longitudinal Myocardial Strain

HER-2 – Epidermal growth factor receptor 2

Hs-cTn – High-sensitivity cardiac troponin

LV – Left Ventricular

LVEF – Left Ventricular Ejection Fraction

LVSD – Left Ventricular Systolic Dysfunction

MUGA – Multigated Acquisition Scan

NRG – Neuregulin

NT-pro-BNP – N-Terminal-pro-B-type Natriuretic Peptide

PARP-1 – Poly (ADP) - Ribose Polymerase 1

PET – Positron Emission Tomography

PK-C – Protein Kinase C

RNA – Ribonucleic Acid

RNS – Reactive Nitrogen Species

ROS – Reactive Oxygen Species

SPECT – Single-Photon Emission Computerized Tomography

Top2 – Topoisomerase II

TRA – Trastuzumab

List of Tables

1. Introduction

Cancer patients' management and outcome have been revolutionizing throughout the last decades, either due to the development of new and more efficient therapies, improvements in diagnosis capacity as well as increasing investment in prevention. These advances are particularly evident in breast cancer therapy and all combined have led to the reduction in cancer-related mortality. Thus, today's patients diagnosed with breast cancer have more therapy options available and consequently, more probability of cure and better outcomes (Kubota et al., 2021).

However, many of the chemotherapeutic agents used in breast cancer therapies are associated with an increased risk of cardiovascular toxicity presenting as long-term complications appearing during or after therapy either through the form of asymptomatic or symptomatic ventricular dysfunction eventually leading to heart failure, as well as other complications, such as arrhythmias or myocardial ischemia (Lakhani et al., 2021). Due to the increase in life expectancy, chemotherapy-induced cardiotoxicity is becoming more prevalent in these patients and is associated with great comorbidity, representing the primary cause of death in breast cancer survivors. According to a study by Cho et al., in 2020, the cumulative incidence of cardiotoxicity at 2 years following therapy for breast cancer in patients who received doxorubicin was 6.1% and 20.2%, if combined therapy with trastuzumab was administered (Cho et al., 2020). Hence, the adverse cardiac effects related to cancer therapies are negatively affecting prognosis by limiting its clinical use and thus reducing the efficacy of these therapies in treating the tumor, while inducing premature morbidity and death among survivors (Zamorano et al., 2016).

Given this rising concern, there is an urgent need to address and periodically monitor these potential cardiac adverse effects of chemotherapy without interfering with the efficacy of cancer treatment itself. Some of the monitoring options available to detect cardiotoxicity, enabling calculation of left ventricular ejection fraction and estimation of myocardial deformation, include cardiac imaging techniques as well as biomarkers assessment to identify myocardial damage (Kostakou et al., 2019; Zamorano et al., 2016). However, the left ventricular ejection fraction, currently used to identify patients at risk for cardiotoxicity, is subject to considerable intra-observer and inter-observer variability, as well as discrepancy across imaging modalities, lacking sensitivity

to detect early subclinical changes or to predict subsequent declines in function along with treatment (Thavendiranathan et al., 2013).

There is also a need to develop efforts and design studies to better understand the mechanisms responsible for this cardiac toxicity, to early identify subclinical cardiac dysfunction, to establish cardioprotective and risk stratification strategies to prevent the interruption or discontinuation of cancer therapy, and to reduce earlier and late cardiovascular and oncological morbidity and mortality (Chung et al., 2018).

It is important that the risks associated with chemotherapy are weighed against the benefits so that one fatal disease is not replaced by another. Therefore, it makes sense to adopt a multidisciplinary approach to these patients, involving cardiologists, oncologists and radiotherapists in a cardio-oncology program. The goal should be that a breast cancer survivor of today does not become the heart failure patient of tomorrow (Chung et al., 2018).

2. Aim

In this dissertation, we aim to review the existing literature on chemotherapy-induced cardiotoxicity, with a particular focus on breast cancer chemotherapies and their potential cardiac toxicities.

Furthermore, it is our purpose to discuss the mechanisms potentially involved in chemotherapy-induced cardiotoxicity, as well as to review the methods for assessing and monitoring cardiotoxicity and the potential strategies to prevent and/or attenuate chemotherapy-related cardiotoxicity. Future perspectives in the field of cardio-oncology will also be discussed.

3. Search strategy and selection criteria

Data was collected through searches using databases such as PubMed and Cochrane Library from November 15, 2020 until October 24, 2021.

Some of the subject headings used are “Cardiotoxicity”, “Breast Cancer” and “Antineoplastic therapies” combined with various keywords.

Searches in databases were limited to reviews, systematic reviews, guidelines, clinical trials, meta-analyses and randomized control trials published in English.

The bibliographic references list of the articles found also provided additional and relevant publications. The “related articles” feature of PubMed was also used to identify other citations in the literature.

4. Definition of cardiotoxicity

Chemotherapy-induced cardiotoxicity is an important and serious complication of chemotherapeutic agents limiting its therapeutic use and simultaneously leading to increased morbidity and mortality (Angsutararux et al., 2015; Shaikh & Shih, 2012).

Throughout many years, there have been several alternative definitions regarding cardiotoxicity as well as changes in the concept over time, as shown in Table 1.

Table 1- Evolution of the definition of Cardiotoxicity over time. Adapted from (Alexandre et al., 2020; Angsutararux et al., 2015; D. M. Cardinale et al., 2020; Chung et al., 2018; Lambert & Thavendiranathan, 2016; López-Sendón et al., 2020; Pardo Sanz & Zamorano, 2020; Shaikh & Shih, 2012; Zamorano et al., 2016).

Year	Definition of Cardiotoxicity
1946	"…cardiac toxicity regarding cardiac function or tissue induced by **local anesthetics, mercurial diuretics and digitalis**…"
1970	"…cardiac complications related to **cancer treatments**, particularly anthracyclines, 5-fluorouracil and combination with radiotherapy…"
2012	"…also **indirect effects that chemotherapy has on the cardiovascular system**, such as thrombotic status or hemodynamic impairment…"
2016	"…**direct cardiac toxic effects of breast cancer treatments,** affecting either its function and\or structure… it can also **accelerate the development or progression of pre-existing CV diseases**…"
2020	"…**new onset or worsening in ventricular function or myocardial damage**, from baseline after initiation of chemotherapy…" presenting by "serial absolute decrease or decline below a certain level in LVEF" (see table 3) or "GLS drop from baseline" (see table 3) and/or "rise in cardiac troponin levels from baseline exceeding the upper limit of normal"

This term was first used in 1946 to describe the cardiac toxicity regarding cardiac function or tissue induced by local anesthetics, mercurial diuretics and digitalis. Later, in the 1970s, cardiotoxicity began to be applied to cancer chemotherapy describing the cardiac complications related to these treatments, particularly those using certain classes like anthracyclines, combination with radiotherapy, and 5-fluorouracil which are

more frequently associated with cardiac adverse effects (Angsutararux et al., 2015; D. M. Cardinale et al., 2020; Chung et al., 2018).

One may try to define cardiotoxicity related to breast cancer as the direct toxic effects that breast cancer treatments have on the heart, affecting either its function and/or structure. Furthermore, given the case of pre-existing cardiovascular risk factors, it can also accelerate the development or progression of cardiovascular diseases (Zamorano et al., 2016). Apart from these direct effects, some consider that cardiotoxicity also encompasses the indirect effects that chemotherapy has on the cardiovascular system, being often associated with thrombotic status or hemodynamic impairment (Shaikh & Shih, 2012).

Alternatively, Chung *et al*. suggested the classification of cardiovascular side effects according to its time of onset as either "acute", "early-chronic" or "late-chronic" as well as whether the population involved is adult or pediatric and the chemotherapeutic class responsible (Chung et al., 2018; Shaikh & Shih, 2012).

Zamorano *et al.* proposed a new approach to cardiovascular toxicity related to chemotherapy by categorizing side effects into 9 categories, respectively 1) Myocardial dysfunction and heart failure (HF); 2) Coronary artery disease (CAD); 3) Valvular disease; 4) Arrhythmias; 5) Arterial hypertension; 6) Thromboembolic disease; 7) Peripheral artery disease and stroke; 8) Pulmonary hypertension; 9) Pericardial complications (Table 2) (Zamorano et al., 2016).

Table 2 – Categories of CV side-effects of chemotherapy. CAD, Coronary artery disease; HF, Heart Failure. Adapted from (Zamorano et al., 2016).

Categories of CV side-effects of chemotherapy	**1.** Myocardial dysfunction and HF
	2. CAD
	3. Valvular disease
	4. Arrhythmias
	5. Arterial hypertension
	6. Thromboembolic disease
	7. Peripheral artery disease and stroke
	8. Pulmonary hypertension
	9. Pericardial complications

More recently, López Sendón et al. suggested an alternative definition in his "CARDIOTOX" study where chemotherapy-induced cardiotoxicity was defined as a new

onset or worsening in ventricular function or myocardial damage, from baseline after initiation of chemotherapy (López-Sendón et al., 2020).

Despite the lack of precision in its definition and the fact that there seems not to be a high level of consensus between organizations at the moment, they all seem to agree on defining cardiotoxicity by a serial absolute decrease or decline below a certain level in left ventricular ejection fraction (LVEF), as the true divergence relates to the thresholds considered by each of them (Chung et al., 2018; Lambert & Thavendiranathan, 2016; Pardo Sanz & Zamorano, 2020).

The American Society of Echocardiography / European Association of Cardiovascular Imaging (ASE/EACVI) considers that in oncology patients a 10% decrease of the LVEF to an absolute value of <53% should be the cut-off point considered. However, the cut-off accepted by the European society of cardiology (ESC) slightly differs as it considers a 10% fall or higher from baseline to a LVEF value of <50% or, in alternative, a LVEF drop of >20 % which is in accordance with the definition of cardiotoxicity frequently used in trials and studies in oncology patients (Alexandre et al., 2020; López-Sendón et al., 2020; Pardo Sanz & Zamorano, 2020). Alternatively, according to the European society of medical oncology (ESMO), a symptomatic decline in LVEF of at least 5% to <55% or asymptomatic decline in LVEF of at least 10% to <55% should be interpreted as cardiotoxicity (Chung et al., 2018).

Apart from the LVEF assessment, a rise in cardiac troponin levels (hs-cTn) from baseline that exceeds the upper limit of reference can also be included in defining cardiotoxicity related to cancer treatment and enable its early diagnosis (Alexandre et al., 2020).

An alternative form of detecting this myocardial toxicity is to access the left ventricular Global Longitudinal Strain (GLS), as ESMO considers that an absolute GLS drop ≥5% or a relative drop ≥12% or, in alternative and according to the "2016 ESC Position paper on cancer treatments and cardiovascular toxicity", a GLS relative drop of >15% from baseline may suggest a risk of cardiotoxicity before significant changes in LVEF occur, representing an early opportunity to start cardioprotective therapy. Table 3 summarizes the different cut-off values of LVEF and GLS for chemotherapy-induced cardiotoxicity considered by different sources.

Table 3 – Different cut-off values of LVEF and GLS for chemotherapy-induced cardiotoxicity. Adapted from Alexandre et al., 2020; D. M. Cardinale et al., 2020; Curigliano et al., 2020; Zamorano et al., 2016.

LVEF cut-off considered	
ASE/EACVI	10% decrease of the LVEF to an absolute value of <53%
ESC	≥ 10% fall from baseline to a LVEF value of <50%
	>20 % decrease in LVEF
ESMO	≥ 5% symptomatic decrease in LVEF to a value of <55%
	≥ 10% asymptomatic decrease in LVEF to a value of <55%.
Left ventricular GLS cut-off considered	
ESC	GLS relative decrease of >15% from baseline
ESMO	≥5% GLS absolute decrease from baseline
	≥12% GLS relative decrease from baseline

Therefore, some authors have suggested an update of the definition of cardiotoxicity stating that it should include a combination of both cardiac imaging techniques and laboratory tests for better diagnostic sensitivity and specificity outcomes and also to improve prevention of chemotherapy-related adverse cardiac events (Alexandre et al., 2020; D. M. Cardinale et al., 2020; Curigliano et al., 2020; Zamorano et al., 2016).

5. Mechanisms of chemotherapy-induced cardiotoxicity

Cardiotoxic drugs are responsible for the induction of cardiac dysfunction causing ventricular dysfunction, arrhythmias, defects in the conduction system, ischemia and/or endothelial dysfunction through the induction of either reversible or irreversible myocardial injury (D. M. Cardinale et al., 2020).

Several studies have shown that the type of chemotherapy drugs influences the probability of inducing cardiotoxicity. Also, the risk of inducing cardiotoxicity is further influenced by factors such as the route of administration of chemotherapy, the duration of treatments using cardiotoxic drugs also referred to as "cumulative dose" and the dosage used (Angsutararux et al., 2015; Shaikh & Shih, 2012).

Many chemotherapy agents induce cardiac dysfunction which frequently resolves over time, however, some do not and remain permanently damaged. Therefore, several pathophysiological mechanisms have been proposed to try to explain these adverse effects of cancer therapies.

Suter and Ewer et al. divided cardiotoxic drugs into two categories according to the type of damage, "Type I" defining drugs with the potential to cause irreversible histological damages that can lead to progressive cardiovascular disease; and "Type II" drugs which are agents associated with potentially reversible damage which are usually transient (Suter & Ewer, 2013).

Type I irreversible damages are usually caused by direct cellular toxicity leading to cellular death and occurring either due to necrosis or apoptosis, depending on whether the process is energy-independent or dependent. Its more widely accepted pathophysiological mechanism to explain cardiotoxicity involves the mechanism of oxidative stress in which occurs an imbalance between reactive oxygen species (ROS) and the antioxidant capacities of the cells resulting in elevated ROS that damages cells and induce cumulative myocardial injury, resulting in both irreversible diastolic and systolic dysfunction (Angsutararux et al., 2015; Koutsoukis et al., 2018; Shaikh & Shih, 2012).

On the other hand, Type II drugs usually work by inhibiting the physiological function of myocardial cells, therefore, inducing cellular dysfunction, for example, mitochondrial and protein changes either through impaired excitation/contraction

coupling, intracellular calcium homeostasis and/or mitochondrial function or by increasing cell permeability with cell wounds resulting in a “stunned” myocardium and significant but likely reversible ventricular dysfunction. For this reason, therapy with type II agents can sometimes be reintroduced after cardiac recovery. Arrhythmogenic effects are also frequent side effects and can be due to either cellular toxicity or dysfunction with cellular membrane channels (Angsutararux et al., 2015; Koutsoukis et al., 2018; Shaikh & Shih, 2012).

Unfortunately, there are some limitations regarding this classification and it is no longer much used because these two mechanisms sometimes overlap and some type II cardiotoxic drugs have been identified as inductors of irreversible injury in about 20% of cases rather than reversible ones, either when associated with the presence of comorbidities or through potentiation of the toxic effects of type I drugs if used in combination therapies. Therefore, the most recent guidelines from American Society of Clinical Oncology (ASCO) do not recommend this distinction (D. M. Cardinale et al., 2020; Curigliano et al., 2012; Keulenaer & Leite-moreira, 2013; Koutsoukis et al., 2018; Kubota et al., 2021; S. Gillespie et al., 2012; Suter & Ewer, 2013).

Cardiotoxicity in breast cancer can appear at different timings throughout cancer treatment. It is defined as acute or subacute if cardiac dysfunction or myocardial injury develops between the initiation of treatment until up to two weeks after its ending. Arrhythmias, abnormalities in repolarization and QT intervals, acute coronary syndromes, or pericardial disease are some examples. Chronic cardiotoxicity appears afterward and can further be divided into early chronic cardiotoxicity if occurs within the first year after the end of chemotherapy, or late chronic cardiotoxicity if it appears later on (Shaikh & Shih, 2012).

Most breast cancer therapies currently available are listed below on Table 4 and of all, this review will only focus on those presenting with available and relevant data regarding cardiotoxicity, namely chemotherapy agents and human epidermal growth factor receptor-2 (HER2) targeted drug therapies.

Table 4- **Breast Cancer Therapies**. Adapted from American Cancer Society, 2019 and Waks & Winer, 2019.

<table>
<tr><th colspan="3">Breast Cancer Therapies</th></tr>
<tr><td rowspan="5">Chemotherapy</td><td colspan="2">Anthracyclines – Doxorubicin, Epirubicin or Idarubicin</td></tr>
<tr><td colspan="2">Fluoropyrimidines – 5-Fluorouracil (5-FU) or Capecitabine</td></tr>
<tr><td rowspan="2">Microtubule inhibitors</td><td>Taxanes – Paclitaxel or Docetaxel</td></tr>
<tr><td>Vinca alkaloids – Vinorelbine</td></tr>
<tr><td rowspan="2">Alkylating Agents</td><td>Platinum agents – Cisplatin or Carboplatin</td></tr>
<tr><td>Cyclophosphamide</td></tr>
<tr><td>HER2 Targeted drug therapy</td><td colspan="2">Monoclonal Antibodies – Trastuzumab or Pertuzumab</td></tr>
<tr><td rowspan="2">Hormone therapy</td><td colspan="2">Tamoxifen</td></tr>
<tr><td colspan="2">Aromatase Inhibitors</td></tr>
<tr><td>Immunotherapy</td><td colspan="2">Immune checkpoint inhibitors – PD-1 inhibitor</td></tr>
</table>

5.1. Anthracyclines

Anthracyclines are one of the most frequent drugs associated with chemotherapy-induced cardiotoxicity inducing myocyte damage that trigger continuous progressive decline of LVEF, as well as, progressive cardiac remodeling, eventually leading to symptomatic cardiomyopathy. Apart from its high cardiac toxicity, anthracyclines are highly efficient for the treatment of many tumors including breast cancer therefore, avoiding their use may have a negative impact on prognosis (D. Cardinale et al., 2020; Zamorano et al., 2016).

Anthracyclines target topoisomerase-II (Top2), inducing double-strand-deoxyribonucleic acid (ds-DNA) breaks and thus, inhibiting DNA synthesis leading to cancer cell's death by apoptosis (Beretta & Zunino, 2007). The most commonly used anthracycline drugs are Doxorubicin, Epirubicin and Idarubicin (Koutsoukis et al., 2018). Recently, new alternatives to classic anthracyclines such as Epirubicin and Idarubicin were developed through structural changes of the classic compounds with lower toxicity associated, enabling administration of higher doses. Epirubicin is a semi-synthetic epimer of Doxorubicin while Idarubicin is a structural analogue of Daunorubicin (Adão et al., 2013).

Regarding the type of cardiotoxicity induced by anthracyclines, it is referred to as being a typical example of type I cardiotoxicity which is characterized by irreversible histological changes in heart biopsies such as disorganization of myofibrils and vacuoles as well as necrosis (Fiúza, 2009; Koutsoukis et al., 2018). Its incidence in women receiving monotherapy with anthracyclines is about 4–36%. From these, 6% are clinically symptomatic while 18% present with subclinical cardiotoxicity (Lakhani et al., 2021). This type of cardiotoxicity is also dose-dependent (Chung et al., 2018; Koutsoukis et al., 2018).

As mentioned above, the primary cellular targets of anthracycline toxicity in the heart are from far cardiomyocytes, whose damage contributes to the progressive development of cardiac dysfunction, an adverse effect strongly associated with Anthracyclines' therapies. Recently, other cell types such as cardiac progenitor cells, cardiac fibroblasts, and endothelial cells have been identified as potential additional targets (D. Cardinale et al., 2020).

The underlying cellular mechanisms responsible for anthracycline-induced cardiotoxicity are not yet fully understood. Throughout the years, several hypotheses have been developed to try to explain the mechanisms involved in this type of cardiotoxicity and the reason for their particularly predilection for cardiomyocytes (Adão et al., 2013; Lakhani et al., 2021; Zamorano et al., 2016).

Anthracyclines are thought to affect cardiac tissues mostly through mechanisms involving ROS formation, DNA damage through interaction with topoisomerase II and inhibition of protein synthesis. Of those, the oxidative stress hypothesis is the most accepted pathophysiological mechanism. The latter hypothesis suggests that cardiomyocytes are damaged by the production of ROS which increases the concentration of calcium in cells' cytosol inducing cardiac dysfunction and, at higher concentrations, DNA damage. Oxidative stress further leads to lipid peroxidation and associated membrane injury (D. Cardinale et al., 2020; De Azambuja et al., 2009; Lakhani et al., 2021; Shaikh & Shih, 2012). The triggering of oxidative stress and associated DNA damaging is thought to lead to the activation of the nuclear enzyme poly (ADP) - ribose polymerase 1 (PARP-1), resulting in cellular depletion of NADþ and adenosine triphosphate triggering cell death, either through apoptotic or necrotic pathways. Moreover, PARP-1 genetic deletion as well as pharmacological inhibition with PARP inhibitors has been shown to be protective against doxorubicin-induced cardiotoxicity in mice and can possibly be combined with doxorubicin to increase chemotherapeutic efficacy and reduce cardiotoxicity (Ferdinandy et al., 2019).

Anthracyclines are also thought to suppress the synthesis of DNA, ribonucleic acid (RNA), proteins and other important transcription factors which lead to the deterioration of myocytes and sarcomere proteins and therefore, cardiac sarcopenia (Adão et al., 2013). Furthermore, other mechanisms may also play an important role in anthracyclines' cardiac toxicity such as apoptosis; transcriptional changes in intracellular adenosine triphosphate (ATP) production in cardiac myocytes; downregulation of messenger RNA expression for sarcoplasmic reticulum calcium-ATPase, which decreases cardiac contractility; prolonged drug-related depression in cardiac glutathione peroxidase activity; respiratory defects associated with mitochondrial DNA damage; and topoisomerase II inhibition, which is usually responsible for generating transient DNA

double-strand breaks during replication, transcription, or recombination (D. M. Cardinale et al., 2020; Nitiss, 2009; Shaikh & Shih, 2012).

More recently, the latter hypothesis, through its interaction with Topoisomerase II, has been suggested to be the critical mediator of anthracycline's cardiac toxicity, namely doxorubicin-induced cardiotoxicity. There are two Top2 enzymes: Top2α and Top2β. Doxorubicin targets the enzyme Top2α in cancer cells and acts by binding to both Top2α and DNA forming the Top2-doxorubicin-DNA cleavage complex, which initiates cell death and is responsible for the antineoplastic effect. However, Doxorubicin also targets the enzyme Top2β, which is expressed by cardiomyocytes and is responsible for its cardiac toxicity. In cardiac cells, the formation of the former complex induces DNA double-strand breaks, leading to activation of cell death pathways as well as ROS generation leading to doxorubicin-mediated cardiomyopathy. This theory is corroborated by studies showing that cardiomyocyte-specific deletion of the Top2β gene, which encodes the Top2β enzyme, has a protective effect on cardiomyocytes, preventing them from doxorubicin-induced DNA double-strand breaks and thus, from the development of progressive heart failure. Assuming that Top2β does not contribute to doxorubicin's antineoplastic activity, drugs that selectively target only the Top2α isozyme, should be less cardiotoxic without interfering with its therapeutic effect (D. M. Cardinale et al., 2020; Zhang et al., 2012).

There is considerable variability in patients' susceptibility to the adverse cardiotoxic effects of anthracyclines. The presence of some risk-factors can influence anthracycline-related cardiotoxicity and can help to explain these different degrees in vulnerability to similar therapies using the same class of chemotherapeutic agents. The following risk factors are only associated with early and late chronic toxicity as acute forms do not usually relate with the former (Adão et al., 2013; Zamorano et al., 2016).

The total cumulative lifetime dose of anthracyclines represents the main risk factor responsible for the induction of cardiotoxicity and consequently the development of heart failure (Shaikh & Shih, 2012). Other important risk factors include 1) the route of administration; 2) duration of therapy; 3) type of anthracycline agent used; 4) previous mediastinal irradiation; 5) concomitant use of other agents with known cardiotoxicity; 6) concomitant cardiovascular (CV) disease; 7) female sex; 8) genetic predisposition; 9) age, including older than 65 years or children and young adults; 10)

increase in time that passed since the end of therapy; 11) increase in cardiac biomarkers, during and after administration (Adão et al., 2013; Curigliano et al., 2012; S. Gillespie et al., 2012; Zamorano et al., 2016).

As discussed above, patients with higher expression of Topoisomerase IIβ in cardiomyocytes may be more susceptible to doxorubicin-induced cardiotoxicity (Zhang et al., 2012).

When referring to the doses administered the higher the cumulative dose, the greater the risk of inducing cardiotoxicity. Higher single doses also increase the risk (Curigliano et al., 2012). The lifetime dose of doxorubicin recommended is usually limited to 450 to 550 mg/m^2. Some consider a lower threshold of 250 mg/m^2. Several retrospective cohort studies have shown that the incidence of heart failure progressively increases with cumulative doses superior to the threshold of 400 mg/m^2, with its incidence being 5% at a dose of 400 mg/m^2 and rising up to 48% when a cumulative dose of 700 mg/m^2 is reached. However, it is important to note that lower doses, within the dosage recommended, are also associated with a considerable risk for cardiotoxicity (Alexandre et al., 2020; Koutsoukis et al., 2018; Mitchell & Lenihan, 2020). Similar to doxorubicin, the amount of cumulative dose of epirubicin administered also increases the risk of inducing cardiac adverse effects. In patients with breast cancer, a cumulative dose higher than 600 mg/m2 has a significantly higher risk of cardiac dysfunction (Mitchell & Lenihan, 2020).

If one or more of these risk factors are present, the cumulative dose vs. cardiotoxicity risk curve will shift to the left and alternative chemotherapy agents should be considered or closer monitoring of these patients is recommended (Zamorano et al., 2016).

Regarding the route of administration, intravenous bolus administration is associated with a higher risk of cardiotoxicity, when compared to slower infusion protocols. In an attempt to reduce anthracyclines absorption in cardiac cells, new alternatives to passive deliver agents such as Doxycycline and Daunorubicin to the tumor site through their incorporation into liposomes are being developed. These liposomal anthracyclines, such as pegylated liposomal doxorubicin, have comparable effectiveness and are significantly less cardiotoxic, as the amount of drug encapsulated

increases and its elimination time extends, presenting a potential alternative choice for high-risk patients (Adão et al., 2013; Koutsoukis et al., 2018; S. Gillespie et al., 2012).

As mentioned above, the type of anthracycline agent used also plays an important role in the degree of adverse cardiac effects. In this setting, doxorubicin is considered the most cardiotoxic, whereas epirubicin and mitoxantrone are associated with lesser toxicity (S. Gillespie et al., 2012).

In terms of adverse effects, cardiac dysfunction represents the most common manifestation of cardiotoxicity related to the use of anthracyclines. Additionally, these chemotherapeutic agents can also be responsible for electrical changes and the rise in biomarkers among others, referred below (table 5) (Chung et al., 2018; Koutsoukis et al., 2018).

According to the timing of occurrence, anthracyclines-induced cardiotoxicity can be classified as “Acute” and “Chronic”, with the latter subdividing in “Early-onset” and “late-onset” (Curigliano et al., 2012).

Acute anthracycline-related cardiotoxicity is rare, developing in less than 1% of patients immediately within the beginning of treatment until 14 days after its end. It is usually temporary and reverts within weeks after discontinuation of therapy regardless of the dosage administered. However, some may induce permanent myocyte injury and progress to chronic toxicity. It is not possible to identify with certainty which will evolve to permanent cardiotoxicity but, the elevation of cardiac biomarkers can be helpful in identifying patients at greater risk (Adão et al., 2013; D. M. Cardinale et al., 2020; Zamorano et al., 2016).

In terms of clinical presentation, acute anthracycline-related cardiotoxicity frequently manifests as transient left ventricular (LV) dysfunction characterized by a decline in ejection fraction. Other forms of presentation include electrocardiographic changes, such as alterations of ventricular repolarization, changes in QT interval and ventricular and supraventricular arrhythmias or atrial fibrillation. Myocardial ischemia, acute coronary syndromes, acute heart failure and less frequently myocarditis and pericarditis can also occur (Adão et al., 2013; Koutsoukis et al., 2018; Shaikh & Shih, 2012).

Chronic anthracycline-related cardiotoxicity can be divided into early or late, according to the time of symptom onset. Early-onset chronic cardiac toxicity is the

principal form of anthracycline-induced cardiotoxicity occurring in 1.6% to 2.1% of patients within the first year, with a peak in incidence at 3 months after treatment and progressive decline in FEVE throughout time. Late-onset chronic cardiotoxicity develops years to decades afterward in 1.6% to 5% of patients and may only become clinically apparent after 10 to 20 years. Unlike Acute cardiotoxicity, both types are dose-dependent and irreversible, usually with reduced response to therapy and poorer prognosis (Adão et al., 2013; D. M. Cardinale et al., 2020; Curigliano et al., 2012; Shaikh & Shih, 2012).

Clinically, chronic cardiotoxicity usually presents as progressive ventricular dysfunction, either systolic or diastolic, that can evolve to severe heart failure and possibly death (Adão et al., 2013; Shaikh & Shih, 2012).

Table 5 summarizes the principal aspects regarding the cardiotoxicity of Anthracyclines discussed in this chapter.

Table 5 - Cardiotoxicity of Anthracyclines: incidence, adverse effects and mechanisms.
ACS, Acute coronary syndromes; Eo, Early-onset; HF, Heart Failure; Lo, Late-onset; LV, Left-Ventricular; MI, Myocardial Ischemia; ROS, Reactive oxygen species; VT/SVT, Ventricular/supraventricular. Adapted from Adão et al., 2013; D. Cardinale et al., 2020; Fiúza, 2009; Koutsoukis et al., 2018; Lakhani et al., 2021; Mitchell & Lenihan, 2020; Nitiss, 2009; Purkayastha et al., 2017; Shaikh & Shih, 2012; Zamorano et al., 2016.

Cardiotoxicity of Anthracyclines			
Anthracycline agent	**Incidence**	**CV adverse effects**	**Mechanisms of cardiotoxicity**
Anthracyclines, in general	**Overall:** 4-36% -Acute: <1% -Eo-chronic: 1.6-2.1% -Lo-chronic: 1.6-5%	**Type I; Dose dependent.** **Acute:** HF, ventricular repolarization and QT interval abnormalities, arrhythmias (VT/SVT), myocardial ischemia, ACS, myocarditis, pericarditis. **Chronic:** LV dysfunction	**Main target:** cardiomyocytes -**ROS formation:** Oxidative stress, mitochondrial DNA damage, PARP-1 activation, Apoptosis **-Inhibition of topoisomerase IIβ:** DNA damage **-Apoptosis** **-Inhibition of proteins' synthesis:** Apoptosis, cardiac sarcopenia, decreased contractility.
Doxorubicin	400 mg/m^2: >5% 700 mg/m^2: 48% 800 mg/m^2: 100%		
Epirubicin	900 mg/m^2: >5%		
Idarubicin	150 mg/m^2: >5%		
Daunorubicin	800 mg/m^2: >5%		

5.2. HER-2 Inhibitors

Human epidermal growth factor receptor-2 (HER2) gene, also named erbB2 or HER2/neu is an oncogene and one of four tyrosine kinases of the epidermal growth factor (EGF) receptor (EGFR) or ERBB family, whose main function is to mediate cell proliferation, differentiation and survival. Its regulation is accomplished by limiting receptor expression as well as the quantity and affinity of ligands. Imbalance at any of these points leads to oncogenesis. Thus, ERBB2 gene amplification and/or overexpression of HER2 receptor is present in up to 30% of breast cancers and carries a poor prognosis (Cote et al., 2012; Dawood et al., 2010; Fiúza, 2009).

Trastuzumab is a humanized monoclonal antibody that binds selectively to the extracellular domain of the HER2 receptor triggering its internalization and degradation. Other HER2 inhibitors include Pertuzumab and lapatinib (Alexandre et al., 2020; Dawood et al., 2010; Vu & Claret, 2012).

Unlike anthracyclines, the cardiotoxicity associated with trastuzumab and other HER-2 inhibitors is not dose-dependent and, in adult cardiac cells the associated LV dysfunction induced by these agents is usually transient and reversible with the interruption of therapy and/or initiation of heart failure therapies, as they do not lead to cell death nor induce structural changes on histological exams and are therefore associated with type II cardiotoxicity resulting in better prognosis (Adão et al., 2013; De Azambuja et al., 2009; Fiúza, 2009; Zamorano et al., 2016).

Despite their high efficacy in treating HER2+ breast cancer, cardiotoxicity was the main reason for suspending therapy in 13.5% of patients who interrupted chemotherapy due to adverse effects associated with HER2 inhibitors (Zamorano et al., 2016). Monotherapy with trastuzumab is associated with a 1.7%-7% risk of cardiac adverse effects with its incidence increasing up to 27% if concomitant therapy with an anthracycline is added. Nevertheless, it is important to note that most patients who were treated with monotherapy using trastuzumab, were previously exposed to anthracyclines and for this reason, it is not clear whether the cardiotoxicity observed in this group can be related to trastuzumab or entirely a consequence of previous anthracycline exposure (Cote et al., 2012; Fiúza, 2009; Guarneri et al., 2006; Keefe, 2002; Zamorano et al., 2016). As described in table 6, other anti-HER2-targeted therapies such

as bevacizumab and pertuzumab appear to have a similar risk of inducing cardiotoxicity, but more studies are needed in this matter (Zamorano et al., 2016).

Unfortunately, the **mechanisms** underlying the cardiotoxicity associated with these agents are still not well understood. Multiple hypotheses on potential mechanisms responsible for trastuzumab-induced cardiotoxicity have been proposed and include: 1) Interference with cardiomyocyte survival pathways; 2) Immune-mediated destruction of cardiomyocytes; 3) Impaired HER-2 signalling required for the maintenance of cardiac contractility, and 4) Drug interaction with anthracyclines (De Azambuja et al., 2009).

Regarding the inhibition of Neuregulin-ERBB (NRG–ERBB), an HER2 receptor, signalling and interference with cardiomyocyte survival, this pathway is essential for cardiac development and important for cardiomyocyte survival in the context of acute cardiac stress, in which it leads to increased HER2 expression and activation of HER2 receptors by neuregulin (NRG). In cardiac cells, overexpression of HER2 and/or NRG-mediated activation of the HER2 signalling pathway increases protection against oxidative stress and prevents cell death. Thus, inhibition of HER2 by trastuzumab disrupts this pathway and induces ventricular dysfunction as shown in studies using mice with inactivation of Erbb2 or Nrg1, in which consequent inhibition of NRG–ERBB signalling led to the development of dilated cardiomyopathy with impaired contractility, relaxation, reduced adaptation to pressure overload and greater sensitivity to anthracycline toxicity. Following the inactivation of the referred pathway, other alternative survival pathways are activated and are usually sufficient to prevent the cell loss and progression to heart failure. However, in certain patients with pre-existing cardiovascular disease or who are under treatment with anthracyclines following previous therapy with trastuzumab, irreversible loss of cardiomyocytes can develop as cardiac stress signals are probably already activated (Adão et al., 2013; Chien, 2006; Cote et al., 2012; Keefe, 2002).

Nevertheless, there is evidence that the cardiotoxic mechanism of trastuzumab not only involves HER2 inhibition, as studies demonstrated low cardiac toxicity followed by the use of Lapatinib, a tyrosine kinase inhibitor of both HER2 and EGF. Trastuzumab, particularly its IgG1 domain, is thought to trigger cytotoxic immune reactions on cardiomyocytes, leading to Immune-mediated destruction of cardiomyocytes. Another

mechanism can involve cardiac contractility as impairment of HER-2 signaling, induced by trastuzumab after binding to HER2, modulates mitochondrial integrity via the BCL-X family of proteins, leading to ATP depletion and contractile dysfunction (Adão et al., 2013; De Azambuja et al., 2009).

More recent studies reported that HER-2 signalling also affects the sympathovagal control systems of the heart as NRG is associated with antiadrenergic effects, reducing cardiac output as well as blood pressure. Experiences using neuregulin-deficient mice and inhibition of HER-2 signalling are thought to be associated with an increase in the resting sympathetic tone as well as with the decreased response to beta-agonists therapy (De Azambuja et al., 2009; Keefe, 2002).

Several risk factors increase the risk of HER-2 inhibitors-induced cardiotoxicity. Amongst all, concomitant use of anthracyclines is the primary risk factor. As already discussed, anthracycline can induce oxidative stress, however, cardiac injury is frequently attenuated by the cardioportective effects of HER2/neu receptor signaling. Hence, by inhibiting HER2/neu signaling, HER2 inhibitors substantially worsen anthracycline-induced cardiac injury through unopposed oxidative stress (De Azambuja et al., 2009). Furthermore, the risk of cardiotoxicity is higher, the higher the cumulative dose of anthracyclines previously administered, particularly at cumulative doses superior to >400 mg/m^2, according to Fiúza, 2009, while other authors like Adão *et al*. consider a lower threshold of >300mg/m^2. The short time interval between anthracycline and HER-2 inhibitor therapy is also considered a risk factor as well as previous exposure to other drugs such as paclitaxel (Adão et al., 2013; Fiúza, 2009; Zamorano et al., 2016). However, in patients without cardiac disease, the use of trastuzumab after anthracyclines or the choosing of an anthracycline-free chemotherapy regimen significantly reduces the incidence of clinical ventricular dysfunction and remains a valid option as adjuvant therapy for breast cancer therapy (De Azambuja et al., 2009; Zamorano et al., 2016).

Other risk factors that have been identified to increase the risk of trastuzumab cardiac toxicity include: 1) Pre-existing cardiovascular disease, particularly low LVEF and arterial hypertension, 2) Age >60 years, and 3) Obesity or body mass index superior to 25 (Adão et al., 2013; Fiúza, 2009; Zamorano et al., 2016).

From those 13.5% of patients who suspended chemotherapy due to cardiotoxicity, 30% manifested clinical heart failure, whereas 70% presented with asymptomatic left ventricular systolic dysfunction (LVSD). Regarding the timing of occurrence, the cardiotoxic effects associated with trastuzumab are usually acute and typically occur during treatment (Zamorano et al., 2016). Concerning late cardiotoxicity, long-term follow-up studies (up to 10 years) showed a reassuring low incidence of late-onset HF in patients with low cardiovascular risk who received treatment with trastuzumab (Advani et al., 2016; De Azambuja et al., 2014; Goldhirsch et al., 2013; Romond et al., 2012).

Frequent adverse effects include a decline in LVEF, congestive heart failure and arrhythmias and are listed in Table 6, together with the cardiotoxicity incidence and mechanisms associated according to the HER-2 inhibitor agent (Angsutararux et al., 2015; Moslehi, 2016).

Table 6 - Cardiotoxicity of HER-2 Inhibitors. ANT, Anthracycline; HF, Heart Failure; HER-2, Human epidermal growth factor receptor-2; LVEF, Left-Ventricular Ejection Fraction; TRA, Trastuzumab. Adapted from Adão et al., 2013; Angsutararux et al., 2015; De Azambuja et al., 2009; Fiúza, 2009; Keefe, 2002; Moslehi, 2016 and Zamorano et al., 2016.

Cardiotoxicity of HER-2 Inhibitors			
HER-2 Inhibitors	**Incidence**	**CV adverse effects**	**Mechanisms of cardiotoxicity**
Trastuzumab	Monotherapy: 1.7-7% +ANT association: 20.1-27%	**Type II; Dose-independent. Acute >> Late** LVEF decline; HF; arrhythmias.	**-HER-2 receptor (NRG–ERBB) inhibition:** Cardiomyocyte survival pathways, contractility and sympathovagal impairments. **-Immune-mediated cytotoxic reactions**, by TRA's IgG1 domain.**-Drug interaction with ANT:** ANT toxicity potentiation by HER2 inhibition through unopposed oxidative stress.
Bevacizumab	1.6-4%		
Pertuzumab	0.7-1.2%		

5.3. Fluoropyrimidines

Fluoropyrimidines are antimetabolites that suppress DNA synthesis by inhibiting the action of thymidylate synthase, thus impairing cell survival (Jurczyk et al., 2021; Mendes, 2018).

Despite being the third most used class of chemotherapeutic agents, these drugs have important adverse effects including the ability to induce cardiotoxicity. In fact, fluoropyrimidines-associated cardiotoxicity is one of the most common and serious side effects of these therapies and is associated with a mortality rate oscillating from 1.6% to 10.2% (Jurczyk et al., 2021; Mendes, 2018).

Drugs that form part of this class of chemotherapeutic agents include, for example, fluorouracil (5-FU) and capecitabine. While 5-FU is administered intravenously, its prodrug, capecitabine, presents as an oral alternative.

Although still unclear, cardiotoxicity associated with the use of fluoropyrimidines does not appear to be dependent on the dosage administered and tends to be reversible as some symptoms, such as chest pain, usually improve after treatment interruption (Koutsoukis et al., 2018; Shiga & Hiraide, 2020). However, it is usually irreversible, if a myocardial infarction occurs (Purkayastha et al., 2017).

According to the ESC, the incidence of myocardial ischemia in patients treated with fluoropyrimidines can reach up to 10% of patients, varying according to the dosages and route of administration (Jurczyk et al., 2021; Mendes, 2018). It is also important to note that this incidence may be underestimated because up to 7–10% of patients may present with silent myocardial ischemia (Zamorano et al., 2016).

Unfortunately, the precise mechanisms of cardiotoxicity induced by fluoropyrimidines remain not completely understood. However, several hypotheses have been suggested believing that fluoropyrimidines-associated cardiotoxicity is caused by a combination of multiple mechanisms including endothelial damage, direct myocardial injury and coronary arterial vasospasm, among others (Jurczyk et al., 2021; Koutsoukis et al., 2018; Zamorano et al., 2016). Of these and according to the ESC, the main pathophysiological mechanism to be considered is endothelial damage and consequent induction of coronary vasospasm (Jurczyk et al., 2021; Shiga & Hiraide, 2020).

A possible mechanism that has been suggested evokes that endothelial damage induced by 5-FU leads to endothelial dysfunction which decreases the production of nitric oxide and consequent impairs nitric oxide-dependent vasodilation (Mendes, 2018). Another possibility that may contribute to vasoconstriction is acetylcholine-induced smooth muscle contraction as due to endothelial dysfunction acetylcholine induces paradoxical vasoconstriction instead of vasodilation. Moreover, primary smooth muscle dysfunction can also occur and is thought to be mediated by the activation of protein kinase C (PK-C) which causes endothelium-independent vasoconstriction of smooth muscle in the presence of a normal endothelium and is independent of angiotensin II (Jurczyk et al., 2021; Shiga & Hiraide, 2020).

On the other hand, endothelial-dependent damage secondary to therapy with fluoropyrimidines can also lead to an increase of vasoconstrictors in plasma, such as urotensin-2 and endothelin-1, a peptide with vasoconstrictor properties involved in the regulation of coronary artery tonus, whose plasma levels were found to be elevated in patients presenting with 5-FU-related cardiovascular toxicity. The resulting coronary contraction leads to a decrease in blood oxygen supply which can evolve to cardiomyocytes' ischemia. Furthermore, endothelial lesions also increase the risk of thrombotic events in these patients (Jurczyk et al., 2021; Koutsoukis et al., 2018; Shiga & Hiraide, 2020).

A decrease in oxygen binding to hemoglobin and its transportation is another potential mechanism that, similar to endothelial damage, is believed to contribute to the reduction of blood supply to cardiac tissue. This may be due to impairments in the energetic metabolism of erythrocytes secondary to a rapid increase in oxygen consumption as well as a decrease in ATP levels inducing structural changes of cell membranes, such as irreversible echinocytosis or increased membrane fluidity and reduction of erythrocytes' ability to deliver oxygen (Jurczyk et al., 2021; Koutsoukis et al., 2018; Shiga & Hiraide, 2020).

An alternative mechanism of 5-FU cardiotoxicity concerns direct cellular damage secondary to the production of ROS and oxidative stress which induces mitochondrial disturbances and blocks cell proliferation cycles leading to the activation of apoptosis pathways in endothelial cells and cardiomyocytes. It is important to note that, given their high number of mitochondria, cardiomyocytes are particularly vulnerable to

damage. Other mechanisms such as reduction in the activity of glutathione and lipid peroxidation are also believed to be involved (Jurczyk et al., 2021; Shiga & Hiraide, 2020).

There is also a theory that suggests that the degradation of 5-FU results in highly toxic metabolites that can interfere with the Krebs cycle by inhibiting the production of energy in mitochondria leading to cardiotoxicity (Shiga & Hiraide, 2020).

In summary, the resulting coronary arteries contraction leads to hypoperfusion of myocardial tissue and consequent reduction in oxygen supply to cardiomyocyte which, combined with an increase in oxygen demand of cardiac cells secondary to mitochondria disturbances results in myocardial dysfunction and evolution to ischemia (Shiga & Hiraide, 2020).

There are several risk factors that increase the risk of fluoropyrimidines-induced cardiotoxicity, one of which concerns the route of administration as continuous infusion of 5-FU is associated with a 7 to 18% higher risk of cardiac adverse effects when compared with intravenous bolus. Unfortunately, studies have shown that bolus administration is not as effective as a continuous infusion in cancer therapy (Jurczyk et al., 2021).

Other factors that are associated with a higher probability of inducing cardiac toxicity following therapy with fluoropyrimidines include re-exposure to the agent in question, preexisting cardiovascular disease or cardiovascular risk factors, other pre-existing conditions like renal insufficiency, chemotherapy with combined therapeutic schemes using multiple drugs, for example, taxanes or anthracyclines (Adão et al., 2013; Jurczyk et al., 2021; Mendes, 2018). There are also genetic enzymatic polymorphisms in drugs' catabolizing pathways that are believed to increase vulnerability to 5-FU-induced cardiotoxicity namely dihydropyrimidine dehydrogenase (DPD) deficiency, which has been detected in 3% to 5% of patients treated with 5-FU, and is associated with a decreased capacity to its degradation, resulting in a higher risk of toxicity (Jurczyk et al., 2021).

Regarding the agent used, there seems to be no difference between 5-FU and capecitabine in terms of cardiac adverse effects associated with therapy using each of these agents (Shiga & Hiraide, 2020).

According to some studies, cardiotoxicity after 5-FU therapy can be classified as acute cardiotoxicity, as it usually develops after the first dose. The time of onset of the first symptoms is not consensual in the literature (Jurczyk et al., 2021). According to Jurczyk et al. symptoms usually develop within 72 hours after the start of therapy, but some adverse effects may only appear up to 2 days later. Alternative sources consider an earlier onset at 12 hours. On the other hand, Koutsoukis *et al.* and other authors agree that if a continuous intravenous infusion is given, the onset of 5-fluorouracil-induced cardiac toxicity can manifest from the second up to the fifth day of treatment.

In terms of adverse effects, cardiotoxicity of 5-FU treatment has been associated with myocardial ischemia and coronary artery disease that can become clinically evident as acute coronary syndromes, heart failure, Prinzmetal's angina, coronary dissection, cardiomyopathy, arrhythmias, QT interval prolongation, ST changes or even sudden death. Furthermore, it can also be responsible for peripheral arterial disease, including Raynaud's phenomenon. Of all, angina is the most common symptom reported (Jurczyk et al., 2021; Khouri et al., 2013; Shiga & Hiraide, 2020).

The Table 7 summarizes the principal aspects regarding the cardiotoxicity of fluoropyrimidines discussed in this chapter.

Table 7 - Cardiotoxicity of HER-2 Inhibitors. 5-FU, 5-Fluorouracil; ACS, Acute coronary syndrome; CAD, Coronary artery disease; Hb, hemoglobin; HF, Heart Failure; MI, Myocardial Ischemia; NO, Nitric Oxide; PAD, Peripheral arterial disease; ROS, Reactive Oxygen Species; SMI, Silent Myocardial Ischemia. Adapted from Adão et al., 2013; Angsutararux et al., 2015; Jurczyk et al., 2021; Khouri et al., 2013; Mendes, 2018; Purkayastha et al., 2017; Shiga & Hiraide, 2020; Zamorano et al., 2016.

Cardiotoxicity of Fluoropyrimidines			
Fluoropyrimidines	**Incidence**	**CV adverse effects**	**Mechanisms of cardiotoxicity**
5-FU	Up to 10% (may be underestimated due to SMI)	**Dose-independent (?) Irreversible, if MI.** MI (Angina, ACS, CAD); SMI; Prinzmetal's angina; congestive HF; arrhythmias; coronary dissection; PAD; Raynaud's phenomenon; QT interval prolongation; ST changes.	-**Endothelial damage**: - **Coronary spasm**: decreased NO production of and NO-dependent vasodilation inducing MI. - **Increase of plasmatic vasoconstrictors** (urotensin-2 and endothelin-1): impairment in coronary artery tonus. -**Direct cardiomyocyte damage**: ROS production, oxidative stress. -**Decrease in oxygen binding to Hb**: reduced blood supply and MI.
Capecitabine			

5.4. Microtubule inhibitors

Antimicrotubular drugs have been used in breast cancer therapy for a long time and they work by disturbing microtubule dynamics, particularly disrupting the mitotic spindle and consequently interrupting their division processes, therefore interfering with cell cycles leading to cell death (Fanale et al., 2015; Joshi et al., 2021).

According to their mechanism of action, drugs within this class can be classified as microtubule-stabilizing and microtubule destabilizing agents. Taxanes, which include agents like paclitaxel and docetaxel, are an example of microtubule-stabilizing drugs and they work by promoting the polymerization of microtubules, through the binding of the agent to the tubulin polymer leading to stabilization of microtubules. On the other hand, the class classified as microtubule destabilizing agents works by binding to tubulin dimers and destabilizing microtubules through induction of microtubule depolymerization. Examples of this group are vinca alkaloids in which vinorelbine is the agent still in use, particularly in patients with metastatic breast cancer. It is important to note that cardiotoxicity related to therapy using vinca alkaloids is rare (Bomzer, 2014; Fanale et al., 2015; Joshi et al., 2021).

Taxanes are classified as type I cardiotoxicity agents but, in the majority of patients, these cardiotoxic effects associated with antimicrotubule agents are usually asymptomatic or mild (Mladěnka et al., 2018).

Although the incidence of severe cardiac adverse effects is low, it is important to consider that the majority of studies concerning antimicrotubules-induced cardiotoxicity included patients receiving therapy with multiple chemotherapeutic agents, namely associations combining taxanes and anthracyclines and hence, it is difficult to discern the true cause of cardiac toxicity and more studies are needed on this matter (Joshi et al., 2021).

Regarding the differences in the incidence of some cardiac adverse effects between agents within the same class, according to Zamorano *et al.* docetaxel was associated with a 2.3% to 13% risk of developing LVEF when compared with the less than 1% risk attributed to paclitaxel (Zamorano et al., 2016). Alternatively, according to a study by Han *et al.*, 5% of patients who received paclitaxel developed cardiotoxicity (Han et al., 2017).

Concerning the mechanisms through which microtubule inhibitors induce cardiotoxicity and despite the lack of robust evidence in the literature, some hypotheses have been formulated throughout the years. Given that taxanes are strongly associated with the development of conduction abnormalities particularly bradycardia as well as heart block, it was hypothesized that microtubules could have an important role in the regulation of intracellular calcium. Studies in patients receiving therapy with paclitaxel have shown that exposure to this agent was associated with a temporal reduction between the state of maximum contraction to relaxation of cardiomyocytes caused by an acceleration of spontaneous calcium release in these cells, increasing the risk of inducing cardiac arrhythmias. This premature calcium release is believed to be triggered through the interaction of these agents with the neuronal calcium sensor 1, a calcium-binding protein known to regulate the inositol-1,4,5- trisphosphate receptor. Mladěnka et al., for instance, proposes an alternative theory suggesting that taxanes are responsible for blocking both Na+ and Ca2+ channels in cardiomyocytes (Chaulin et al., 2020; Han et al., 2017; Mladěnka et al., 2018). Another proposed theory regarding the mechanisms of antimicrotubule agents-related cardiotoxicity concerns the induction of massive histamine release which was supported by studies showing that stimulation of histamine receptors in animal cardiomyocytes led to the generation of arrhythmias. Furthermore and regarding paclitaxel, the arrhythmogenic effects of this agent can also be related to the delivery vehicle in which it is diluted, namely polyethoxylated castor oil (Cremophor EL) which triggers histamine release (Albini et al., 2010; Bomzer, 2014; Chaulin et al., 2020).

Taxanes-induced cardiotoxicity is also believed to occur through direct myocardial injury via effects on cellular organelles, leading to a later decrease in LVEF. It has also been hypothesized that the microtubule inhibitors´ effects on cellular cycles could preferentially impact cardiac endothelium and therefore potentiate ischemia (Albini et al., 2010; Joshi et al., 2021).

Several risk factors have been identified to increase the generation of cardiotoxicity in the setting of therapies using microtubule inhibitors such as taxanes. From those, one of the most relevant is the use of therapeutic schemes combining multiple classes of chemotherapeutic agents, particularly those combining a taxane with an anthracycline agent. Taxanes appear to potentiate the cardiotoxicity of anthracyclines, with paclitaxel

being associated with a greater risk when compared to docetaxel. Studies have demonstrated that monotherapy with a taxane has a significantly lower cardiotoxic risk when compared with combined regimens (Joshi et al., 2021; Zamorano et al., 2016).

Taxanes appear to potentiate the metabolism of anthracyclines to toxic metabolites, such as doxorubicinol by reducing doxorubicin elimination and enhancing the enzymatic conversion of doxorubicin, possibly through allosteric modulation of carbonyl or aldo/keto reductases. Given this and when needed, it is recommended to separate the administration of both classes, administering first the anthracycline followed by the taxane agent and/or, in alternative, limit the cumulative dose of anthracycline administered to 360 mg/m^2 (Minotti et al., 2001; Zamorano et al., 2016).

Apart from anthracyclines, concomitant therapy with other agents like trastuzumab, an Her2-inhibitor, potentiates the cardiotoxic effects, mainly through inhibition of the ErbB2/neuregulin pathway leading to an additive dysfunction in cardiomyocytes (Chaulin et al., 2020; Joshi et al., 2021).

Cardiotoxicity secondary to therapies using antimicrotubule agents, particularly taxanes, frequently manifests with heart failure, myocardial ischemia, and/or conduction abnormalities, such as bradyarrhythmias (sinus bradycardia and atrioventricular block) and tachyarrhythmias (ventricular tachycardia and supraventricular arrhythmias, including atrial fibrillation, atrial flutter and atrial tachycardia). The latter is the most commonly described cardiotoxic adverse effect associated with antimicrotubules-induced cardiotoxicity, with asymptomatic sinus bradycardia being the most frequent clinical presentation, occurring in up to 31% of patients, according to Han et al. (Albini et al., 2010; Chaulin et al., 2020; Han et al., 2017; Joshi et al., 2021).

Nevertheless, arrhythmias secondary to taxanes-induced cardiotoxicity are usually benign and transient as it has not yet been identified any association with the development of fatal arrhythmias and they frequently resolve after the end of therapy (Chaulin et al., 2020; Joshi et al., 2021).

When referring to the incidence of myocardial ischemia according to the literature available, it has been identified in 3 to 5% of patients receiving taxanes and who also had pre-existing conditions identified as risk factors for the development of

cardiotoxicity. As for heart failure, chemotherapy using taxanes have been associated with a 2 to 8% risk (Joshi et al., 2021).

Studies in patients receiving therapy with paclitaxel have shown that prolongation of the QT interval is the most significant acute arrhythmogenic adverse effect seen on electrocardiography with bradycardia and heart block being the most frequent clinical presentation in this setting. On the other hand and according to Bomzer *et al.* these adverse effects have not been identified with docetaxel (Bomzer, 2014; Osman & Elkady, 2017).

Vinca alkaloids are rarely associated with cardiotoxicity effects however, although infrequent, there is some evidence that vincristine may be associated with pulmonary hypertension as well as myocardial ischemia, particularly if administrated in combination with other chemotherapeutic drugs (Joshi et al., 2021).

Table 8 summarizes the principal aspects regarding the cardiotoxicity of microtubule inhibitors discussed in this chapter.

Table 8 - Cardiotoxicity of Microtubule inhibitors. AF, Atrial fibrillation; ANT, Anthracycline; AT, Atrial tachycardia; AV, Atrioventricular; CHF, Congestive heart failure; DTX, Docetaxel; HT, Hypertension; MI, Myocardial ischemia; PTX, Paclitaxel; SVT, Supraventricular tachycardia; VT, Ventricular tachycardia. Adapted from (Adão et al., 2013; Albini et al., 2010; Angsutararux et al., 2015; Bomzer, 2014; Chaulin et al., 2020; Fanale et al., 2015; Han et al., 2017; Joshi et al., 2021; Moslehi, 2016; Purkayastha et al., 2017; Zamorano et al., 2016)

Cardiotoxicity of Microtubule inhibitors			
Drug examples	**Incidence**	**CV adverse effects**	**Mechanisms of cardiotoxicity**
Taxanes (DTX,PTX)	**DTX:** 2.3-13% **PTX:** 1-5% **+ ANT:** 18%	**Type I.** **Conduction abnormalities:** - Bradyarrhythmias: sinus bradycardia, AV-block. - Tachyarrhythmias (VT, SVT-AF, atrial flutter, AT) - [PTX]: QT prolongation **CHF** (2-8%), **MI** (3-5%).	**- Impaired regulation of intracellular calcium:** premature calcium release (neuronal calcium sensor 1) - **Histamine release** through stimulation of its receptors in cardiomyocytes: arrhythmias. **- Direct myocardial injury**
Vinca alkaloids (vinorelbine, vincristine)	Unknown (rare)	MI, coronary spasm, raynaud's phenomenon, Pulmonary HT.	

5.5 Alkylating agents

Alkylating agents such as cyclophosphamide, cisplatin or ifosfamide are part of a frequently used class of chemotherapeutic drugs that works by binding to negatively charged DNA sites, inducing strand breaks and cross-linking (Han et al., 2017).

The risk of cardiotoxicity is dose-dependent and hence classified as type I cardiotoxicity, usually occurring within the first 10 days after the first administration (Curigliano et al., 2012; Han et al., 2017; Madeddu et al., 2016; Mladěnka et al., 2018).

Cardiac toxicity arising from therapy with cyclophosphamide is estimated to occur in 7% to 28% of patients. Regarding Ifosfamide, cardiotoxicity is present in 0.5% to 17% of patients. Cisplatin is also relatively rarely associated with overt cardiotoxicity occurring, approximately, in 2% of patients (Zamorano et al., 2016).

Regarding the mechanisms through which the therapeutic administration of alkylating agents leads to cardiotoxicity, some degree of uncertainty about the exact mechanisms involved remains. Nevertheless, it is believed that the cardiotoxicity of these agents such as cyclophosphamide or cisplatin arises from direct endothelium injury and consequent leakage of metabolites and their toxic effects on cardiomyocytes.

Alternatively, these chemotherapeutic agents have also been associated with mitochondrial dysfunction through impairment of cellular respiration mostly due to ROS production and oxidative stress leading to injury of mitochondrial membranes of cardiac cells and eventually triggering cell death.

The generation of oxidative stress is also associated with the induction of a prothrombotic state and additionally, cisplatin is also thought to induce platelet activation and aggregation as well as to increase levels of von Willebrand factor and triggering vasospasm (Madeddu et al., 2016).

A study in animal models concluded that cisplatin was associated with a significant rise in serum troponin I as well as a concomitant decrease in antioxidants thus leading to injury of mitochondrial and nuclear DNA. Furthermore, cardiac ischemic disease associated with cyclophosphamide therapy can also result from generation of intracapillary microemboli (Adão et al., 2013; Madeddu et al., 2016).

Ifosfamide's cardiotoxic mechanisms appear to be similar to those of cyclophosphamide (Madeddu et al., 2016).

Risk factors believed to contribute to an increasing risk of cardiac toxicity include total bolus administration regimens, older age, therapeutic schemes combining multiple chemotherapeutic drugs as well as concomitant thoracic radiation (Han et al., 2017; Zamorano et al., 2016). As mentioned before, by being dose-dependent, cardiac toxicity arising from therapy with cyclophosphamide is uncommon at low doses, with most cases occurring at higher doses whose cutoff value considered is ≥140 mg/kg, according to Zamorano *et al.* In alternative, Curigliano *et al.* suggest a higher value of >150 mg/kg or 1.5 g/m^2/day (Bomzer, 2014; Curigliano et al., 2012; Zamorano et al., 2016). Concerning Ifosfamide, for doses <10 g/m^2, the risk is also relatively low affecting only 0.5% of patients. In contrast, doses ≥12.5 g/m^2 are associated with a 17% risk of cardiotoxicity (Zamorano et al., 2016).

Clinically, following administration of high doses, the cardiotoxicity of cyclophosphamide frequently manifests as acute heart failure, which is secondary to the induction of left ventricular dysfunction that can sometimes be reversible. Mild functional mitral regurgitation has also been identified. Other symptoms of cardiotoxicity identified in the literature include frequently asymptomatic pericardial effusion as well as acute pericarditis, myopericarditis and arrhythmias (Adão et al., 2013; Albini et al., 2010; Bomzer, 2014; Han et al., 2017; Madeddu et al., 2016).

Cisplatin is also relatively rarely associated with overt cardiotoxicity. Nonetheless, when it occurs, it is associated with both acute and late-onset conditions such as angina, acute myocardial infarction, hypertension, occurring in 15% to 50% of patients, arrhythmias, myocarditis, cardiomyopathy, congestive heart failure as well as deep vein thrombosis and pulmonary embolism. Asymptomatic systolic dysfunction is also frequent (Bomzer, 2014; Han et al., 2017; Madeddu et al., 2016; Purkayastha et al., 2017).

Regarding ifosfamide-associated cardiotoxicity, like other alkylating agents, it generally presents with symptoms of heart failure secondary to left ventricular dysfunction. Partial reversible myocardial dysfunction and malignant arrhythmias can occur (Curigliano et al., 2012; Madeddu et al., 2016; Zamorano et al., 2016).

Recently, alkylating agents have also been associated with the development of veno-occlusive disease of pulmonary small vessels, frequently presenting as pulmonary hypertension or pulmonary thromboembolism (Zamorano et al., 2016).

Table 9 summarizes the principal aspects regarding the cardiotoxicity of alkylating agents discussed in this chapter, namely incidence, cardiovascular adverse effects and possible mechanisms of cardiotoxicity.

Table 9 - Cardiotoxicity of Alkylating agents. CHF, Congestive heart failure; DVT, Deep vein thrombosis; HF, Heart failure; HT, Hypertension; MI, Myocardial Ischemia; PE, Pulmonary embolism; ROS, Reactive oxygen species; VWF, Von Willebrand factor. Adapted from Adão et al., 2013; Albini et al., 2010; Angsutararux et al., 2015; Bomzer, 2014; Han et al., 2017; Madeddu et al., 2016; Moslehi, 2016; Purkayastha et al., 2017; Zamorano et al., 2016

Cardiotoxicity of Alkylating agents			
Drug examples	**Incidence**	**CV adverse effects**	**Mechanisms of cardiotoxicity**
Cyclophosphamide	7-28%	**Type I** Acute HF; Mild functional mitral regurgitation; pericardial effusion; pericarditis; myopericarditis; arrhythmias.	**- Direct endothelium injury:** leakage of metabolites toxic to cardiomyocytes. **- ROS production + oxidative stress:** metabolites toxic to cardiomyocytes; Prothrombotic state (platelet activation + increased VWF); vasospasm – [cisplatin+]
Cisplatin	≈2%	**HT** (15-50%); MI (angina, acute myocardial infarction); Arterial thrombosis; DVT; PE; arrhythmias; myocarditis; CHF.	
Ifosfamide	<10 g/m^2=0.5% 12.5-16g/m^2=17%	HF; arrhythmias.	

6. Comorbidities in chemotherapy-related cardiotoxicity

The risk of cardiac toxicity increases in patients presenting with pre-existing cardiovascular conditions such as hypertension, diabetes mellitus, liver diseases as well as previous cardiac disease (Angsutararux et al., 2015). Studies enrolling women with breast cancer and pre-existing cardiovascular comorbidities who received therapy with anthracyclines revealed that these patients presented a higher risk of developing cardiotoxicity (Subramaniam et al., 2021).

Patients with comorbidities, particularly coronary artery disease and hypertension, benefit from intensive management of pre-existing conditions before and during therapy because chemotherapy is known to possibly worsen them. For example, anthracyclines and anti-HER2 therapies are known to worsen pre-existing heart failure (Curigliano et al., 2012; Sarfati et al., 2016).

Furthermore, the presence of multiple comorbidities frequently has a negative impact on cancer therapy and prognosis because it influences patients' tolerability and response to chemotherapy. When Batra *et al.* compared patients with pre-existing comorbidities to those without previously known conditions, he concluded that non-cancer-dependent survival was shorter in the first group of patients (Batra et al., 2020; Sarfati et al., 2016).

Considering the growing need for guidance, some organizations including the ASCO, the American College of Cardiology (ACC) and the American Heart Association (AHA), have recently developed a guideline on cardiovascular disease management in cancer patients, in which it is recommend that physicians perform a thorough cardiovascular exam in every patient who receives a cancer diagnosis as well as, a screening for modifiable cardiovascular risk factors prior to starting potentially cardiotoxic treatments (Subramaniam et al., 2021).

Another important aspect refers to the fact that most drugs used in cancer therapies are usually tested for potential cardiotoxicity only in the healthy heart and despite some of them appearing relatively harmless in these settings, its cardiac toxicity can sometimes be "hidden" and only become apparent in previously injured cardiac tissue, frequently in the settings of an underlying disease state or a pre-existing comorbidity. In these circumstances, cardiac tissue already presents with a certain

degree of weakness and thus it is more prone to the direct toxic effects of some of these agents further damaging these cells which would otherwise not be sufficient to induce harm in the normal heart. Hence, this concept named “hidden toxicity” defines cardiotoxicity that only manifests in the presence of pre-existing comorbidities or in the setting of a state of disease, such as myocardial ischemia, diabetes, dyslipidemia, obesity, among others. This explains why patients who already have an asymptomatic or latent cardiac disease are more prone to drug cardiotoxicity (D. M. Cardinale et al., 2020; Ferdinandy et al., 2019; Kosalka et al., 2019).

7. Methods for monitoring cardiotoxicity

Cardiac imaging techniques are crucial to provide important information on cardiac function and structure. In people undergoing potential cardiotoxic therapies, it is important to establish a monitoring program to detect chemotherapy-induced cardiomyopathy. Most of the existing recommendations for monitoring only encompass treatments with anthracyclines and trastuzumab, as there is few guidance for other cardiotoxic agents (Stone et al., 2021).

Nowadays, there are several and some readily available methods for detection and monitoring of cardiotoxicity in people undergoing chemotherapy for breast cancer. These include cardiac imaging techniques, which allow for the detection of ventricular dysfunction evidenced by a decrease in LVEF resulting from structural damage; biomarkers; electrocardiographic studies and others (S. Gillespie et al., 2012; Stone et al., 2021; Zamorano et al., 2016).

7.1.Cardiac imaging techniques

It is recommended that cardiovascular imaging is used for screening and monitoring of cardiotoxicity before, during and after chemotherapy when potential cardiotoxic drugs are used (S. Gillespie et al., 2012; Stone et al., 2021; Zamorano et al., 2016).

Preference should be given to the same imaging technique for future assessments throughout treatment rather than switching between modalities to allow for comparison and to minimize discrepancies (S. Gillespie et al., 2012; Stone et al., 2021; Zamorano et al., 2016).

The modality chosen usually depends upon local availability and operator experience. Imaging methods with the best reproducibility, good quality image and radiation-free as well as the ones providing additional clinically relevant information should be preferred (S. Gillespie et al., 2012; Stone et al., 2021; Zamorano et al., 2016).

7.1.1. Echocardiography

Echocardiography remains the most frequently chosen imaging method for monitoring and screening of chemotherapy-induced cardiotoxicity mostly due to its good safety profile and wide availability, allowing for detection and monitoring of myocardial dysfunction, through the assessment of LVEF before, during and after chemotherapy treatments (S. Gillespie et al., 2012; Stone et al., 2021; Zamorano et al., 2016).

It has the great advantage of not using any radiation and also being useful in the identification of other complications of cancer therapy like pericardial or valvular diseases and pulmonary hypertension. Unfortunately, it can be of limited use in patients who have undergone chest radiation, had a mastectomy or are obese. When available and performed by an experienced operator, **3D echocardiography** is believed to be the best echocardiographic method for LVEF measurement providing that endocardial definition is clear and is also associated with the best reproducibility. Nevertheless, today **2D echocardiography biplane Simpson method** is more widely available, so it remains of great importance because it allows for the estimation of left ventricular volumes and ejection fraction. Any identified changes in LVEF should be confirmed by serial repeated imaging conducted 2 to 3 weeks afterward the initial study showing the decrease in LVEF to allow for the diagnosis of cardiotoxicity. The main limitation regarding 2D echocardiography is its lower reproducibility in comparison with 3D echocardiography (Stone et al., 2021; Zamorano et al., 2016).

There are many other echocardiographic techniques to consider including **contrast echocardiography**, which is useful to improve image reproducibility when there is a suboptimal delineation of the left ventricle endocardial borders; **stress echocardiography**; **doppler myocardial imaging**; and **global longitudinal myocardial strain** (GLS), which similarly to the latter can be used to accurately predict a subsequent decrease in LVEF and allow for early detection of ventricular dysfunction in this context. GLS imaging by echocardiography can detect cardiotoxicity at an asymptomatic early stage. However, despite looking promising, it still requires standardization among centers (Stone et al., 2021; Zamorano et al., 2016).

7.1.2. Cardiac magnetic resonance imaging (CMRI)

CMRI is considered the reference method in the evaluation and measurement of ventricular volumes and function mostly because of its high accuracy. Nonetheless, when it comes to its routine use in the assessment of cardiotoxicity, there is currently limited studies regarding its applicability as it is now mostly used in situations when there is a need to clarify discrepancies in the degree of fall in ventricular function, when it is borderline or if other techniques are non-diagnostic and the quality of some images are suboptimal. It is, however, strongly discouraged to access LVEF using multiple methods, as they are not interchangeable. Some also believe CMRI can have a role in the identification of small changes in LVEF during chemotherapy (Lambert & Thavendiranathan, 2016; Stone et al., 2021; Zamorano et al., 2016).

7.1.3. Nuclear cardiac imaging

As mentioned above, there are several other cardiac imaging techniques of value to detect myocardial toxicity like nuclear cardiac imaging in which positron emission tomography (**PET**)**,** single-photon emission computerized tomography (**SPECT**), multigated acquisition (**MUGA**) **scan** and other techniques are included. The latter has a good accuracy, reproducibility and it correlates well with CMRI and 3D-echocardiography imaging. However, it has the disadvantage of being associated with a clinically significant cumulative radiation exposure and only providing limited structural and hemodynamic information (Zamorano et al., 2016).

7.2.Biomarkers

Apart from imaging methods, biomarkers are of great importance as myocardial injury can also be identified by elevated values of high-sensitivity cardiac troponin (**hs-cTn**). Although they do not replace imaging techniques, cardiac troponins have proven to be important in the evaluation of cardiotoxicity if used in combination with imaging techniques, allowing for a significant increase in the prevention of adverse cardiac

events secondary to chemotherapy-related toxicity and increasing both diagnostic specificity and sensitivity (D. M. Cardinale et al., 2020).

In addition to cardiac troponin I and T, other laboratory biomarkers such as natriuretic peptides (B-type Natriuretic Peptide (**BNP**); N-Terminal-pro-B-type Natriuretic Peptide (**NT-pro-BNP**)) also play an important role at early stages when there is only minor damage, or the affected area is still small, making it difficult to detect myocardial injury even using more sophisticated cardiac imaging techniques. In these situations so called subclinical toxicity, cardiac injury may remain asymptomatic for long periods, but once it becomes manifest it carries a very poor prognosis (D. M. Cardinale et al., 2020). Recently discovered biomarkers such as soluble suppression of tumorigenicity-2, myeloperoxidase, growth differentiation factor-15, galectin-3 and endothelin-1 can also have potential to predict early cardiotoxicity but, at present time, they still lack robust evidence (W & M, 2021).

It is also well known that natriuretic peptides are very useful in detecting heart failure, but their routine role in monitoring cardiac injury related to chemotherapy is still not established (Zamorano et al., 2016).

Some limitations regarding biomarkers' use related to cardiotoxicity still remain namely, the need to define timings of screening during chemotherapy as well as the definition of the upper limit of normal for each specific biomarker, the use of different laboratory assays and which strategy to follow given the case of an abnormal result, as there is currently a lack of evidence to support decisions on whether to stop or continue chemotherapy in these situations (D. M. Cardinale et al., 2020; Zamorano et al., 2016).

Despite not being sufficient to diagnose cardiotoxicity, a newly abnormal result can predict future toxicity, thus allowing identification of patients who are at increased risk of developing cardiac dysfunction and will probably have a poorer prognosis, urging them to start preventing therapies (D. M. Cardinale et al., 2020; Zamorano et al., 2016).

Table 10 summarizes each method previously mentioned, their advantages as well as their limitations.

Table 10 - **Methods for monitoring chemotherapy-induced cardiotoxicity.** 2D, two-dimensional; 3D, three-dimensional; BNP, B-type Natriuretic Peptide; CMRI, Cardiac magnetic resonance imaging; GLS, Global longitudinal myocardial strain; HD, Hemodynamic; HF, heart failure; hs-cTn, high-sensitivity cardiac troponin; LVEF, Left ventricular ejection fraction; MUGA, Multigated Acquisition Scan; NCI, Nuclear cardiac Imaging; NT-pro-BNP, N-Terminal-pro-B-type Natriuretic Peptide; PET, Positron Emission Tomography; SPECT, Single-Photon Emission Computerized Tomography. Adapted from (Adão et al., 2013; D. M. Cardinale et al., 2020; Lambert & Thavendiranathan, 2016; S. Gillespie et al., 2012; Stone et al., 2021; Zamorano et al., 2016)

Methods for monitoring chemotherapy-induced cardiotoxicity			
Technique		**Advantages**	**Limitations**
Cardiac imaging	**Echocardiography:** -3D -2D Simpson's -Stress - with Contrast -with Doppler -GLS	- Safe profile, Radiation free. - Wide availability. - Assessment cancer therapy-related complications, other than LVEF dysfunction.	- Operator-dependent (intra- and inter-observer variability). Limited reproducibility. - Image quality. - Obese patients, previous chest radiation or mastectomy. - **GLS:** Lack standardization among centers.
	CMRI	- High accuracy and reproducibility: ventricular volumes; small changes in LVEF (useful when other techniques are non-diagnostic). - Safe profile, Radiation free. - Detection of myocardial fibrosis.	- Limited availability. - Patient's related: long acquisition time, claustrophobia.
	NCI: -PET -SPECT -MUGA scan	- Good accuracy and reproducibility. (**MUGA**+) - Good correlation with CMRI and 3D-echocardiography. (**MUGA**+)	- Radiation exposure. - Only limited structural and HD information
Cardiac biomarkers	- hs-cTn - Troponin I	In Complement to imaging techniques: - Accuracy. - Reproducibility. - Non-invasive. - Early identification of subclinical toxicity and patients at increased risk. - **Natriuretic peptides**:identification of HF + monitoring (?)	- Lack of evidence to establish clinical significance of abnormal values. - Role for routine screening unclear and timings not established. - Lack of concise definition of the upper limit of normal and variations with different laboratory assays.
	Natriuretic peptides -BNP -NT-pro-BNP		
Electrocardiography		- Non-invasive - Low cost - Useful for arrhythmia screening, repolarization changes among others.	- Limited use as a marker of cardiotoxicity - Lack of LVEF assessment. - Intra- and inter-observer variability

8. Prevention of chemotherapy-induced cardiotoxicity

As already mentioned, and according to the ESMO, cancer patients with pre-existing cardiovascular disease or known risk factors have an increased risk of developing cardiac toxicity related to chemotherapy. Thus, the development and adoption of preventive measures are of great importance in these patients (Jurczyk et al., 2021).

The first step in which one can intervene to try to prevent or minimize the risk of cardiac toxicity related to many cancer therapies is to identify patients who are at greater risk before initiating chemotherapy. Although, there are no robust guidelines for risk assessment at the present, there is a certain consensus among several entities that screening and treatment of cardiovascular risk factors such as hypertension, diabetes, dyslipidemia, obesity and smoking should be always performed and complemented by a detailed clinical history, physical examination as well as baseline assessment of cardiac function through echocardiogram imaging. Additionally, measurements of biomarkers can be considered, though controversial (Jurczyk et al., 2021; Lambert & Thavendiranathan, 2016; Mitchell & Lenihan, 2020).

This initial clinical and functional cardiac assessment is of great importance because it enables the detection of pre-treatment subclinical cardiac dysfunction, which may influence future decisions concerning the chemotherapy chosen, the eventual need for cardioprotection and increase the frequency of surveillance exams.

Furthermore, this initial assessment of cardiac function grants comparison material that can later be used to identify eventual changes that could develop during therapy enabling proper interpretation and management of complications. It is also important to emphasize the importance of close cooperation between the medical specialties involved in this field, such as cardiologists and oncologists (Jurczyk et al., 2021; Lambert & Thavendiranathan, 2016; Mitchell & Lenihan, 2020).

It is also important to consider that the risk of cardiotoxicity during anticancer treatment is also influenced by other clinical factors such as prior anthracycline treatment, age older than 75 years, prior chest radiotherapy, elevated cardiac biomarkers before initiation of therapy and baseline systolic left ventricular dysfunction as well as factors referred to the chemotherapy regimen itself, such as the route of drug

administration, cumulative dose and combination of multiple agents (Angsutararux et al., 2015; Curigliano et al., 2020).

Some general preventive strategies believed to reduce the risk of cardiotoxicity include avoidance of potentially cardiotoxic therapies if equivalent alternatives exist, without compromising cancer prognosis. Regardless of this fact, if a potentially cardiotoxic drug is administered patients should be actively screened for the cardiovascular risk factors mentioned above. If mediastinal radiotherapy is necessary, it is recommended to select lower radiation doses and reduce radiation fields to the minimum required, when possible (Koutsoukis et al., 2018; Mitchell & Lenihan, 2020).

Non-pharmacological therapies that prevent the incidence of cardiac toxicity should be strongly recommended and include lifestyle changes to reduce cardiovascular risk of these patients. Such measures include blood pressure control, lower cholesterol levels as well as adopting an healthy diet including low fat and sodium restriction, smoking cessation, weight loss and frequent physical exercising, mainly aerobic exercise (Avila, M. S., Siqueira, S., Ferreira, S., & Bocchi, 2019; Curigliano et al., 2020). The latter carries important prognostic value regarding survival in cancer patients and it is associated with a substantial decrease in the incidence of cardiovascular side effects in non-metastatic breast cancer patients (Jones et al., 2016). According to Radonjic *et al.*, a prospective study comparing non-metastatic breast cancer patients who exercised several hours daily with those presenting with limited physical activity, the first group of patients had a 23% reduction in the risk of cardiac adverse effects compared to the sedentary patient group (Radonjic et al., 2020).

However, according to Zamorano *et al.* the preventive impact on LV function of regular moderate aerobic exercise is more evident in patients receiving therapy with anthracyclines, whereas it has not shown a reduction in the incidence of trastuzumab-induced cardiotoxicity (Zamorano et al., 2016).

Other measures include avoidance of drugs that prolong QT interval and antihistamines, minimization of the risk of radiation exposure and the treatment of comorbidities (Cruz et al., 2015).

Regarding the use of prophylactic cardioprotective medication in preventing LV dysfunction, further evidence is needed concerning the efficacy of these preventive therapies, except for dexrazoxane, an intracellular iron-chelating agent, which is the

only Food and drug administration (FDA) and European medicines agency (EMA) approved medication for cardioprotection use in patients with advanced or metastatic breast cancer which works by reducing the production of free radicals, therefore preventing the decrease in LV function particularly that caused by cumulative dose of >300 mg/m^2 of doxorubicin or >540 mg/m^2 of epirubicin (Zamorano et al., 2016).

However, despite the current lack of formal recommendations recent studies have demonstrated that particularly some groups of patients may benefit from these cardioprotective therapies as well as patients undergoing therapy with high-risk cardiotoxic chemotherapy regimens, mainly because follow-up studies demonstrated that cardioprotective drugs prevented a decrease in LVEF, within 6-month. Thus, clinicians should consider the administration of such therapies in selected patients (Curigliano et al., 2020; Koutsoukis et al., 2018; Mitchell & Lenihan, 2020; Zamorano et al., 2016).

When choosing which cardioprotective agent to use, some studies concluded that angiotensin-converting enzyme inhibitors, angiotensin receptor blockers and/or beta-blockers such as carvedilol and nebivolol are the drugs preferred. Regarding Beta-blockers, both nebivolol and carvedilol have been studied for the prevention and treatment of cardiotoxicity (Curigliano et al., 2020; Koutsoukis et al., 2018; Mitchell & Lenihan, 2020; Zamorano et al., 2016).

As mentioned, apart from optimization of risk factors control, initial screening and baseline assessment, prophylactic cardioprotective agents can potentially be indicated for prevention of cardiac toxicity, in particularly situations, including scenarios in which there is a high baseline cardiotoxicity risk due to causes such as preexisting cardiovascular conditions and/or poorly controlled cardiovascular risk factors or previous therapy with anthracyclines. In patients presenting with an elevation in troponin levels, preventive pharmacological therapy may be considered, especially in therapies using high doses of anthracyclines. Alternatively, patients presenting with low basal risk but who will undergo therapy with high total cumulative anthracycline doses (>250–300 mg/m^2 doxorubicin or equivalent) are also believed to benefit from preventive treatment with angiotensin-converting enzyme inhibitors (ACEis) or angiotensin receptor blockers (ARBs) and/or beta-blockers. Patients already presenting with pre-existing heart failure or LV dysfunction at baseline assessment, should be

referenced to a cardiologist. Given the higher risk of cardiotoxicity in this group, further options to reduce cardiac toxicity include the selection of less cardiotoxic chemotherapy agents when possible, switching from bolus to intravenous continuous infusions, eventual dose reduction or selection of preparations with lower toxicity such as liposomal formulation of doxorubicin as well as additional cardioprotective therapy, mentioned above (Curigliano et al., 2020; Mitchell & Lenihan, 2020; Shiga & Hiraide, 2020; Zamorano et al., 2016).

Regarding the example of anthracycline-induced cardiotoxicity, despite the lack of proper guidelines some measures have been proposed to try to reduce its potential for cardiac toxicity such as close monitoring, reduction in the cumulative dose to levels lower than 450 mg/m^2, use of continuous infusions up to 96 h, preference for the use of analogues like epirubicin and idarubicin with lower toxicity or liposomal doxorubicin formulations as well as, the use of cardioprotective medications such as dexrazoxane previously mentioned above (Adão et al., 2013; Keefe, 2002; Zamorano et al., 2016).

As referred previously taxanes, particularly paclitaxel, interfere with the metabolism of anthracyclines and reduce its elimination, resulting in higher plasma levels further enhancing its cardiotoxicity. Therefore, to risk reduction it is recommended the administration of anthracyclines before paclitaxel or to limit its dosage to 360 mg/m^2 (Zamorano et al., 2016).

According to Alexandre et al. in patients presenting with a LVEF lower than 40%, anthracyclines should be contraindicated and preference must be given to alternative chemotherapeutic agents, unless there is no effective alternative therapy. For LVEF higher than 40% but less than 50% as well as in patients with cardiovascular risk factors who are exposed to multiple cardiotoxic therapies regardless of their cardiac function, anthracyclines may be administered but should be combined with cardioprotective agents such as ACEis or ARBs and/or β-blockers (Alexandre et al., 2020).

Alternatively, the prophylactic use of such cardioprotective drugs in patients with cardiovascular risk factors and/or increase in cardiac biomarkers but presenting with a normal LVEF remains controversial and further studies are needed (Fiúza, 2009).

Since these agents' cardiac toxicity impair key antioxidant mechanisms, drugs such as antioxidants, iron-chelating agents and lipid-lowering drugs have been tested but their cardioprotective effects are still uncertain and further studies are needed to

clarify this topic. Thus, administration of mitochondrially targeted antioxidants aimed to increase antioxidant defense systems and to neutralize the mitochondrial reactive oxygen species/reactive nitrogen species (ROS/RNS) have proven cardioprotective in animal models without interfering with the efficacy of cancer treatment. Furthermore, carvedilol appears to have antioxidant properties as studies demonstrated that it could prevent cardiac toxicity induced by doxorubicin (Adão et al., 2013; Curigliano et al., 2020; Ferdinandy et al., 2019).

It has also been suggested that PARP inhibitors can also reduce the incidence of cardiotoxicity without interfering with cancer therapy, when combined with doxorubicin or cisplatin (Adão et al., 2013; Curigliano et al., 2020; Ferdinandy et al., 2019). As already discussed, dexrazoxane may have cardioprotective effects, possibly through the reduction of superoxide radicals produced by anthracyclines. Unfortunately, its use is limited due to its interference with the therapeutic efficacy of anthracyclines as well as an increase in secondary tumors (Adão et al., 2013; Curigliano et al., 2020; Koutsoukis et al., 2018).

Strategies to reduce the cardiotoxicity associated with trastuzumab include avoidance of the concomitant therapeutic association of trastuzumab and anthracyclines. Regardless of this fact, the cardiotoxic risk of the latter association can be reduced if a drug-free interval between the administration of both drugs is implemented (Fiúza, 2009; Zamorano et al., 2016). Studies suggest that prophylactic therapy with beta-blockers before initiating chemotherapy with trastuzumab reduces the risk of developing heart failure in patients with breast cancer and normal LVEF. Trials analyzing the potential preventive role of candesartan, lisinopril–carvedilol and perindopril–bisoprolol combinations in decreasing the incidence of trastuzumab-induced cardiotoxicity are currently being developed (Zamorano et al., 2016). Keefe *et al.* suggested that identification of patients with a higher predisposition to toxicity of trastuzumab could be achieved through administration of a tracer dose of radiolabeled trastuzumab and subsequent screen of cardiac uptake using scintigraphy (Keefe, 2002).

Although not consensual, in cases concerning patients previously treated with fluoropyrimidines who develop cardiotoxic adverse effects mainly coronary spasms, re-exposure with the same agent is associated with a 90% risk of recurrence and thus it is not recommended for patients with a previous history of fluoropyrimidines toxicity. On

the other hand, there is not strong evidence that the use of coronary dilators such as nitrate and calcium antagonists reduce the risk of fluoropyrimidines induced cardiotoxicity. However, when re-exposure to fluoropyrimidines is mandatory, administration of nitrates and calcium antagonists may be considered after careful evaluation of the patient's risk and benefit (Shiga & Hiraide, 2020).

Furthermore, recent studies testing probucol, a drug with antioxidant and lipid lowering properties, demonstrated a potential cardioprotective effect during therapy with 5-FU (Jurczyk et al., 2021).

Despite the recommendations and scientific evidence, there are gaps in the current knowledge about the use of prophylactic cardioprotective medications and more research is needed to define beneficial interventions to prevent cardiovascular disease in breast cancer patients.

9. Treatment of cardiotoxicity induced by chemotherapy

In patients manifesting with anthracycline-induced-cardiotoxicity, either in the form of symptomatic heart failure or an asymptomatic decrease in LVEF of >10% from baseline or to <50%, it may be necessary to interrupt chemotherapy and recommended to start cardioprotection with the administration of beta-blockers combined with an angiotensin converting enzyme inhibitor or, in alternative, an angiotensin receptor blocker in order to prevent further deterioration of cardiac function or progression to symptomatic heart failure (Pardo Sanz & Zamorano, 2020; Zamorano et al., 2016). Thus, treatment of cardiac toxicity arising from cancer therapies resembles the standard therapy for other forms of heart failure and it should be initiated regardless of the presence of symptoms (Curigliano et al., 2012; Fiúza, 2009).

Cardioprotective therapy combining both beta-blockers and ACE inhibitors has shown higher efficacy than monotherapy alone, when administered in patients who present with anthracycline-induced cardiotoxicity (Zamorano et al., 2016).

Unlike anthracyclines, in the case of trastuzumab-induced cardiotoxicity, there are no available trials that prove the therapeutic efficacy of administering heart failure drugs. However, despite this lack of strong evidence, it is believed that ventricular dysfunction induced by trastuzumab would likely improve with therapy using ACE inhibitors and beta-blockers. If after suspending chemotherapy and starting cardioprotection the LVEF improves to >49%, therapy with trastuzumab may be reinitiated. Nevertheless, if LVEF continues to decrease regardless of cardioprotective therapy, patients should be referred to cardio-oncology (Zamorano et al., 2016).

In fluoropyrimidine-induced cardiotoxicity, it is recommended to stop cancer therapy with the causal agent and to initiate therapy with nitrates, beta-blockers, and calcium channel blockers which are believed to reduce coronary spasm and thus improve ischemic symptoms and angina (Jurczyk et al., 2021; Shiga & Hiraide, 2020). Recently, some studies suggested that therapy with Glucagon-like peptide-1 (GLP-1) analogs may be helpful in treating fluoropyrimidine-related cardiovascular

toxicity by antagonizing the negative effect that drugs such as 5-FU have in the expression levels of endothelial nitric oxide synthase (Shiga & Hiraide, 2020).

10. Future perspectives in Cardio-Oncology

Clinical trials in the Cardiology field do not usually tend to include oncological patients in the populations enrolled. Often the reason behind the under-representation of this group of patients relates to the fact that, historically, they are frequently associated with poorer prognosis. Nevertheless, the prognosis of oncological patients has been improving throughout the years, mostly due to advances in scientific knowledge and better cancer therapies available (Trapani et al., 2020). Therefore, the number of long-term cancer survivors is improving but, the development of long-term chemotherapy-induced adverse effects, particularly cardiac toxicity and associated cardiovascular comorbidities are increasing simultaneously.

Recently, to better assess these patients a new field combining both oncology and cardiology was developed and thus, cardio-oncology emerged aiming to provide better treatment for cardiovascular side effects of cancer therapies (Campia et al., 2019; Zamorano et al., 2016). The scope of cardio-oncology is wide, including prevention, diagnosis and monitoring of cardiac toxicity as well as the will to develop new anticancer therapies with less cardiovascular impact. However, because of being a recent discipline currently under development, cardio-oncology still presents many unmet needs and gaps in knowledge to guide best practices in clinical settings. Of these, guidelines on prevention, diagnosis or treatment allowing for optimization of clinical practice and standardization are still lacking and there is a major need for more validated data to allow for better outcomes by reducing the development of cardiovascular complications or exacerbation of pre-existing cardiac conditions during or after cancer therapies in patients at risk (Curigliano et al., 2020; Zamorano et al., 2016).

Given this increasing need for guidance, sometimes clinicians are faced with uncertainty regarding what clinical decision would be the best given the lack of evidence on many matters, for example, whether to maintain or interrupt cancer therapy because of the risk of cardiotoxicity although chemotherapy might be lifesaving, among others. This calls for a clearer definition of cardiotoxicity itself, better selection of the best methods to be used for its detection, therefore improving sensitivity and early detection, as well as further improvements on preventive strategies to reduce the risk of cardiac toxicity (Curigliano et al., 2020; Zamorano et al., 2016).

The ASCO also emphasized the increasing need for guidance with a particular focus on the assessment of cardiac dysfunction either symptomatic or subclinical and the risk factors influencing its incidence. Furthermore, compared to diastolic dysfunction, systolic dysfunction is far more assed in studies and thus, more data on the former is also needed (Mitchell & Lenihan, 2020).

Future perspectives in this field are diverse but they share the common aim of developing a more personalized and precise medicine enabling better management and treatment of the cardiac adverse effects of therapies in oncological patients (Curigliano et al., 2012). To allow for a better understanding of cardiovascular toxicities associated with known cardiotoxic drugs of both patients and clinicians, some entities are looking forward to developing educational resources such as free access online platforms as well as designing clinical trials to identify the most effective treatments in cases of cardiovascular toxicity (Alexandre et al., 2020; Campia et al., 2019).

The future of Cardio-Oncology and precision medicine face several challenges, one of which starts with the need of a universally structured definition of cardiotoxicity itself as referred above, as well as, development of better diagnostic methods and defined criteria particularly, the investment in more validated approaches regarding early cardiac toxicity detection, given that the current diagnosis techniques of cardiotoxicity present limited sensitivity by only assessing left ventricular ejection fraction or due to insufficient data to attest reliability in the case of cardiovascular biomarkers assessment (Chung et al., 2018; Zamorano et al., 2016).

Biomarkers' evaluation has the potential to identify early cardiotoxicity, preventing progressive and sometimes irreversible damage of cardiac cells as well as optimize cancer therapy and risk stratification of patients. Recently advances in areas, such as mass spectrometry, have led to important cardiovascular biomarker discoveries through proteomic profiling that is being applied in cardio-oncology studies, like the one that identified immunoglobin E as a marker of doxorubicin and trastuzumab cardiotoxicity (Zaha et al., 2021).

Similarly to the latter, metabolomic profiling has enabled the detection of mitochondrial dysfunction through the detection of small-molecule metabolites that reflect cell activity and thus, can potentially be used as biomarkers of cardiac dysfunction (Zaha et al., 2021). Further investigation challenges regard RNA molecules,

such as circular and microRNAs, and focus on its role on gene expression and epigenetics, amongst other processes, which could also serve as potential biomarkers (Zaha et al., 2021).

Another important question that needs to be answered in the future concerns preventive strategies particularly who would benefit from primary prevention, apart from populations with high cardiovascular risk factors or who are receiving therapies associated with higher-risk of cardiotoxicity. The data available on this topic is limited and thus, it is uncertain whether "universalization" of primary prevention would be cost effective and justified inclusion of patients with low-risk for cardiac toxicity. In contrast, secondary prevention presents more defined indications in guidelines. However, there is still some evidence lacking further elucidation as well as a need to deepen the knowledge of the underlying pathophysiology to determine patients at increased risk and develop a more personalized approach (Campia et al., 2019; Zamorano et al., 2016).

Development of strategies to better stratify patients at risk for cardiotoxicity is another important challenge. To achieve this goal, it is needed to review cardiovascular risk factors and their influence on the incidence of cardiotoxicity, develop better strategies to detect subclinical toxicity and establish associated clinical manifestations and prognosis (Campia et al., 2019; Zamorano et al., 2016).

Another challenge concerns the disparity in clinical research between frequent cardiotoxic agents such as anthracyclines and lesser-known classes also associated with cardiac toxicity, though more uncommon. Hence, further investigation addressing its mechanisms of cardiotoxicity and clinical manifestations are required (Trapani et al., 2020).

Another field that is starting to be a focus of growing research on Cardio-Oncology is the influence of genetics on the risk of developing cardiotoxicity. Oncological patients with the same diagnosis who are receiving the same therapy frequently present different susceptibility to cardiotoxicity that can only be partially explained by traditional factors, suggesting a possible underlying genetic factor. Given this heterogeneity, genome sequencing can be used to identify mutations that confer increased susceptibility to cancer therapies and thus support risk stratification before treatment and assist on lesser cardiotoxic chemotherapeutic schemes selection. An example of gene mutations thought to be associated with the induction of

chemotherapy-induced cardiotoxicity is the identification of a mutated Titin gene. Furthermore, future research is needed to develop molecular approaches to better understand each patient's susceptibility (Alexandre et al., 2020; Campia et al., 2019; Kubota et al., 2021; Zaha et al., 2021).

Finally, it is also important to define standard protocols for long-term cardiac monitoring in survivors (Alexandre et al., 2020).

11. Conclusion

Nowadays due to the development of novel and better therapies, most patients diagnosed with breast cancer are expected to live longer and concomitantly cardiac complications following chemotherapy are becoming more prevalent, so further attention must be given to its management including prevention, early detection, monitoring and treatment.

Addressing chemotherapy-induced cardiotoxicity has recently started to be seen as a high priority in the field of cardio-oncology, since cardiotoxicity related to some chemotherapeutic agents used in breast cancer therapies is frequent and impacts prognosis, either due to the need to suspend cancer therapy consequently reducing its efficacy or by leading to the developing of usually severe cardiac complications weeks, months or even years after the end of cancer treatments which shortens life-time expectancy and increases comorbidities in these patients.

Of all chemotherapeutic agents used in breast cancer, anthracyclines are by far the most frequent class associated with cardiac toxicity mostly through the induction of usually asymptomatic progressive ventricular dysfunction that eventually ends up leading to symptomatic heart failure. Apart from cardiac dysfunction, other cardiac adverse effects are associated with chemotherapeutic agents used in breast cancer and some of these include arrhythmias, myocardial ischemia, myopericarditis, among others. Some agents, like trastuzumab generally induce transient cardiac dysfunction, which is referred to as type II cardiotoxicity, whereas others like doxorubicin induce permanent damages, therefore associated with type I cardiotoxicity. Nevertheless, the later classification is starting to fall into disuse.

Unfortunately, evidence and clarification of the mechanisms responsible for cardiotoxicity are still lacking and further studies are needed to allow for better management of the cardiovascular complications of cancer therapies in these patients.

Nowadays protocolized guidelines regarding the management of cardiovascular complications of cancer therapies are still scarce and clinicians are frequently taking decisions based on clinical consensus rather than evidence. Due to these lack of studies and evidence-based orientations to guide decisions on to whether suspend or continue cancer therapies in the setting of increased cardiotoxicity, it could carry a non-

neglectable impact on prognosis. Hence, firstly a clear and universally accepted definition of cardiotoxicity is needed as well as further evidence to support more informed decisions on when to initiate therapy, early diagnosis or, which patients would benefit from prophylactic measures. Further investigation of less cardiotoxic chemotherapeutic agents as well as a better definition of its pathophysiological mechanism are also some of the many challenges in the field of cardio-oncology for the future to allow for better outcomes in these patients.

References

Adão, R., De Keulenaer, G., Leite-Moreira, A., & Braś-Silva, C. (2013). Cardiotoxicity associated with cancer therapy: Pathophysiology and prevention strategies. *Revista Portuguesa de Cardiologia*, *32*(5), 395–409. https://www.revportcardiol.org/en-pdf-S2174204913000895

Advani, P. P., Ballman, K. V., Dockter, T. J., Colon-Otero, G., & Perez, E. A. (2016). Long-term cardiac safety analysis of NCCTG N9831 (Alliance) adjuvant trastuzumab trial. *Journal of Clinical Oncology*, *34*(6), 581–587. https://doi.org/10.1200/JCO.2015.61.8413

Albini, A., Pennesi, G., Donatelli, F., Cammarota, R., De Flora, S., & Noonan, D. M. (2010). Cardiotoxicity of anticancer drugs: The need for cardio-oncology and cardio-oncological prevention. *Journal of the National Cancer Institute*, *102*(1), 14–25. https://doi.org/10.1093/jnci/djp440

Alexandre, J., Cautela, J., Ederhy, S., Damaj, G. L., Salem, J. E., Barlesi, F., Farnault, L., Charbonnier, A., Mirabel, M., Champiat, S., Cohen-Solal, A., Cohen, A., Dolladille, C., & Thuny, F. (2020). Cardiovascular toxicity related to cancer treatment: A pragmatic approach to the american and european cardio-oncology guidelines. *Journal of the American Heart Association*, *9*(18). https://doi.org/10.1161/JAHA.120.018403

American Cancer Society. (2019). Breast Cancer: Treating Breast Cancer. *American Cancer Society*, 1–120. https://www.cancer.org/cancer/breast-cancer/treatment.html

Angsutararux, P., Luanpitpong, S., & Issaragrisil, S. (2015). Chemotherapy-Induced Cardiotoxicity: Overview of the Roles of Oxidative Stress. *Oxidative Medicine and Cellular Longevity*, *2015*. https://doi.org/10.1155/2015/795602

Avila, M. S., Siqueira, S., Ferreira, S., & Bocchi, E. A. (2019). Prevention and Treatment of Chemotherapy-Induced Cardiotoxicity. *Methodist DeBakey Cardiovascular Journal*, *15*(4), 267–273. https://doi.org/10.1016/s1098-3597(00)90004-8

Batra, A., Sheka, D., Kong, S., & Cheung, W. Y. (2020). Impact of pre-existing cardiovascular disease on treatment patterns and survival outcomes in patients with lung cancer. *BMC Cancer*, *20*(1), 1–16. https://doi.org/10.1186/s12885-020-

07487-9

Beretta, G. L., & Zunino, F. (2007). Molecularmechanisms of anthracycline activity. *Topics in Current Chemistry*, *283*(October 2007), 1–19. https://doi.org/10.1007/128_2007_3

Bomzer, C. A. (2014). Cardiovascular Toxicity of Common Chemotherapy Drugs Used to Treat Breast Cancer: An Overview. *Journal of Patient-Centered Research and Reviews*, *1*(3), 133–136. https://doi.org/10.17294/2330-0698.1027

Campia, U., Moslehi, J. J., Amiri-Kordestani, L., Barac, A., Beckman, J. A., Chism, D. D., Cohen, P., Groarke, J. D., Herrmann, J., Reilly, C. M., & Weintraub, N. L. (2019). Cardio-Oncology: Vascular and Metabolic Perspectives: A Scientific Statement From the American Heart Association. *Circulation*, *139*(13), E579–E602. https://doi.org/10.1161/CIR.0000000000000641

Cardinale, D., Iacopo, F., & Cipolla, C. M. (2020). Cardiotoxicity of Anthracyclines. *Frontiers in Cardiovascular Medicine*, *7*(March), 3–5. https://doi.org/10.3389/fcvm.2020.00026

Cardinale, D. M., Zaninotto, M., Cipolla, C. M., Passino, C., Plebani, M., & Clerico, A. (2020). Cardiotoxic effects and myocardial injury: The search for a more precise definition of drug cardiotoxicity. *Clinical Chemistry and Laboratory Medicine*, *59*(1), 51–57. https://doi.org/10.1515/cclm-2020-0566

Chaulin, A. M., Abashina, O. E., & Duplyakov, D. V. (2020). Pathophysiological mechanisms of cardiotoxicity in chemotherapeutic agents. *Russian Open Medical Journal*, *9*(3), 1–9. https://doi.org/10.15275/rusomj.2020.0305

Chien, K. R. (2006). Herceptin and the Heart — A Molecular Modifier of Cardiac Failure. *New England Journal of Medicine*, *354*(8), 789–790. https://doi.org/10.1056/nejmp058315

Cho, H., Lee, S., Sim, S. H., Park, I. H., Lee, K. S., Kwak, M. H., & Kim, H. J. (2020). Cumulative incidence of chemotherapy-induced cardiotoxicity during a 2-year follow-up period in breast cancer patients. *Breast Cancer Research and Treatment*, *182*(2), 333–343. https://doi.org/10.1007/s10549-020-05703-5

Chung, R., Ghosh, A. K., & Banerjee, A. (2018). Cardiotoxicity: Precision medicine with imprecise definitions. *Open Heart*, *5*(2), 2–5. https://doi.org/10.1136/openhrt-2018-000774

Cote, G. M., Sawyer, D. B., & Chabner, B. A. (2012). ERBB2 Inhibition and Heart Failure. *New England Journal of Medicine*, *367*(22), 2150–2153. https://doi.org/10.1056/nejmcibr1203156

Cruz, M., Duarte-Rodrigues, J., & Campelo, M. (2015). Cardiotoxicity in anthracycline therapy: Prevention strategies. *Revista Portuguesa de Cardiologia*, *35*(6), 359–371. https://doi.org/10.1016/j.repc.2015.12.004

Curigliano, G., Cardinale, D., Suter, T., Plataniotis, G., De azambuja, E., Sandri, M. T., Criscitiello, C., Goldhirsch, A., Cipolla, C., & Roila, F. (2012). Cardiovascular toxicity induced by chemotherapy, targeted agents and radiotherapy: ESMO clinical practice guidelines. *Annals of Oncology*, *23*(SUPPL. 7). https://doi.org/10.1093/annonc/mds293

Curigliano, G., Lenihan, D., Fradley, M., Ganatra, S., Barac, A., Blaes, A., Herrmann, J., Porter, C., Lyon, A. R., Lancellotti, P., Patel, A., DeCara, J., Mitchell, J., Harrison, E., Moslehi, J., Witteles, R., Calabro, M. G., Orecchia, R., de Azambuja, E., … Jordan, K. (2020). Management of cardiac disease in cancer patients throughout oncological treatment: ESMO consensus recommendations. *Annals of Oncology*, *31*(2), 171–190. https://doi.org/10.1016/j.annonc.2019.10.023

Dawood, S., Broglio, K., Buzdar, A. U., Hortobagyi, G. N., & Giordano, S. H. (2010). Prognosis of women with metastatic breast cancer by HER2 status and trastuzumab treatment: An institutional-based review. *Journal of Clinical Oncology*, *28*(1), 92–98. https://doi.org/10.1200/JCO.2008.19.9844

De Azambuja, E., Bedard, P. L., Suter, T., & Piccart-Gebhart, M. (2009). Cardiac toxicity with anti-HER-2 therapies-what have we learned so far? *Targeted Oncology*, *4*(2), 77–88. https://doi.org/10.1007/s11523-009-0112-2

De Azambuja, E., Procter, M. J., Van Veldhuisen, D. J., Agbor-Tarh, D., Metzger-Filho, O., Steinseifer, J., Untch, M., Smith, I. E., Gianni, L., Baselga, J., Jackisch, C., Cameron, D. A., Bell, R., Leyland-Jones, B., Dowsett, M., Gelber, R. D., Piccart-Gebhart, M. J., & Suter, T. M. (2014). Trastuzumab-associated cardiac events at 8 years of median follow-up in the herceptin adjuvant trial (BIG 1-01). *Journal of Clinical Oncology*, *32*(20), 2159–2165. https://doi.org/10.1200/JCO.2013.53.9288

Fanale, D., Bronte, G., Passiglia, F., Calò, V., Castiglia, M., Di Piazza, F., Barraco, N., Cangemi, A., Catarella, M. T., Insalaco, L., Listì, A., Maragliano, R., Massihnia, D.,

Perez, A., Toia, F., Cicero, G., & Bazan, V. (2015). Stabilizing versus destabilizing the microtubules: A double-edge sword for an effective cancer treatment option? *Analytical Cellular Pathology*, *2015*(Figure 1). https://doi.org/10.1155/2015/690916

Ferdinandy, P., Baczkó, I., Bencsik, P., Giricz, Z., Görbe, A., Pacher, P., Varga, Z. V., Varró, A., & Schulz, R. (2019). Definition of hidden drug cardiotoxicity: Paradigm change in cardiac safety testing and its clinical implications. *European Heart Journal*, *40*(22), 1771-1777C. https://doi.org/10.1093/eurheartj/ehy365

Fiúza, M. (2009). Cardiotoxicity associated with trastuzumab treatment of HER2+ breast cancer. *Advances in Therapy*, *26*(SUPPL.1), 9–17. https://doi.org/10.1007/s12325-009-0048-z

Goldhirsch, A., Gelber, R. D., Piccart-Gebhart, M. J., De Azambuja, E., Procter, M., Suter, T. M., Jackisch, C., Cameron, D., Weber, H. A., Heinzmann, D., Lago, L. D., McFadden, E., Dowsett, M., Untch, M., Gianni, L., Bell, R., Köhne, C. H., Vindevoghel, A., Andersson, M., … Baselga, J. (2013). 2 years versus 1 year of adjuvant trastuzumab for HER2-positive breast cancer (HERA): An open-label, randomised controlled trial. *The Lancet*, *382*(9897), 1021–1028. https://doi.org/10.1016/S0140-6736(13)61094-6

Guarneri, V., Lenihan, D. J., Valero, V., Durand, J. B., Broglio, K., Hess, K. R., Michaud, L. B., Gonzalez-Angulo, A. M., Hortobagyi, G. N., & Esteva, F. J. (2006). Long-term cardiac tolerability of trastuzumab in metastatic breast cancer: The M.D. Anderson Cancer Center experience. *Journal of Clinical Oncology*, *24*(25), 4107–4115. https://doi.org/10.1200/JCO.2005.04.9551

Han, X., Zhou, Y., & Liu, W. (2017). Precision cardio-oncology: Understanding the cardiotoxicity of cancer therapy. *Nature Communications*, *1*(1), 1–10. https://doi.org/10.1038/s41698-017-0034-x

Jones, L. W., Habel, L. A., Weltzien, E., Castillo, A., Gupta, D., Kroenke, C. H., Kwan, M. L., Quesenberry, C. P., Scott, J., Sternfeld, B., Yu, A., Kushi, L. H., & Caan, B. J. (2016). Exercise and Risk of cardiovascular events in women with nonmetastatic breast cancer. *Journal of Clinical Oncology*, *34*(23), 2743–2749. https://doi.org/10.1200/JCO.2015.65.6603

Joshi, A. M., Prousi, G. S., Bianco, C., Malla, M., Guha, A., Shah, M., Brown, S. A., & Patel,

B. (2021). Microtubule Inhibitors and Cardiotoxicity. *Current Oncology Reports*, *23*(3). https://doi.org/10.1007/s11912-021-01014-0

Jurczyk, M., Król, M., Midro, A., Kurnik-łucka, M., Poniatowski, A., & Gil, K. (2021). Cardiotoxicity of fluoropyrimidines: Epidemiology, mechanisms, diagnosis, and management. *Journal of Clinical Medicine*, *10*(19). https://doi.org/10.3390/jcm10194426

Keefe, D. L. (2002). Trastuzumab-associated cardiotoxicity. *Cancer*, *95*(7), 1592–1600. https://doi.org/10.1002/cncr.10854

Keulenaer, G. De, & Leite-moreira, A. (2013). *Cardiotoxicity associated with cancer therapy: Pathophysiology and prevention*. *32*(5).

Khouri, M. G., Douglas, P. S., Mackey, J. R., Martin, M., Scott, J. M., Scherrer-crosbie, M., & Jones, L. W. (2013). Cancer : Addressing the Unresolved Issues. *Circulation*, *126*(23), 2749–2763. https://doi.org/10.1161/CIRCULATIONAHA.112.100560.Cancer

Kosalka, P., Johnson, C., Turek, M., Sulpher, J., Law, A., Botros, J., Dent, S., & Aseyev, O. (2019). Effect of obesity, dyslipidemia, and diabetes on trastuzumab-related cardiotoxicity in breast cancer. *Current Oncology*, *26*(3), e314–e321. https://doi.org/10.3747/co.26.4823

Kostakou, P. M., Kouris, N. T., Kostopoulos, V. S., Damaskos, D. S., & Olympios, C. D. (2019). Cardio-oncology: a new and developing sector of research and therapy in the field of cardiology. *Heart Failure Reviews*, *24*(1), 91–100. https://doi.org/10.1007/s10741-018-9731-y

Koutsoukis, A., Ntalianis, A., Repasos, E., & Kastritis, E. (2018). *Management and Comorbidities Cardio-oncology : A Focus on Cardiotoxicity Chemotherapy-associated Cardiotoxicity*. 64–69.

Kubota, S., Hara, H., & Hiroi, Y. (2021). Current status and future perspectives of onco-cardiology: Importance of early detection and intervention for cardiotoxicity, and cardiovascular complication of novel cancer treatment. *Global Health & Medicine*, *3*(4), 214–225. https://doi.org/10.35772/ghm.2021.01024

Lakhani, H. V., Pillai, S. S., Zehra, M., Dao, B., Tirona, M. T., Thompson, E., & Sodhi, K. (2021). Detecting early onset of anthracyclines-induced cardiotoxicity using a novel panel of biomarkers in West-Virginian population with breast cancer. *Scientific*

Reports, *11*(1), 1–11. https://doi.org/10.1038/s41598-021-87209-8

Lambert, J., & Thavendiranathan, P. (2016). *Controversies in the Definition of Cardiotoxicity: Do We Care? - American College of Cardiology*. American College of Cardiology. https://www.acc.org/latest-in-cardiology/articles/2016/07/07/14/59/controversies-in-the-definition-of-cardiotoxicity

López-Sendón, J., Álvarez-Ortega, C., Zamora Auñon, P., Buño Soto, A., Lyon, A. R., Farmakis, D., Cardinale, D., Canales Albendea, M., Feliu Batlle, J., Rodríguez Rodríguez, I., Rodríguez Fraga, O., Albaladejo, A., Mediavilla, G., González-Juanatey, J. R., Martínez Monzonis, A., Gómez Prieto, P., González-Costello, J., Serrano Antolín, J. M., Cadenas Chamorro, R., & López Fernández, T. (2020). Classification, prevalence, and outcomes of anticancer therapy-induced cardiotoxicity: The CARDIOTOX registry. *European Heart Journal*, *41*(18), 1720–1729. https://doi.org/10.1093/eurheartj/ehaa006

Madeddu, C., Deidda, M., Piras, A., Cadeddu, C., Demurtas, L., Puzzoni, M., Piscopo, G., Scartozzi, M., & Mercuro, G. (2016). Pathophysiology of cardiotoxicity induced by nonanthracycline chemotherapy. *Journal of Cardiovascular Medicine*, *17*(type II), S12–S18. https://doi.org/10.2459/JCM.0000000000000376

Mendes, M. (2018). Acute coronary syndrome in the oncology patient: An avoidable event? *Portuguese Journal of Cardiology*, *37*(9), 4–7.

Minotti, G., Menna, P., Saponiero, A., Licata, S., Calafiore, A. M., Teodori, G., & Gianni, L. (2001). Paclitaxel and docetaxel enhance the metabolism of doxorubicin to toxic species in human myocardium. *Clinical Cancer Research*, *7*(6), 1511–1515.

Mitchell, J., & Lenihan, D. J. (2020). *Practical Integration of the ASCO Guidelines for Prevention and Monitoring of Cardiac Dysfunction in Survivors of Adult Cancers*. 1–11.

Mladěnka, P., Applová, L., Patočka, J., Costa, V. M., Remiao, F., Pourová, J., Mladěnka, A., Karlíčková, J., Jahodář, L., Vopršalová, M., Varner, K. J., & Štěrba, M. (2018). Comprehensive review of cardiovascular toxicity of drugs and related agents. *Medicinal Research Reviews*, *38*(4), 1332–1403. https://doi.org/10.1002/med.21476

Moslehi, J. J. (2016). Cardiovascular Toxic Effects of Targeted Cancer Therapies. *New*

England Journal of Medicine, *375*(15), 1457–1467. https://doi.org/10.1056/nejmra1100265

Nitiss, J. L. (2009). DNA topoisomerase II and its growing repertoire of biological functions. *Nature Reviews Cancer*, *9*(5), 327–337. https://doi.org/10.1038/nrc2608

Osman, M., & Elkady, M. (2017). A prospective study to evaluate the effect of paclitaxel on cardiac ejection fraction. *Breast Care*, *12*(4), 255–259. https://doi.org/10.1159/000471759

Pardo Sanz, A., & Zamorano, J. L. (2020). Cardiotoxicity': Time to define new targets? *European Heart Journal*, *41*(18), 1730–1732. https://doi.org/10.1093/eurheartj/ehaa013

Purkayastha, K., Seth, R., Seth, S., & Lyon, A. (2017). Cancer therapy-induced cardiotoxicity: Review and algorithmic approach toward evaluation. *Journal of the Practice of Cardiovascular Sciences*, *3*(2), 82. https://doi.org/10.4103/jpcs.jpcs_33_17

Radonjic, T., Simonovic, N., & Turnic, T. N. (2020). *REVIEW PAPER REVIJALNI RAD REVIEW PAPER REVIJALNI RAD REVIEW PAPER AN OVERVIEW OF PHARMACOLOGICAL AND NON □ PHARMACOLOGICAL TREATMENT AS A USEFUL TOOL FOR THE PROTECTION TRETMANA U PREVENCIJI KARDIOTOKSIČNOSTI*. 263–270. https://doi.org/10.2478/sjecr

Romond, E. H., Jeong, J. H., Rastogi, P., Swain, S. M., Geyer, C. E., Ewer, M. S., Rathi, V., Fehrenbacher, L., Brufsky, A., Azar, C. A., Flynn, P. J., Zapas, J. L., Polikoff, J., Gross, H. M., Biggs, D. D., Atkins, J. N., Tan-Chiu, E., Zheng, P., Yothers, G., … Wolmark, N. (2012). Seven-year follow-up assessment of cardiac function in NSABP B-31, a randomized trial comparing doxorubicin and cyclophosphamide followed by paclitaxel (ACP) with ACP plus trastuzumab as adjuvant therapy for patients with node-positive, human epidermal gr. *Journal of Clinical Oncology*, *30*(31), 3792–3799. https://doi.org/10.1200/JCO.2011.40.0010

S. Gillespie, H., J. McGann, C., & D. Wilson, B. (2012). Noninvasive Diagnosis of Chemotherapy Related Cardiotoxicity. *Current Cardiology Reviews*, *7*(4), 234–244. https://doi.org/10.2174/157340311799960672

Sarfati, D., Koczwara, B., & Jackson, C. (2016). The impact of comorbidity on cancer and its treatment. *CA: A Cancer Journal for Clinicians*, *66*(4), 337–350.

https://doi.org/10.3322/caac.21342

Shaikh, A. Y., & Shih, J. A. (2012). Chemotherapy-induced cardiotoxicity. *Current Heart Failure Reports*, *9*(2), 117–127. https://doi.org/10.1007/s11897-012-0083-y

Shiga, T., & Hiraide, M. (2020). Cardiotoxicities of 5-Fluorouracil and Other Fluoropyrimidines. *Current Treatment Options in Oncology*, *21*(4). https://doi.org/10.1007/s11864-020-0719-1

Stone, J. R., Kanneganti, R., Abbasi, M., & Akhtari, M. (2021). Monitoring for Chemotherapy-Related Cardiotoxicity in the Form of Left Ventricular Systolic Dysfunction: A Review of Current Recommendations. *JCO Oncology Practice*, *17*(5), 228–236. https://doi.org/10.1200/op.20.00924

Subramaniam, S., Kong, Y.-C., Zaharah, H., Uiterwaal, C. S., Richard, A., Tab, N. A., Deniel, A., Chee, K.-H., Bustamam, R. S., See, M.-H., Fong, A., Yip, C.-H., & Bhoo-Pathy, N. (2021). Baseline cardiovascular comorbidities, and the influence on cancer treatment decision-making in women with breast cancer. *Ecancermedicalscience*, *15*, 1–11. https://doi.org/10.3332/ecancer.2021.1293

Suter, T. M., & Ewer, M. S. (2013). Cancer drugs and the heart: Importance and management. *European Heart Journal*, *34*(15), 1102–1111. https://doi.org/10.1093/eurheartj/ehs181

Thavendiranathan, P., Grant, A. D., Negishi, T., Plana, J. C., Popović, Z. B., & Marwick, T. H. (2013). Reproducibility of echocardiographic techniques for sequential assessment of left ventricular ejection fraction and volumes: Application to patients undergoing cancer chemotherapy. *Journal of the American College of Cardiology*, *61*(1), 77–84. https://doi.org/10.1016/j.jacc.2012.09.035

Trapani, D., Zagami, P., Nicolò, E., Pravettoni, G., & Curigliano, G. (2020). Management of cardiac toxicity induced by chemotherapy. *Journal of Clinical Medicine*, *9*(9), 1–18. https://doi.org/10.3390/jcm9092885

Vu, T., & Claret, F. X. (2012). Trastuzumab: Updated mechanisms of action and resistance in breast cancer. *Frontiers in Oncology*, *2 JUN*(June), 1–6. https://doi.org/10.3389/fonc.2012.00062

W, Y., & M, Z. (2021). Biomarkers with Potential Predictive Value for Cardiotoxicity in Anticancer Treatments. *Chinese Medical Sciences Journal = Chung-Kuo i Hsueh k'o Hsueh Tsa Chih*, *36*(4), 1. https://doi.org/10.24920/003790

Waks, A. G., & Winer, E. P. (2019). Breast Cancer Treatment: A Review. *JAMA - Journal of the American Medical Association*, *321*(3), 288–300. https://doi.org/10.1001/jama.2018.19323

Zaha, V. G., Hayek, S. S., Alexander, K. M., Beckie, T. M., Hundley, W. G., Kondapalli, L., Ky, B., Leger, K. J., Meijers, W. C., Moslehi, J. J., Shah, S. H., & American Heart Association Cardio-Oncology Subcommittee of the Council on Clinical Cardiology; and Council on Genomic and Precision Medicine. (2021). Future Perspectives of Cardiovascular Biomarker Utilization in Cancer Survivors: A Scientific Statement From the American Heart Association. *Circulation*, CIR0000000000001032. https://doi.org/10.1161/CIR.0000000000001032

Zamorano, J. L., Lancellotti, P., Rodriguez Muñoz, D., Aboyans, V., Asteggiano, R., Galderisi, M., Habib, G., Lenihan, D. J., Lip, G. Y. H., Lyon, A. R., Lopez Fernandez, T., Mohty, D., Piepoli, M. F., Tamargo, J., Torbicki, A., Suter, T. M., Achenbach, S., Agewall, S., Badimon, L., … Szmit, S. (2016). 2016 ESC Position Paper on cancer treatments and cardiovascular toxicity developed under the auspices of the ESC Committee for Practice Guidelines. *European Heart Journal*, *37*(36), 2768–2801. https://doi.org/10.1093/eurheartj/ehw211

Zhang, S., Liu, X., Bawa-Khalfe, T., Lu, L. S., Lyu, Y. L., Liu, L. F., & Yeh, E. T. H. (2012). Identification of the molecular basis of doxorubicin-induced cardiotoxicity. *Nature Medicine*, *18*(11), 1639–1642. https://doi.org/10.1038/nm.2919