Review

Prognostic Factors for Cardiotoxicity Among Children With Cancer: Definition, Causes and Diagnosis With Omics Technologies

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Prognostic Factors for Cardiotoxicity among Children with Cancer: Definition, Causes and Diagnosis with Omics' Technologies'

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Abstract: Improvements in the treatment of childhood cancer have considerably enhanced survival rates over the last decades, to over 80% as per today. However, this great achievement has been accompanied by the occurrence of several early and long-term treatment-related complications major of which is the cardiotoxicity. The chemotherapeutic agents that have been implicated as a cause of cardiotoxicity, are mostly anthracyclines and mitoxantrone, alkylating agents, proteasome inhibitors and to a lesser extent anti-microtubule agents, cisplatin, monoclonal antibodies, small molecular tyrosine kinase inhibitors, and nucleotide synthesis inhibitors. Routine diagnosis and monitoring of cardiotoxicity rely on electrocardiography and echocardiography. For the early detection of cardiotoxicity, in recent years, major studies have been conducted using biomarkers such as troponin, N-terminal pro b-natriuretic peptide, etc. Despite refinements in diagnostics still severe limitations exist, due to the increase of the above-mentioned biomarkers, only after significant cardiac damage has occurred. Lately, research has been expanded by introducing new technologies and finding new markers by omics approach. These new markers could be used not only for early detection, but also for early prevention of cardiotoxicity. The omics science, which includes genomics, transcriptomics, proteomics, and metabolomics, offers new opportunities for biomarker discovery in cardiotoxicity and may provide understanding of mechanisms of cardiotoxicity, beyond traditional technologies. This article reviews the contemporary definition of cardiotoxicity, older and newer chemotherapeutic agents mainly involved in cardiotoxicity and the methods of early and preventive diagnosis, using omics technology.

Keywords: cardiotoxicity; childhood cancer; chemotherapeutics agents; biomarkers; omics technology

Introduction

With the induction of new chemotherapeutic agents over the years, 5-year survival rate for childhood malignancies exceeds 80%. Caring for these patients includes not only early survival but also later outcomes. Chronic health conditions and health-related quality of life are noted among them as many long-term treatment related complications have resulted in increasing morbidity and mortality rates. Cardiotoxicity represents the most serious non-hematological toxicity of chemotherapeutic drugs. It is noted that childhood cancer survivors have an 8x higher risk in mortality due to cardiovascular disease and 6x increased risk of congestive heart failure. Most importantly early cardiotoxicity may affect the design of chemotherapy and the omission of radiotherapy resulting in incomplete cancer treatment and consequently inferior outcome. The study of cardiotoxicity among the pediatric population is, as expected, of particular importance due to the long-life expectancy. [1-6]

Cardiotoxicity

Definition

As a term, cardiotoxicity was first described in 1946 as the damage on the heart caused from local anesthetics, mercurial diuretics, and digitalis. Later in the 1970s, the term broadened to encompass cardiac complications related with anthracyclines (doxorubicin and daunorubicin), combination therapies like doxorubicin and radiation and drugs like for 5-fluorouracil. Presently there is an increased research interest both basic and clinical aiming in detecting and managing cardiotoxicity as early as possible.

The definition of cardiotoxicity has a great significance for patient's management. Based on International Cardio-Oncology Society (IC-OS) the cardiovascular complications of chemotherapy can be separated into the following clinical entities and/or categories: i) cardiac dysfunction: cardiomyopathy/ heart failure (HF), ii) vascular toxicity, iii) myocarditis, iv) arterial hypertension and v) arrhythmias and QT prolongation. [7,8]

The most preponderant diagnosis of cardiotoxicity is based on the changes found in left ventricular (LV) systolic function measured by left ventricular ejection fraction (LVEF). Different organizations have defined cardiotoxicity in several ways using different threshold changes in LVEF [8]. The need to harmonize all these definitions satisfied by the International Cardio-Oncology Society (IC-OS) and supported by 2022 ESC Guidelines. [7,8]

Cardiotoxicity can be categorized according to the time of presentation as acute, early onset and late onset. Cardiotoxicity could be reversible if addressed while ints early stages, [9] Acute (<1%) toxicity could occur either after administrating a single dose, or after a course of chemotherapeutic agents, as long as the onset of clinical manifestations is within the first two weeks following the end of the administration. If presented within the first year of treatment, it is characterized as Early onset (1-18%). Late or chronic onset is manifested years or even decades following the treatment. [9] The percentage of late onset cardiotoxicity varies in literature mainly due to the different definitions used, the detection methods of cardiotoxicity, the population monitored, and also due to the study design. It seems that over 50% of pediatric cancer survivors show subclinical decline in myocardial function and over 16% show symptoms of clinical HF, especially those who have been exposed to anthracyclines. [9]

Abnormalities of ventricular repolarization and electrocardiographic QT-interval alterations, supraventricular and ventricular arrhythmias, acute coronary syndromes, and pericarditis and/or myocarditis-like syndromes are the hallmarks of acute or early onset cardiotoxicity. [9] In contrast, asymptomatic systolic and/or diastolic LVD, which can result in dilated cardiomyopathy, is the most typical indicator of chronic cardiotoxicity. [9,10] Clinical and sub-clinical cardiovascular damage, coronary artery disease, and cerebrovascular events are other conditions linked to treatment-related complications. Survivors had an almost six-fold higher risk of heart failure, a five-fold higher risk of myocardial infarction, a six-fold higher risk of pericardial disease, and an almost five-fold higher risk of valvular abnormalities compared to their siblings. [11-13]

Chemotherapeutic drugs

Anthracyclines

Anthracyclines, primarily doxorubicin but also daunomycin, epirubicin and idarubicin, are some of the commonest used agents for both hematologic and solid tumors. Acute cardiotoxicity due to anthracyclines may present as hypotension, tachycardia, arrhythmia, transient depression of left ventricular function, myocarditis, pericarditis, and acute coronary syndrome. The late onset cardiotoxicity caused by high cumulative dose of anthracyclines, mainly includes signs and symptoms of cardiomyopathy and chronic heart failure. [9]

Mitoxantrone is a an anthracenedione, or anthracycline analogue and has similar anthracycline mechanisms of action. Mitoxantrone might cause a wide variety of heart conditions, such as disturbances of cardiac rhythm, chronic heart failure, and persistent diastolic dysfunction in the absence of an impairment of left ventricular ejection fraction. [10]

The prevalent concept of how anthracycline action may cause heart damage, involves the production of oxygen radicals, which in turn damage the DNA, proteins, and lipids and lead to cellular dysfunction and myocyte death. [14,15-16]

Cardiolipins are abundantly found on the inner mitochondrial cell membrane. By having an increased affinity for anthracyclines, they in turn allow for their increased cell entry. Upon cell entry by passive diffusion, they can reach much higher intracellular concentrations compared to extracellular compartments. Within the cell, they form complexes by binding to iron thus producing free radicals and reactive oxygen species, which in turn cause cell damage and death. By peroxidizing lipids of the cell membrane, those elements may also damage the cell membrane. As cardiomyocytes, contain an abundance of mitochondria, they are more susceptible to anthracycline damage because of the depletion of glutathione peroxidase (antioxidant). [15]

Other mechanisms of cardiotoxicity include alterations to gene expression and nitric oxide synthase activity, which lead to reduced creatine kinase activity and function in mitochondria and ultimately cell death. [15] After exposure to anthracyclines, many of these subcellular sequelae continue to develop for weeks, shedding light on the mechanisms of chronic cardiomyopathy.[14]

Another identified mechanism of doxorubicin-mediated cardiotoxicity is changes in topoisomerase-II (Top2). Topoisomerase II (TOP2) is a molecule that anthracyclines bind to and inhibit, preventing the growth of tumors. DNA's phosphate backbone is broken, twisted, and then resealed by topoisomerases, allowing the double helix's tension to be changed during transcription and replication. Anthracyclines intercalate into DNA, forming complexes with TOP2 that halted the enzyme's activity and trigger a DNA-damage reaction that results in cell death. [14,15,17].

The mechanisms of mitoxantrone associated cardiotoxicity remain yet to be completely understood, the formation of reactive oxygen species in myocardial cells is thought to lead to tissue damage through interactions with cellular iron metabolism, [10].

Nucleotide synthesis inhibitors

The clinical presentation of methotrexate and Fluorouracil (5-FU)-induced cardiotoxicity includes myocardial ischemia, cardiogenic shock, heart failure and cardiomyopathy [10]. Coronary spasm is the most frequently reported mechanism of 5-FU-induced cardiotoxicity [10]. Data derived from animal models indicate that these chemotherapeutic agents induce oxidative stress and subsequent apoptosis of cardiomyocytes and endothelial cells. [10,17]

Alkylating agents

Adjuvant DNA alkylating agents, such as cyclophosphamide (CP) and ifosfamide (IFO), suspend DNA synthesis in cancer cells. These two agents are similar in structure and engender a similar pattern of cardiotoxic effects causing acute heart failure, hemorrhagic myopericarditis and arrhythmia. [10,18] CP- and IFO-induced acute cardiotoxicity is attributed mainly due to a raise in free oxygen radicals and the lower antioxidant defense mechanism in the myocardium. A recent study by Sayed-Ahmed MM et al demonstrated that CP- and IFO-induced cardiotoxicity is due to the inhibition of long-chain fatty acid oxidation via repression of carnitine palmitoyl transferase I and fatty acid binding protein. [19]

Tyrosine kinase inhibitors

Dasatinib, imatinib, lapatinib, sorafenib, nilotinib, and sunitinib are examples of small molecule tyrosine kinase inhibitors (TKIs) that suppress cancer cell proliferation and induce apoptosis of cancer cells. Imatinib, dasatinib, and nilotinib are the three FDA-approved TKIs for use as first-line chronic myeloid leukemia therapy in pediatrics. Also, sorafenib is used in young adults. The pathophysiological mechanism of TKI-induced cardiotoxicity is mitochondrial impairment and cardiomyocyte apoptosis. [10, 18, 20] Each of the above drugs is associated with a different type of cardiotoxicity. For example, dasatinib is more often associated with pleural effusion and less with hypertension, HF, pericardial effusion, and pulmonary hypertension. Nilotinib is associated with

peripheral artery disease, hypertension and prolonged QTc. In contrast, imatinib is related with less cardiotoxicity than the others TKIs.[7]

Anti- microtubule agents

Anti- microtubule agents, including docetaxel, paclitaxel and vinca alkaloids, prevent the polymerization or depolymerization of microtubules. [17] The clinical feature of the cardiotoxicity induced by anti-microtubule agents is mostly ischemia, and arrhythmia. Among all anti-microtubule agents in clinical use, paclitaxel induces the release of histamine, which in turn, activates specific cardiac receptors raising the myocardium's oxygen need, leading to coronary vasoconstriction. Also, Zhang et al reported that the frequency of spontaneous calcium concentration in cardiomyocytes was significantly increased after paclitaxel treatment. This finding could be of great significance as fluctuations of blood calcium levels are linked to arrhythmogenesis. [10,21]

Cisplatin

Cisplatin is an efficacious chemotherapeutic drug with a strong antitumor effect against a wide range of neoplasms. However, the drug's acute and cumulative cardiotoxicity, including electrocardiograph (ECG) abnormalities, angina and acute myocardial infarction, hypertension and hypotension, arrhythmias, myocarditis, cardiomyopathy, and congestive heart failure, is a significant factor that restricts cisplatin treatment. Cisplatin cardiotoxicity can be caused by reactive oxygen species generation, which leads to the creation of oxidative stress and the transition to a prothrombotic state, or by a direct toxic effect on cardiac myocytes.[10]

Monoclonal antibodies

Monoclonal antibodies, including bevacizumab (Avastin) and trastuzumab not used in children, inhibit angiogenesis. Bevacizumab blocks vascular endothelial growth factor (VEGF), while trastuzumab inhibits human epidermal growth factor receptor 2 (HER2) in cancer cells. Bevazicumab causes mostly hypertension, congestive heart failure, thromboembolic events of artery and vein through the mechanic of oxidate stress-induced by the cardiomyocyte apoptosis. Monoclonal antibodies are not widely used in children with malignancy, so we will not discuss them further. [10,18, 20]

Proteasome inhibitors

A new therapeutic option for the treatment of Acute Lymphoblastic Leukemia (ALL) includes proteasome inhibitors. [18, 20] Bortezomib and carfilzomib are two newly prescribed drugs with the potential to cause cardiac dysfunction. [22] Comparatively to carfilzomib (up to 25%), bortezomib has a lower incidence of heart failure (up to 4%). The pathogenesis of proteasome inhibitor cardiotoxicity is not currently well understood. Exposure to proteasome inhibitors in a prenatal mouse model has shown that they can induce oxidative stress leading to myocardial dysfunction. Carfilzomib is also known to induce renal toxicity and microangiopathy, as a consequence of endothelial dysfunction. Combining these studies reveals a complicated mechanism of cardiotoxicity linked to proteasome inhibitors, including alterations to the heart's muscle and vasculature that may be more severe with carfilzomib than bortezomib due to the irreversible nature of proteasome inhibition of carfilzomib. [23-24]

Risk factors

A significant number of factors contribute to the onset of cardiotoxicity. Several types of chemotherapeutic drugs may cause cardiotoxicity, as referred above. These drugs act on cancer cells through a variety of mechanisms and promote cardiotoxicity with distinctive clinical symptoms and underlying mechanisms. (Table 1) [7, 10, 16]

Table 1. Chemotherapeutic drugs and cardiovascular toxicity.**(**Adjusted by Rochette et al, Trends Pharmacol Sci. 2015 Jun;36(6):326-48 [112]).

Medicine/Cardiotoxicity	**Incidence (%)**	**Arrhythmia**	**Myocardial ischemia**	**Vascular toxicity**	**Heart failure**	**QT prolongation**	**Arterial Hypertension**
Anthracyclines							
Doxorubicin	3-26	×××	X	NE	Xxx	NE	X
Doxorubicin Liposomal	2	×	xx	NE	x	NE	X
Epirubicin	0,9-3,3	×	X	NE	X	NE	X
Daunorubicin		××	X	NE	X	NE	X
Idarubicin	5-18	×××	X	NE	xx	NE	X
Antibiotics							
Mitoxantrone	0,2-30	×××	xx	NE	Xx	NE	Xx
Mitomycin-c	10	Xx	xx	NE	Xx		NE
Monoclonal antibody							
Trastuzumab	1,7-8	Xx	X	Xx	Xxx	NE	Xx
Bevacizumab	1,6-4	Xx	xx	xxx	xx	NE	Xx
Pertuzumab	0,7-1,2	X	X	X	Xx	NE	x
dinutuximab beta		NE	xx	NE	Xx	NE	Xx
Rituximab		X	xx	Xxx	X	NE	Xx
Tyrosine kinase inhibitors							
Dasatinib	2-4	xxx	xx	Xx	Xx	xx	Xx
Nilotinib	1	xx	NE	x	Xx	xx	Xxx
Vermurafenib		xx	xx	Xx	x	NE	Xx
Sorafenib	2-28	X	xx	Xx	Xx	NE	xx
Sunitinib	2,7-15	X	xx	Xx	Xxx	x	Xxx
Erlotinib	7-11	NE	xx	Xx	NE	NE	NE
Lapatinib	0,2-1,5	NE	xx	X	NE	xxx	NE
Pazopanib	7-11	NE	xx	Xx	X	NE	Xxx
Imatinib	0,2-2,7	NE	xxx	Xx	Xx	NE	NE
Proteasome inhibitors							
Bortezomib	2-5	X	X	X	X	NE	X
Carfilzomib	11-25	Xx	xx	NE	X	NE	X
Antimetabolite							
5-fluorouracil	2-20	xxx	xxx	NE	X	NE	NE
Capecitabine		xxx	xxx	Xx	NE	NE	NE
Clofarabine	27					NE	
Alkylating agents							
Cyclophosphamide	7-28	NE	NE	X	NE	NE	NE
Ifosfamide	0,5-17	NE	NE	X	Xx	NE	NE
Cisplatin	rare	NE	NE	Xx	NE	NE	NE
Antimicrotubule agent							
Paclitaxel	<1	xx	X	NE	X	NE	x

nab-paclitaxel		xx	NE	X	NE	NE	X
Docetaxel	2,3-13	xx	xx	NE	X	NE	Xx
Alkaloids of vinca							
Vincristine	25	xx	X	NE	NE	xx	X
Vinblastine	NE		X	NE	NE	NE	X
Vindesin	NE		NE	NE	NE	NE	NE
Vinorelbin	NE		X	NE	NE	NE	NE

The overall dose and mode of administration of each chemotherapeutic agent play an aggravating role in causing cardiac damage. For example, anthracycline induced cardiotoxicity is known to be both cumulative and dose-related, indicating that each administered dose induces sequential or additional damage. [18,25] The cumulative total anthracycline dose is the most important risk factor for cardiac dysfunction. [26]. In retrospective research, Von Hoff et al. [27] observed that when a patient receives a combined doxorubicin dose of 400, 550, and 700 mg/m2, the incidence of cardiotoxicity is 3, 7, and 18%, respectively, with dose-limiting toxicity. Another study in adolescents found that even at dosages of 180–240 mg/m2, 30% of participants experienced subclinical episodes 13 years after therapy [29]. These results imply that there is no anthracycline dose that is considered safe. Reduced cardiac function has been correlated with dosages as low as 100 mg/m2. [28, 30-31].

In addition, female gender, age (under <5 years old), clinical condition of the patient (extent of disease, infection), genetic background, pre-existing cardiac disease, and combinations of cardiotoxic drugs, play an important role in causing cardiac damage [32] (Table 2). The pediatric population is more homogeneous, as a study population, since there are no confounding cardiovascular risk factors (diabetes, smoking, arterial hypertension). [7, 14, 33]

Table 2. Risk Factors.

Risk factors related in child	Risk factors related in therapy
• Female sex	
• Age < 5 years	• Anthracycline > 250 mg/m2 equivalent doxorubicin
• genetic background	• Cumulative dose
• pre-existing cardiac disease	• Irradiation
• Cardiovascular risk factors (Diabetes, obesity, hyperlipidemia, Hypertension)	• combination of cardiotoxic drugs

The risk of developing cardiotoxicity is also increased by concurrent radiation exposure to the chest. In addition to the myocardium, radiation therapy has the potential for damaging the pericardium, heart vessels, and conductive tissue. [18]

Carriers of certain genetic mutations are also more susceptible to cardiotoxicity. [9,18] Our understanding of genetic susceptibility to anthracyclines-related cardiotoxicity has been influenced by a sizable body of research, as we will describe below.

Imaging

Diagnostic approaches for chemotherapy-induced cardiotoxicity include electrocardiography and echocardiography, used as methods of monitoring cardiac function before, during and after treatment.

Electrocardiography (ECG) can be used to identify any early signs of cardiac toxicity, such as resting tachycardia, ST-T wave abnormalities, conduction disturbances, QT interval prolongation, or arrhythmias. However, these ECG findings could be induced by several factors unrelated to cardiotoxic treatment. These ECG abnormalities may be reversed and are not always related to the development of chronic cardiomyopathy. [7,26 ,33]

The two-dimensional Echocardiography (2D) is the most used imaging technique to monitor cardiac function. It is non-invasive, cheap, readily available and does not expose the patient to further radiation. However, standard echocardiographic parameters like LVEF may lack sensitivity for the detection of systolic dysfunction. [33]

Considering the poor sensitivity of 2D LVEF measurement, the use of global systolic longitudinal myocardial strain (GLS) analysis has become an area of interest. [33-34] A pathogenic percentage reduction of GLS greater than 15% from baseline is regarded as a sign of early LV dysfunction. When possible, it is preferable to use these sophisticated echocardiographic measures as the foundation for clinical decision.

Other methods in the monitoring of these patients are cardiac magnetic resonance imaging (CMR), nuclear cardiac imaging (MUGA) and myocardial perfusion imaging (MPI). There are 2 techniques for MPI: single photon emission computed tomography (SPECT) and position emission tomography (PET). All these methods take considerably longer than a follow-up echocardiography and might not be as accessible at all pediatric facilities. [33].

Biomarkers

Several biomarkers have been assessed for their efficacy in early prediction of patients' risk of cardiotoxicity and identification of cardiac dysfunction. The World Health Organization defines biomarkers as any element, structure, or process that can be detected in the body (or its byproducts) and affects or forecasts the development or course of a disease.

In accordance with the literature, troponin and natriuretic peptide are the most studied biomarkers for the detection of both early cardiotoxicity and its later follow-up. Lipshultz et al showed that the elevation of cardiac troponin T and N-terminal pro-brain natriuretic peptide (NT-pro-BNP) in children with acute lymphoblastic leukemia have been associated to notably reduced left ventricular (LV) mass, abnormal LV end-diastolic posterior wall thickness, and abnormal LV thickness-to-dimension ratios, all of which suggested LV remodeling, respectively, 4 years later. [35] However, further research hasn't pointed out an association between acute or chronic troponin release and left ventricular dysfunction, but in contrary found association with NT-pro-BNP in childhood cancer survivors. [36-39]

We should be especially careful in evaluating troponin and natriuretic peptide values in children < 1 year of age due to their normally elevated values at these ages. [40]

Other biomarkers investigated include inflammation markers, such as C-reactive protein (CRP) and growth/differentiation factor 15 (GDF-15) [37], oxidative stress markers such as myeloperoxidase, vascular remodeling markers such as placental growth factor and soluble Fms-like tyrosine kinase receptor 3, and fibrosis markers. (Galectin 3). [35,41-49] More Those conventional biomarkers usually show significant changes only after heart damage occurs.

To determine the proper use of these biomarkers in clinical practice, new prospective and multicenter studies with large populations, well-standardized dosing methodologies, well-defined time of sampling, and cardiologic end points are required.

Omics

In the last decades, new research and clinical studies have attempted to identify possible biomarkers of early cardiac damage by chemotherapeutic agents using omics technology. So, the omics science offers new opportunities for biomarker discovery in cardiotoxicity and may provide understanding of cardiotoxicity beyond traditional technologies. Omics technology includes genomics, transcriptomics, proteomics, and metabolomics.

Genomics

Cumulative anthracycline dose and other related risk factors seem not to be the exclusive risk factor responsible for the significant individual variation in the incidence and severity of heart failure in pediatric cancer survivors. Several studies have revealed how important host genetic

polymorphisms could lead to differential risk of cardiotoxicity among cancer survivors with otherwise identical clinical and treatment-related risk factors by using genome wide association or candidate gene approaches. [50-53] This explains why some patients experience cardiotoxicity while other patients can tolerate high doses of chemotherapy without heart damage.

The cumulative dose of anthracyclines and other risk variables do not appear to be the only ones contributing to the considerable individual variability in the occurrence and severity of heart failure in pediatric cancer survivors

Genomic polymorphisms are small changes in a specific part of the DNA chain. One or more polymorphisms can determine a range of patient characteristics such as their ability to metabolize and eliminate genotoxic substances. Cancer treatment-related cardiovascular toxicity risk may be influenced by genetic variation. Significant efforts using targeted and whole genome correlation studies have been made to reveal the pharmacogenomic causes of this predisposition. [50,68-62]

At least 45 SNPs located in 34 genes have been associated with anthracycline induced cardiotoxicity. Many of these associations require further investigation through replication and/or functional and mechanistic studies to make sure we confirm and better understand the roles of these associated variant in anthracycline-related cardiotoxicity (ACT).[9]

Polymorphisms in solute carrier transporter (SLC) genes are associated with ACT. One of the functions of the SLC family is acting as drug transporters for anthracyclines and thus provides biological support for these genetic associations. Research on childhood cancer survivors has discovered correlations between ACT risk and protective variants in SLC such as SLC28A3, SLC22A17, SCL22A7. These findings were successfully replicated [54-59]. Also, different studies reported protective variants in SLC10A2 and SLC22A1 [55]. SLC22A6 was first mentioned in the context of ACT by Sagi et al in patients treated for childhood ALL [57].

Retinoic acid receptor gamma (RARG) has been involved in cardiac development and remodeling through the repression of Top2b [60]. A recent genome-wide association study, Aminkeng [51] et al, uncovered a non-synonymous variant rs2229774 in *RARG* that was significant associated with ACT in survivors of childhood cancer. Specifically, rs2229774-carriers had a significantly increased risk of developing ACT as compared to non-carriers. [51]

Studies have also revealed an elevated risk brought on by a variation in the UGT1A6 gene, a member of the glucuronosyl transferase family. Through the glucuronidation path, the UGT1A6 play a significant role in the detoxification of drugs, including the metabolites of anthracyclines. [51,54,58]

Polymorphisms in adenosine triphosphate-binding cassette transporter (ABC) genes are related with cardiotoxicity in childhood patient cancers treated with anthracyclines. The ABC genes seems to play a role as efflux transporters of drugs including anthracyclines, so may have important effects in the myocardium. Eight variants in five genes (ABCB1, ABCB4, ABCC1, ABCC2, ABCC5) were associated with cardiotoxicity, especially with reduced ejection fraction. [57, 61-64]

Other studies investigated polymorphisms in carbonyl reductase genes, which have been associated with dose dependent increase in cardiomyopathy risk. Carbonyl reductase (CBR) will reduce anthracyclines to cardiotoxic alcohol metabolites. As Blanco et al has showed, among childhood cancer survivors, homozygosity for G allele in CBR3 leads to increased cardiomyopathy risk associated with low- to moderate-dose anthracyclines. Patients homozygous with the CBR3 V244M G allele have no safe cut off minimum dose. [65-66]

A recent study showed a gene environment interaction between single-nucleotide polymorphism on the CELF4 gene and higher dose of anthracyclines. [67] CELF4 (CUGBP Elav-Like Family Member 4) protein is responsible for pre-mRNA alternative splicing of TNNT2, the gene that encodes for cardiac troponin T.

Aminkeng [58] et al gathered the evidence-based clinical practice recommendations for pharmacogenomic testing and emphasized that the RARG gene rs2229774, UGT1A6 * 4 rs1786378, SLC28A3 rs7853758 have the potential to further discriminate patients at high and lower risk of ACT. A pharmacogenetic test for these genetic variations in RARG, SLC28A3, and UGT1A6 has been released at the British Columbia Children's Hospital since the publication of these guidelines. Based on genetic and clinical risk variables, tested patients were divided into several risk groups, and

therapy adjustments were chosen in accordance with this risk. Early evidence indicates that the British Columbia Children's Hospital's pharmacogenetic testing was effective in lowering the incidence of ACT in children, which should inspire additional clinics to utilize this pharmacogenetic test.

These findings might help develop prediction models that will spot patients who will be particularly susceptible to ACT and who need their therapy modified or who need closer monitoring. Further independent research may make it possible to identify people before treatment with a genetic predisposition to cardiovascular toxicity and for whom more thorough screening or perhaps preventive measures should be implemented. Replication analyses, however, occasionally failed to support the initial findings. Numerous factors, including the variability of cohorts, ambiguities in the definition of ACT, variations in procedures, and the type or dosage of the chemotherapeutic drugs used, may contribute to this. To increase the diagnostic and prognostic role in predicting ACT, more research is required.

Transcriptomics

Another interesting area is the integration of microRNAs in the early detection of cardiotoxicity. Recently, the potential use of circulating MicroRNAs (miRNAs) has been studied as possible specific biomarkers and therapeutic targets of cardiac disease. [73-79]

MicroRNAs are small endogenous non-coding RNAs of 21–24 nucleotides, acting as post-transcriptional gene regulators by inhibiting and/or degrading target messenger RNAs (mRNAs). Bioinformatics data suggest that each miRNA molecule can control hundreds of gene targets, thus indicating the potential effect of miRNAs on virtually any genetic pathway. MiRNAs play a significant role in different biological processes including proliferation, differentiation, development, and cell death. Furthermore, several miRNAs are involved in regulating heart development from embryonic to adult stage and their dysregulation leads to various heart diseases such as, arrhythmias, essential hypertension, heart failure, cardiomyopathy, cardiac hypertrophy, and atherosclerosis. [80-81]

Cardiotoxic effect of chemotherapeutic agents may lead to specific miRNAs with changed expression. These could be used to investigate the toxicity of potential drug candidates on cardiomyocytes and cell lines originating from the heart in a preclinical in vitro setting. The potential use of circulating miRNAs in plasma as indicators of drug-induced cardiotoxicity has undergone much research during the last several years.[80]

Nearly 30 circulating miRNAs have had their levels altered, both increasing and decreasing, and these changes have been linked to HF and associated pathologies. MiRNAs including miR-1, miR-133, miR-208a/b, miR-499, miR-29, and miR-34, which are substantially expressed in the myocardium compared to other tissues, are the ones that are primarily being researched [73]. In addition, a variety of harmful substances alter the miRNA profile in both plasma and cardiac tissue. Even at low toxin concentrations, where other tissue damage biomarkers are not discernible, alterations in miRNAs can be measured. [80] Most studies use data from experimental animals, while those utilizing clinical patient samples are limited.

MiR-1 is a skeletal muscle specific miRNA that has an important role in cardiac development, function, and disease. Abnormal miR-1 levels are associated with acute myocardial infraction, heart failure, arrhythmias, ventricular dysfunction, cardiac hypertrophy, and myocyte hyperplasia. [82] MiR-499 and miR-208 are associated with acute myocardial infraction and HF. [82] Circulating levels of miR-133a have been associated with increased risk of cardiovascular diseases. Increased levels of miR-133a have been detected in patients with acute myocardial infarction earlier than cardiac troponin T increase. [83] MiR-133 are two miRNAs, named miR-133a and miR-133b, highly expressed in human heart and seems to be involved in heart development and myocyte differentiation.

The analysis of circulating miRNAs in breast cancer patients receiving doxorubicin (DOX) identified miR-1 as a potential candidate for the early detection of DOX-induced cardiotoxicity. [84] Leger et al investigated other possible markers of cardiotoxicity in children and young adults treated with anthracycline chemotherapy (AC). Candidate plasma profiling of 24 miRNAs was performed in

33 children before and after a cycle of AC or non-cardiotoxicity chemotherapy. MiR-1, miR-29b and miR-499 were reported to be upregulated in pediatric patients following acute initiation of AC.[85-86]. Monitoring the plasma levels of miR-208a and miR-208b showed an elevaton in patients with myocardial damage and were even detected earlier than cardiac troponins [87]. This is concordance with the findings from other studies. [73-74,87-88] Table 3 provides a summary of the major miRNAs linked to drug-induced cardiotoxicity.

Table 3. Summary of major miRNAs link to drug-induced cardiotoxicity in people.

MiRNA	Drug	Modulation	Species	System	References
miR-1	doxorubicin	increase	female patients	plasma	Riguad et al, Oncotarget 2017
miR-1, miR-29b, miR-499	anthracyclines	increase	children and young adult	plasma	Leger et al, J Am Heart Assoc. 2017
miR1254	bevacizumab	increase	Humans	plasma	Zhao et al, Tumour Biol. 2014
miR29 miR499	doxorubicin	increase	Children	plasma	Oatmen et al, Am J Physiol Heart Circ Physiol, 2018
miR208	doxorubicin	nothing	female patients	plasma	Calvalho et al, J Appl Toxicol 2015

In addition to anthracyclines, other cytotoxic agents have shown cardiotoxic effects and biomarkers of their pathomechanism have been searched for, including miRNAs. Patients with bevacizumab-induced cardiotoxicity when compared with controls were found to have increased levels of five miRNAs. In the validation experiments, two of these (miR-1254 and miR-579) shown valuable specificity. MiR-1254 exhibited the strongest correlation with the clinical diagnosis of bevacizumab-induced cardiotoxicity [89].

With regards to a number of features of drug-induced cardiotoxicity, miRNAs appear to be a promising agent. A potentially successful method for preventing severe problems is the identification of patients with subclinical cardiotoxicity through the detection of cardio-specific miRNAs circulating in plasma that are not present under normal circumstances. A supposedly efficient method for identifying people with subclinical cardiotoxicity is the detection of cardio-specific miRNAs circulating in plasma that are not present under normal circumstances. [80]. Many other research studies should focus on how the miRNAs profile changes when interacting with drugs with proven cardiotoxicity.

Proteomics

The proteomic data available to date on chemotherapy-induced cardiac toxicity are limited, mainly involving anthracyclines, and related to experimental animal studies. [90]

Proteomics is the study of proteins, which are essential components of organisms and have a variety of functions. The proteome consists of all the proteins expressed by a cell, tissue, or organism. Proteomics could give us important information for a number of biological problems.

Ohyama et al identified cellular processes in mouse heart tissue from control rats and rats affected by different Adriamycin and docetaxel dosing protocols using a toxicoproteomic approach. They identified 9 different proteins that were expressed in the control and in the two treatment

groups, and were involved in energy production pathways, such as glycolysis, the Krebs cycle and the mitochondrial electron transport chain. [91]

Kumar et al in 2011 used a rat model of Doxorubicin-induced cardiotoxicity to show the differential regulation of several key proteins, including protein S that are stress responsive (ATP synthase, enolase alpha, alpha B-crystallin, translocation protein 1 and stress-induced phosphoprotein 1), and apoptotic/cell damage markers (p38 alpha, lipocortin, voltage-dependent anion-selective channel protein 2, creatine kinase and MTUS1). [86]

More recently, Desai et al pinpointed possible biomarkers of early cardiotoxicity in plasma from male B6C3F1 mice that have received weekly intravenous dose of 3 mg/kg doxorubicin (DOX) or saline (SAL) for 2, 3, 4, 6, or 8 weeks (corresponding to cumulative doses of 6, 9, 12, 18, or 24 mg/kg DOX).They suggested the neurogenic locus notch homolog protein 1 (NOTCH1) and von Willebrand factor (vWF) as early biomarkers of DOX cardiotoxicity, to address the clinically significant question of identifying cancer patients at risk for cardiotoxicity. [92]

Finally, Yarana et al using a mouse model of DOX-induced cardiac injury, quantified serum extracellular vehicles (EVs), assayed proteomes, counted oxidized protein levels in serum EVs generating following DOX treatment and examined the alteration of EV content. The release of EVs containing brain/heart glycogen phosphorylase (PYGB) before the increase in cardiac troponin in the blood following DOX therapy suggests that PYGB is an early indicator of cardiac damage, according to the proteomic profiling of DOX_EVs. [93]

To find out if these pathways could result in the discovery of early markers of cardiotoxicity, more research in that area is required.

Further studies are needed in that field to investigate if these pathways could result in the discovery of early markers of cardiotoxicity.

Metabolomics

Metabolomics is the upcoming new science with the potential to further increase our knowledge on cancer biology and the search for prognostic biomarkers. Up to now most studies use metabolomic data from experimental animals, while those utilizing clinical patient samples are extremely limited.

Metabolism is more directly related to the phenotype and physiology of a biological system. Metabolomics is the study of all cellular metabolites (hydrocarbons, amino acids, sugars, fatty acids, organic acids, steroids, peptides). It encompasses all levels of cellular regulation, that is, the regulation that occurs at the level of transcription, translation, and post-translational modifications, hence, they can closely reflect the phenotype of an organism at specific time. The human metabolome is thought to be composed of about 3,000 endogenous metabolites at current estimates (Human metabolome project). But the exact size of human metabolome is still debatable. It is also believed that nutritional compounds, xenobiotics, and microbial metabolites must be considered when defining the human metabolome [94]. Therefore, metabolome analysis can be a useful tool used to find diagnostic markers that will help us examine unknown pathological conditions, effectively.

Different analytical techniques can be used in measurement of the metabolites. Such methods are nuclear magnetic resonance (NMR) spectrometry, molecular mass spectrometry (MS), gas chromatography (GC), high performance liquid chromatography (LC) and tricarboxylic acid (TCA). The most common and more high throughput technologies are nuclear magnetic resonance (NMR) spectrometry and molecular mass spectrometry (MS).

Mass spectrometry is an analytical platform for metabolomic analysis. It is a highly sensitive, reproductive, and versatile method as it identifies molecules and their fragments by measurng their masses. This information is obtained by measuring the mass–to–charge ratio (m/z) of ions that are produced by inducing the loss or gain of a charge from a neutral species. The sample, which is comprised of up of a complicated mixture of metabolites, can be introduced into the mass spectrometer either directly or preceded by a separation approach (using liquid chromatography or gas chromatography). [95]

NMR spectroscopy utilizes magnetic properties of nuclei to determine the number and type of chemical entities in a molecule. Proton NMR spectroscopy can detect soluble proton-containing

molecules with a molecular weight of approximately 20 kD or less. The NMR spectra serve as the raw material for pattern recognition analyses, which simplifies the complex multivariate data into 2 or 3 dimensions that can be readily understood and evaluated. Both NMR and liquid chromatography-mass spectrometry (LC-MS) systems can be integrated to in vivo tissues or to biological fluids such as serum, plasma, urine, etc., obtained from humans. The advantages of NMR are that it requires relatively little sample preparation, it is non-destructive, and can provide useful information regarding the exact structure of metabolites. However, NMR sensitivity is related to the magnet's strength, while available instrumentation can unambiguously detect only the most abundant metabolites in plasma. On the other hand, the most important advantage of mass spectrometry coupled with upfront chromatography is of far greater sensitivity than NMR MS-based systems that have been used to resolve compounds in the nanomole to picomole and even femtomole range, whereas identification of compounds by 1H-NMR requires concentrations of 1 nanomole or higher. [96-97]

The main methodologies that are used for metabolomic analysis are untargeted and targeted metabolomics. Untargeted metabolomics allow measuring a wider variety of metabolites present in an extracted sample without prior knowledge of the metabolome. The main advantage is that it provides with an unbiased way to examine the relationship between interconnected metabolites from multiple pathways. By contrast, targeted metabolomic analyses measure the concentrations of predefined set of metabolites and provides higher sensitivity and selectivity than untargeted metabolomics.

An overview of the main metabolomics associated with drug-induced cardiotoxicity detected in plasma/stem cell/heart in mice or people is given in Table 4. The role of carnitine in detection of cardiotoxicity was confirmed by a successive study in which Armenian et al compared a metabolomics analysis in 150 symptom-free childhood cancer survivors that receieved anthracycline treatment. Thirty-five participants were found to have cardiac dysfunction without symptoms. So, they compared the two groups (participants with cardiac dysfunction and with normal systolic function) and discovered 15 metabolites differentially expressed among patients. After adjusting for multiple comparisons, individuals with cardiotoxicity had significantly lower plasma carnitine levels in comparison with those with normal cardiac function. [98]

Table 4. Metabolomics associated with drugs-induced cardiotoxicity.

Metabolite	Plasma	Stem cell	Heart	Mice	People	XRT	Medicine	Dose	Biomarker	References
proline	↓ // ↑		↑	Yes	no		cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
LPC 20:3	↓			Yes	no		cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
linoleic acid	↓			Yes	no		cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
l-carnitine	↑ // ↑			Yes	no		cyclophosphamide/doxo/isoproterenol/5-fluorouracil	200mg/kg// 20mg/kg//5 mg/kg//125 mg/kg		Li et al, J Proteome Res, 2015
19-hydroxycorticosterone	↑ // ↓			Yes	no		cyclophosphamide/doxo/isoproterenol/5-fluorouracil	200mg/kg// 20mg/kg//5 mg/kg//125 mg/kg		Li et al, J Proteome Res, 2015

phytophingosine	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
cholid acid	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
LPC 14:0	↓ // ↓		Yes	no	cyclophosphamide/doxo/isoproterenol/5-fluorouracil	200mg/kg//20mg/kg//5mg/kg//125mg/kg		Li et al, J Proteome Res, 2015
LPC 18:3	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
LPC 16:1	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
LPE 18:2	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
LPC 22:5	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
LPC 22:6	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
LPC 22:4	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
LPC 20:2	↓ // ↓		Yes	no	cyclophosphamide/doxo/isoproterenol/5-fluorouracil	200mg/kg//20mg/kg//5mg/kg//125mg/kg		Li et al, J Proteome Res, 2015
PLE 20:3	↓		Yes	no	cyclophosphamide	200mg/kg		Li et al, J Proteome Res, 2015
pyruvate		↑			Doxorubicin	20mg/kg	troponine T LDH	Andreadou et al, NMR Biomed, 2009 /Chauhari et al, Amino Acids 2017
acetate		↑	↑	Yes	doxorubicin	20mg/kg	troponine T LDH	Andreadou et al, NMR Biomed, 2009 /Chauhari et al, Amino Acids 2017
formate		↑			Doxorubicin	20mg/kg	troponine T LDH	Andreadou et al, NMR Biomed, 2009

								/Chauhari et al, Amino Acids 2017
succinate		↑	↑	Yes		Doxorubicin	20mg/kg	troponine T LDH	Andreadou et al, NMR Biomed, 2009 /Chauhari et al, Amino Acids 2017
lactate	↑ // ↑		↓	Yes		Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
alanine	↑ // ↑		↑ // ↑	Yes		Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
glutamine	↑		↓	Yes		Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
glutamate	↑		no	Yes	↑	Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
creatine			no	Yes		Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
taurine			no	Yes	↓	Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
valine	↑		↓	Yes	↑	Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
leuline	↑		↓	Yes		Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
isoleukine	↑		↓	Yes	↑	Doxorubicin	20mg/kg	troponine T	Andreadou et al, NMR Biomed, 2009
carnitine	↓ // ↑		↓	Yes	**yes**	anthracyclines//doxorubicin		troponine T	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
threitol	↓				yes	anthracyclines			Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
mannose	↓				yes	anthracyclines			Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014

pyroglutamine	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
n-acetylalanine	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
creatine	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
eicosenoate	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
stearidonate	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
arachidonate	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
dihomo-linoleate	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
l-stearoylglcerophoinositol	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
dehydroisoandrosterone sulfate	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
pregnen-dio; disulfate	↓	yes	anthracyclines	Armenian et al, Cancer Epidemiol Biomarkers Prev. 2014
pregn steroid monosulfate	↓	yes	anthracyclines	Armenian et al, Cancer

								Epidemiol Biomarkers Prev. 2014
arginine	↑	↑	Yes		Doxorubicin			Schnackenberg et al, Appl. Toxicol. 2016
asparagine	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
citrulline	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
glycine	↑	↑	Yes	↑	Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
histidine	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
lysine	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
methionine	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
ornithine	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
phenylalanine	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
serine	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
threonine	↑	↑	Yes	↑	Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
trptophan	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
tyrosine	↑	↑	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
acetylornithine	↑	↓	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
hydroxproline	↑	no	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016
citrate	no	no	Yes		Doxorubicin		troponine T	Schnackenberg et al, Appl. Toxicol. 2016

propionylcarnitine	↑	no	Yes		Doxorubicin	troponine T	Schnackenberg et al, Appl. Toxicol. 2016
serotonine	no	↑	Yes		Doxorubicin	troponine T	Schnackenberg et al, Appl. Toxicol. 2016
putrescine	no	↑	Yes		Doxorubicin	troponine T	Schnackenberg et al, Appl. Toxicol. 2016
malate	↑	↑	Yes		Doxorubicin		Tan et al, PLoS One 2011
fructose		↑	Yes		Doxorubicin		Tan et al, PLoS One 2011
glycose		↑	Yes		Doxorubicin		Tan et al, PLoS One 2011
cholesterol		↑	Yes		Doxorubicin		Tan et al, PLoS One 2011
alanine		↑	Yes		Doxorubicin		Tan et al, PLoS One 2011
glutamine			Yes	↓	Doxorubicin		Tan et al, PLoS One 2011
docosahexaenoic acid		↓	Yes		Sunitinib		Jencen et al, Metabolites. 2017
arachidonic acid/eicosapetaenoic acid		↓	Yes		Sunitinib		Jencen et al, Metabolites. 2017
6-hydroxynicotinic acid		↓	Yes		Sunitinib		Jencen et al, Metabolites. 2017
o-phosphocolamine		↓	Yes		Sunitinib		Jencen et al, Metabolites. 2017
ethanolamine	↑		Yes		Sunitinib		Jencen et al, Metabolites. 2017
xenobiotics							

More recently, Li et al [99] identified 39 biomarkers for detecting cardiotoxicity earlier than biochemical analysis and histopathological assessment. They used rats to create cardiotoxicity models in which the toxicity was caused by doxorubicin, isoproterenol, and 5-fluorouracil. The metabolomics analysis of plasma was performed by using ultraperformance liquid chromatography quadrupole time-of-flight mass spectrometry. They used a support vector machine (SVM) trying to deploy a predictive model to confirm more exclusive biomarkers with more important l-carnitine, 19-hydroxydeoxycorticosterone, lysophosphatidylcholine (LPC) (14:0) and LPC (20:2). [99]

Similarly, Schnackenberg et al attempted to discover molecular markers of early stage of cardiotoxicity induced by doxorubicin in mice before the onset of cardiac damage. They discovered 18 metabolites significantly altered in plasma, and another 22 metabolites were increased in cardiac tissue after a cumulative dose of 6mg/kg, while myocardial injury and cardiac pathology were not noticed until after 18 and 24 mg/kg cumulative doses, respectively.[100] Metabolomics analyses of plasma and heart tissue showed significant variations in the levels of many amino acids (among

arginine and citrulline), biogenic amines, acylcarnitine's (carnitine) and tricarboxylic acid cycle (TCA)-related metabolites (lactate, succinate e.g.) .

Tan et al conducted a study using gas chromatography-mass spectrometry to describe the metabolic profile of doxorubicin-induced cardiomyopathy in mice. They indentified 24 metabolites, which were implicated in glycolysis, the citrate cycle and the metabolism of some amino acids and lipid and were selected as possible biomarkers for detection of cardiotoxicity. [101]

Andreadou et al used nuclear magnetic resonance (NMR) spectrometry to describe the metabolic profile of the acute doxorubicin cardiotoxicity in rats and to evaluate the metabolic alterations conferred by co- treatment with oleuropein.[90] The mice were divided into six groups: the first group included the control group, the second group received DOX, and the other four groups of mice received doxorubicin with oleuropein in a different dose and days, regarding oleuropein. Mice hearts were excised 72 hours after doxorubicin administration and H-NMR spectra of aqueous myocardium extracts were monitored. The results of analysis showed the increase of levels of acetate and succinate in DOX group compared to controls, while amino acids levels were lower. The conclusion of the article was that acetate and succinate constitute novel biomarkers for early detection cardiotoxicity. [102-103]

Geng et al in their study, used gas chromatography−mass spectrometry analysis of main targeted tissues (serum, heart, liver, brain, and kidney), to systemically evaluate the toxicity of DOX. Multivariate analyses revealed 21 metabolites in the serum, including cholesterol, D-glucose, D-lactic acid, glycine, L-alanine, L-glutamic acid, L-isoleucine, L-leucine, L-proline, L-serine, L-tryptophan, L-tyrosine, L-valine, N-methylphenylethanolamine, oleamide, palmitic acid, pyroglutamic acid, stearic acid, and urea, were changed in the serum in the DOX group. [104]

Tantawy et al identified lower plasma abundance of pyruvate and higher abundance of lactate in patients with carfilzomib-related cardiovascular adverse events. (CVAEs). They emphasized the significance of the pyruvate oxidation pathway associated with mitochondrial dysfunction. In order to better understand the mechanisms of Carfilzomib - CVAEs further investigation and validation is needed in a larger independent cohort. [105]

Yin et al proposed 15 different metabolites which play important role in cyclophosphamide induced cardiotoxicity. In this study, rat plasma samples were collected and analyzed one, three and five days after cyclophosphamide administration using ultra-performance liquid chromatography quadrupole time-of-flight mass spectrometry (UPLC-QqTOF HRMS). Of biomarkers studied, the proline, linoleic acid and glycerophospholipids changed significantly in three periods and the change associated with increasing time of occurrence of cardiotoxicity from cyclophosphamide. [106]

The study of Jensen et [107] al showed significant decreases in docosahexaenoic acid, arachidonic acid/ eicosatetraenoic acid, O-phosphocolamine and 6-hydroxynicotinic acid after sunitinib treatment with non-targeted metabolomics analysis of mice heart.[30] The same author, also, showed alterations in taurine/hypotaurine metabolism in the hearts and skeletal muscles of mice after sorafenib treatment. [108]

Except for the analysis of plasma and heart tissue, NMR spectroscopy-based metabolomics may detect low molecular weight metabolites in urine and cell culture media. For example, Chaudhari [109] et al showed reduction in the utilization of pyruvate and acetate, and accumulation of formate contrast to control culture medium of human induced pluripotent stem cell-derived cardiomyocytes exposed to doxorubicin. In contrast, Wang et al [110] showed in their study that tryptophan and phenylalanine metabolism in urine was also an important process in the systemic toxicity of doxorubicin. Also, Park et al identified 19 urinary metabolites in rats treated with doxorubicin. [111]

This technology is still under development, it seems obvious that metabolomics holds the potential to revolutionize our ability to profile samples in order to understand biological processes and find useful disease diagnostic biomarkers

Conclusions

Cardiovascular toxicity continues to be the major cause of drug failure during preclinical and clinical treatment models and contributes to drug withdrawal after approval. numerous medication

drugs that have been used frequently in adult clinical practice for a long time, have demonstrated potentially harmful effects on the heart in pediatric patients. The cardiotoxicity of these medications persists as a significant issue, having a negative impact on patients' quality of life as well as the overall survival. Several strategies for early detection of cardiotoxicity have been developed to reduce the numbers of patients with cardiac mortality and morbidity. Of importance, the biomarkers identified by the ''omics'' approach are considered new potential markers, especially in the scenario of diagnosis and risk stratification of acute coronary syndromes induced by chemotherapeutic drugs and may prove helpful in the early detection of anticancer cardiotoxicity.

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Abbreviations

ABC	Adenosine triphosphate-binding cassette transporter
ABCC2	ATP Binding Cassette Subfamily C Member 2
ACT	Anthracycline-related cardiotoxicity
ALL	Acute lymphoblastic leukemia
BNP	B-type natriuretic peptide
CBR	Carbonyl reductase
CELF4	CUGBP Elav-Like Family Member 4
CHF	Congestive heart failure
CP	Cyclophosphamide
CMR	Cardiac magnetic resonance imaging
CRP	C-reactive protein
CVAEs	Cardiovascular adverse events
DOX	Doxorubicin
ECG	Electrocardiography
EVs	Extracellular vesicles
GC	Gas chromatography
GDF-15	Growth/differentiation factor 15
GLS	Global systolic longitudinal myocardial strain
HER2	Human epidermal growth factor receptor 2
HF	Heart failure
IFO	Ifosfamide
LC	Liquid chromatography high performance
LC-MS	Liquid chromatography-mass spectrometry
LPC	Lysophosphatidylcholine
LV	Left verticular
LVD	Left ventricular dysfunction
LVEF	Left ventricular ejection fraction
miRNAs	MicroRNAs
mRNAs	Messenger RNAs
MPI	Myocardial perfusion imaging
MS	Molecular mass spectrometry
MUGA	Nuclear cardiac imaging
NMR	Nuclear magnetic resonance spectrometry
NOTCH1	Neurogenic locus notch homolog protein 1
NT-proBNP	N-terminal pro b-natriuretic peptide
PET	Position emission tomography
PYGB	Glycogen phosphorylase
RARG	Retinoic acid receptor gamma
SAL	Saline
SLC	Solute carrier transporters
SNP	Single-nucleotide polymorphism
SPECT	Single photon emission computed tomography
SVM	Vector machine
TKI	Tyrosine kinase inhibitors
TCA	Tricarboxylic acid

TnT	Troponin T
TOP2	Topoisomerase II
Top2β	Topoisomerase-II β
UGT1A6	Glucuronosyltransferase family
UPLC-QqTOF HRMS	Ultra-performance liquid chromatography quadrupole time-of-flight mass spectrometry
VEGF	Vascular endothelial growth factor
vWF	Von Willebrand factor
2D	Two-dimensional Echocardiography
5-FU	Fluorouracil

References

1. Pritchard-Jones K, Bergeron C, de Camargo B, van den Heuvel-Eibrink MM, Acha T, Godzinski J, Oldenburger F, Boccon-Gibod L, Leuschner I, Vujanic G, Sandstedt B, de Kraker J, van Tinteren H, Graf N; SIOP Renal Tumours Study Group. Omission of doxorubicin from the treatment of stage II-III, intermediate-risk Wilms' tumour (SIOP WT 2001): an open-label, non-inferiority, randomised controlled trial. Lancet. 2015 Sep 19;386(9999):1156-64.
2. Ampatzidou M., Kelaidi C., Dworzak M.N., Polychronopoulou S. Adolescents and young adults with acute lymphoblastic and acute myeloid leukemia. MEMO-Magazine of European Medical Oncology 2018; 11(1):47-53.
3. Ampatzidou M, Panagiotou JP, Paterakis G, Papadakis V, Papadimitriou SI, Parcharidou A, Papargyri S, Rigatou E, Avgerinou G, Tsitsikas K, Vasdekis V, Haidas S, Polychronopoulou S. Childhood acute lymphoblastic leukemia: 12 years of experience, using a Berlin-Frankfurt-Münster approach, in a Greek center. Leuk Lymphoma. 2015 Jan;56(1):251-5.
4. Sophia Polychronopoulou, Margarita Baka, Marina Servitzoglou, Vasilios Papadakis, Apostolos Pourtsidis, Georgia Avgerinou, Mirella Abatzidou, Helen Kosmidis. Treatment and clinical results in childhood AML in Greece. MEMO - Magazine of European Medical Oncology, February 2014, Volume 7, Issue 1, pp 50–55
5. Georgakis MK, Karalexi MA, Agius D, Antunes L, Bastos J, Coza D, Demetriou A, Dimitrova N, Eser S, Florea M, Ryzhov A, Sekerija M, Žagar T, Zborovskaya A, Zivkovic S, Bouka E, Kanavidis P, Dana H, Hatzipantelis E, Kourti M, Moschovi M, Polychronopoulou S, Stiakaki E, Kantzanou M, Pourtsidis A, Petridou ET. Incidence and time trends of childhood lymphomas: findings from 14 Southern and Eastern European cancer registries and the Surveillance, Epidemiology and End Results, USA. Cancer Causes Control. 2016 Nov;27(11):1381-1394.
6. Petridou ET, Dimitrova N, Eser S, Kachanov D, Karakilinc H, Varfolomeeva S, Belechri M, Baka M, Moschovi M, Polychronopoulou S, Athanasiadou-Piperopoulou F, Fragandrea I, Stiakaki E, Pourtsidis A, Stefanadis C, Coebergh JW, Sergentanis TN. Childhood leukemia and lymphoma: time trends and factors affecting survival in five Southern and Eastern European Cancer Registries. Cancer Causes Control. 2013 Jun;24(6):1111-8.
7. Lyon AR, López-Fernández T, Couch LS, Asteggiano R, Aznar MC, Bergler-Klein J, Boriani G, Cardinale D, Cordoba R, Cosyns B, Cutter DJ, de Azambuja E, de Boer RA, Dent SF, Farmakis D, Gevaert SA, Gorog DA, Herrmann J, Lenihan D, Moslehi J, Moura B, Salinger SS, Stephens R, Suter TM, Szmit S, Tamargo J, Thavendiranathan P, Tocchetti CG, van der Meer P, van der Pal HJH; ESC Scientific Document Group. 2022 ESC Guidelines on cardio-oncology developed in collaboration with the European Hematology Association (EHA), the European Society for Therapeutic Radiology and Oncology (ESTRO) and the International Cardio-Oncology Society (IC-OS). Eur Heart J. 2022 Nov 1;43(41):4229-4361.
8. Herrmann J, Lenihan D, Armenian S, Barac A, Blaes A, Cardinale D, Carver J, Dent S, Ky B, Lyon AR, López-Fernández T, Fradley MG, Ganatra S, Curigliano G, Mitchell JD, Minotti G, Lang NN, Liu JE, Neilan TG, Nohria A, O'Quinn R, Pusic I, Porter C, Reynolds KL, Ruddy KJ, Thavendiranathan P, Valent P. Defining cardiovascular toxicities of cancer therapies: an International Cardio-Oncology Society (IC-OS) consensus statement. Eur Heart J. 2022 Jan 31;43(4):280-299.
9. Chow EJ, Leger KJ, Bhatt NS, Mulrooney DA, Ross CJ, Aggarwal S, Bansal N, Ehrhardt MJ, Armenian SH, Scott JM, Hong B. Paediatric cardio-oncology: epidemiology, screening, prevention, and treatment. Cardiovasc Res. 2019 Apr 15;115(5):922-934
10. Madeddu C, Deidda M, Piras A, Cadeddu C, Demurtas L, Puzzoni M, Piscopo G, Scartozzi M, Mercuro G. Pathophysiology of cardiotoxicity induced by nonanthracycline chemotherapy. J Cardiovasc Med (Hagerstown). 2016 May;17 Suppl 1: S12-8.

11. Franco VI, Lipshultz SE. Cardiac complications in childhood cancer survivors treated with anthracyclines. Cardiol Young. 2015 Aug;25 Suppl 2:107-16.
12. Ward E, DeSantis C, Robbins A, et al. Childhood and adolescent cancer statistics, 2014. CA Cancer J Clin 2014; 64: 83–103.
13. Mulrooney DA, Yeazel MW, Kawashima T, et al. Cardiac outcomes in a cohort of adult survivors of childhood and adolescent cancer: retrospective analysis of the Childhood Cancer Survivor Study cohort. BMJ 2009; 339: b4606.
14. Bansal N, Amdani S, Lipshultz ER, Lipshultz SE. Chemotherapy-induced cardiotoxicity in children. Expert Opin Drug Metab Toxicol. 2017 Aug;13(8):817-832. Epub 2017 Jul 13.
15. Minotti G., Menna P, Salvatorelli E, Cairo G, Gianni L. Anthracyclines: molecular advances and pharmacologic developments in antitumor activity and cardiotoxicity. Pharmacol Rev. 2004;56(2):185-229
16. Sawyer DB, Peng X, Chen B, Pentassuglia L, Lim CC. Mechanisms of anthracycline cardiac injury: can we identify strategies for cardioprotection? Prog Cardiovasc Dis. 2010 Sep-Oct;53(2):105-13.
17. Simbre VC, Duffy SA, Dadlani GH, Miller TL, Lipshultz SE. Cardiotoxicity of cancer chemotherapy: implications for children. Paediatr Drugs. 2005;7(3):187-202.
18. Herrmann J, Lerman A, Sandhu NP, Villarraga HR, Mulvagh SL, Kohli M. Evaluation and management of patients with heart disease and cancer: cardio-oncology. Mayo Clin Proc. 2014 Sep;89(9):1287-306.
19. Sayed-Ahmed MM, Aldelemy ML, Al-Shabanah OA, Hafez MM, Al-Hosaini KA, Al-Harbi NO, Al-Sharary SD, Al-Harbi MM. Inhibition of gene expression of carnitine palmitoyltransferase I and heart fatty acid binding protein in cyclophosphamide and ifosfamide-induced acute cardiotoxic rat models. Cardiovasc Toxicol. 2014 Sep;14(3):232-42.
20. Rhea IB, Oliveira GH. Cardiotoxicity of Novel Targeted Chemotherapeutic Agents. Curr Treat Options Cardiovasc Med. 2018 Jun 19;20(7):53.
21. Zhang K, Heidrich FM, DeGray B, Boehmerle W, Ehrlich BE. Paclitaxel accelerates spontaneous calcium oscillations in cardiomyocytes by interacting with NCS-1 and the InsP3R. J Mol Cell Cardiol. 2010 Nov;49(5):829-35
22. Takahashi K, Inukai T, Imamura T, et al. Anti-leukemic activity of bortezomib and carfilzomib on B-cell precursor ALL cell lines. *PLoS One*. 2017;12(12): e0188680.
23. Shah C, Bishnoi R, Jain A, Bejjanki H, Xiong S, Wang Y, Zou F, Moreb JS. Cardiotoxicity associated with carfilzomib: systematic review and meta-analysis. Leuk Lymphoma. 2018 Nov;59(11):2557-2569.
24. Waxman AJ, Clasen S, Hwang W, et al. Carfilzomib-Associated Cardiovascular Adverse Events: A Systematic Review and Meta-analysis. *JAMA Oncol.* 2018;4(3): e174519.
25. Lipshultz SE, Karnik R, Sambatakos P, Franco VI, Ross SW, Miller TL. Anthracycline-related cardiotoxicity in childhood cancer survivors. Curr Opin Cardiol. 2014 Jan;29(1):103-12.
26. Manrique CR, Park M, Tiwari N, Plana JC, and Garcia MJ: Diagnostic strategies for early recognition of cancer therapeutics-related cardiac dysfunction. Clin Med Insights Cardiol 11: 1179546817697983, 2017.
27. Von Hoff DD, Layard MW, Basa P, Davis HL Jr, Von Hoff AL, Rozencweig M and Muggia FM: Risk factors for doxorubicin-induced congestive heart failure. Ann Intern Med 91: 710-717, 1979
28. 14 Nysom K, Holm K, Lipsitz SR, Mone SM, Co- lan SD, Orav EJ, Sallan SE, Olsen JH, Hertz H, Jacobsen JR, Lipshultz SE: Relationship be- tween cumulative anthracycline dose and late cardiotoxicity in childhood acute lympho- blastic leukemia. J Clin Oncol 1998; 16:545– 550
29. 15.Vandecruys E, Mondelaers V, De Wolf D, Benoit Y, Suys B: Late cardiotoxicity after low dose of anthracycline therapy for acute lymphoblastic leukemia in childhood. J Cancer Surviv 2012; 6:95–101.
30. 16 Lipshultz SE, Adams MJ: Cardiotoxicity after childhood cancer: beginning with the end in mind. J Clin Oncol 2010; 28:1276–1281.
31. 17 Van der Pal HJ, van Dalen EC, Hauptmann M, Kok WE, Caron HN, van den Bos C, Ol- denburger F, Koning CC, van Leeuwen FE, Kremer LC: Cardiac function in 5-year survivors of childhood cancer: a long-term follow- up study. Arch Intern Med 2010; 170:1247– 1255.
32. Brickler M, Raskin A, Ryan TD. Current State of Pediatric Cardio-Oncology: A Review. Children (Basel). 2022 Jan 19;9(2):127
33. Loar RW, Noel CV, Tunuguntla H, Colquitt JL, Pignatelli RH. State of the art review: Chemotherapy-induced cardiotoxicity in children. Congenit Heart Dis. 2018 Jan;13(1):5-15.

34. Mornoş C, Manolis AJ, Cozma D, Kouremenos N, Zacharopoulou I, Ionac A. The value of left ventricular global longitudinal strain assessed by three-dimensional strain imaging in the early detection of anthracyclinemediated cardiotoxicity. Hellenic J Cardiol. 2014 May-Jun;55(3):235-44.
35. Lipshultz SE, Miller TL, Scully RE, Lipsitz SR, Rifai N, Silverman LB, et al Changes in cardiac biomarkers during doxorubicin treatment of pediatric patients with high-risk acute lymphoblastic leukemia: associations with long-term echocardiographic outcomes., J Clin Oncol. 2012; 30:1042-9.
36. Mavinkurve-Groothuis AM, Groot-Loonen J, Bellersen L, Pourier MS, Feuth T, Bökkerink JP, Hoogerbrugge PM, Kapusta L. Abnormal NT-pro-BNP levels in asymptomatic long-term survivors of childhood cancer treated with anthracyclines. Pediatr Blood Cancer. 2009 May;52(5):631-6.
37. Sherief LM, Kamal AG, Khalek EA, Kamal NM, Soliman AA, Esh AM. Biomarkers and early detection of late onset anthracycline-induced cardiotoxicity in children. Hematology. 2012 May;17(3):151-6**.**
38. Dixon SB, Howell CR, Lu L, Plana JC, Joshi VM, Luepker RV, Durand JB, Ky B, Lenihan DJ, Jefferies JL, Green DM, Ehrhardt MJ, Mulrooney DA, Folse TE, Partin RE, Santucci AK, Howell RM, Srivastava DK, Hudson MM, Robison LL, Ness KK, Armstrong GT. Cardiac biomarkers and association with subsequent cardiomyopathy and mortality among adult survivors of childhood cancer: A report from the St. Jude Lifetime Cohort. Cancer. 2021 Feb 1;127(3):458-466.
39. Armenian SH, Gelehrter SK, Vase T, Venkatramani R, Landier W, Wilson KD, Herrera C, Reichman L, Menteer JD, Mascarenhas L, Freyer DR, Venkataraman K, Bhatia S. Screening for cardiac dysfunction in anthracycline-exposed childhood cancer survivors. Clin Cancer Res. 2014 Dec 15;20(24):6314-23.
40. Lam E, Higgins V, Zhang L, Chan MK, Bohn MK, Trajcevski K, Liu P, Adeli K, Nathan PC. Normative Values of High-Sensitivity Cardiac Troponin T and N-Terminal pro-B-Type Natriuretic Peptide in Children and Adolescents: A Study from the CALIPER Cohort. J Appl Lab Med. 2021 Mar 1;6(2):344-353.
41. Dhssraj Singh, Akanksha Thakur, and W. H. Wilson Tang, Kaufman Utilizing Cardiac Biomarkers to Detect and Prevent Chemotherapy-induced Cardiomyopathy, Clin Biochem. 2015 March; 48(0): 223–235.
42. Eric S. Christensona, Theodore Jamesa, Vineet Agrawala, and Ben H. ParkbaJohns, Use of biomarkers for the assessment of chemotherapy-induced cardiac toxicity, Curr Heart Fail Rep. 2015 June; 12(3): 255–262.
43. Daniela Cardinale, Gina Biasillo, Michela Salvatici, Maria Teresa Sandri & Carlo Maria Cipolla, using biomarkers to predict and to prevent cardiotoxicity of cancer therapy, Journal: Expert Review of Molecular Diagnostics,
44. Horacek JM, Jebavy L, Vasatova M, Pudil R, Tichy M, Jakl M, Maly J. Glycogen phosphorylase BB as a potential marker of cardiac toxicity in patients treated with anthracyclines for acute leukemia., Bratisl Lek Listy. 2013;114(12):708-10.
45. Horacek JM1, Vasatova M, Tichy M, Pudil R, Jebavy L, Maly J. The use of cardiac biomarkers in detection of cardiotoxicity associated with conventional and high-dose chemotherapy for acute leukemia. Exp Oncol. 2010 Jul;32(2):97-9.
46. Horacek JM1, Tichy M, Jebavy L, Pudil R, Ulrychova M, Maly Use of multiple biomarkers for evaluation of anthracycline-induced cardiotoxicity in patients with acute myeloid leukemia. J. Exp Oncol. 2008 Jun;30(2):157-9.,
47. Horacek JM1, Vasatova M, Pudil R, Tichy M, Zak P, Jakl M, Jebavy L, Maly J. Biomarkers for the early detection of anthracycline-induced cardiotoxicity: current status. Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub. 2014 Dec;158(4):511-7. Epub 2014 Jan 23
48. Cao L, Zhu W, Wagar EA, Meng QH. Biomarkers for monitoring chemotherapy-induced cardiotoxicity. Crit Rev Clin Lab Sci. 2017 Mar;54(2):87-101. Epub 2016 Dec 26.
49. Arslan D1, Cihan T, Kose D, Vatansev H, Cimen D, Koksal Y, Oran B, Akyurek F Growth-differentiation factor-15 and tissue doppler ımaging in detection of asymptomatic anthracycline cardiomyopathy in childhood cancer survivors. Clin Biochem. 2013 Sep;46(13-14):1239-43. Epub 2013 Jul 11
50. Armenian S, Bhatia S. Predicting and Preventing Anthracycline-Related Cardiotoxicity. Am Soc Clin Oncol Educ Book. 2018 May 23; 38:3-12.
51. Aminkeng F, Bhavsar AP, Visscher H, Rassekh SR, Li Y, Lee JW, Brunham LR, Caron HN, van Dalen EC, Kremer LC, van der Pal HJ, Amstutz U, Rieder MJ, Bernstein D, Carleton BC, Hayden MR, Ross CJ, Canadian Pharmacogenomics Network for Drug Safety Consortium. A coding variant in RARG confers susceptibility to anthracycline-induced cardiotoxicity in childhood cancer. Nat Genet. 2015 Sep;47(9):1079-84.

52. Madonna R (2017) Early diagnosis and prediction of anticancer drug-induced cardiotoxicity: from cardiac imaging to "Omics" technologies. Rev Espanol Cardiol 70(7):576–582.
53. Linschoten M, Teske AJ, Cramer MJ, van der Wall E, Asselbergs FW., Chemotherapy-Related Cardiac Dysfunction: A Systematic Review of Genetic Variants Modulating Individual Risk. Circ Genom Precis Med. 2018 Jan;11(1): e001753
54. H. Visscher, MD,C.J.D. Ross, PhD, S.R. Rassekh, MD, MHSc, G.S.S. Sandor, MB, ChB, H.N. Caron, MD, PhD,E.C. van Dalen, MD, PhD,L.C. Kremer, MD, PhD,H.J. van der Pal, MD, P.C. Rogers, MB, ChB, MBA, M.J. Rieder, MD, PhD, B.C. Validation of Variants in SLC28A3 and UGT1A6 as Genetic Markers Predictive of Anthracycline-Induced Cardiotoxicity in Children, Carleton, PharmD,M.R. Hayden, MB, ChB, PhD, and the CPNDS Consortium, Pediatr Blood Cancer 2013;60:1375–1381
55. Visscher H, Rassekh SR, Sandor GS, et al; CPNDS Consor◉um. Gene◉c variants in SLC22A17 and SLC22A7 are associated with anthracycline- induced cardiotoxicity in children. *Pharmacogenomics*. 2015; 16:1065-1076.
56. Henk Visscher, Colin J.D. Ross, S. Rod Rassekh, Amina Barhdadi, Marie-Pierre Dube´, Hesham Al-Saloos, S. Sandor, Huib N. Caron, Elvira C. van Dalen, Leontien C. Kremer, Helena J. van der Pal, Andrew M.K. Brown, Paul C. Rogers, Michael S. Phillips, Michael J. Rieder, Bruce C. Carleton, Michael R. Hayden, and Canadian Pharmacogenomics Network for Drug Safety Consortium, Pharmacogenomic Prediction of Anthracycline-Induced Cardiotoxicity in Children, J Clin Oncol 30:1422-1428.
57. Sági JC, Egyed B, Kelemen A, Kutszegi N, Hegyi M, Gézsi A, Herlitschke MA, Rzepiel A, Fodor LE, Ottóffy G, Kovács GT, Erdélyi DJ, Szalai C, Semsei ÁF. Possible roles of genetic variations in chemotherapy related cardiotoxicity in pediatric acute lymphoblastic leukemia and osteosarcoma. BMC Cancer. 2018 Jul 3;18(1):704
58. 14.Folefac Aminkeng, Colin J. D. Ross, Shahrad R. Rassekh, Soomi Hwang, Michael J. Rieder, Amit P. Bhavsar, Anne Smith, Shubhayan Sanatani, Karen A. Gelmon, Daniel Bernstein, Michael R. Hayden Ursula Amstutz, Bruce C. Carleton2, and CPNDS Clinical Practice Recommendations Group, Recommendations for genetic testing to reduce the incidence of anthracycline-induced cardiotoxicity, Br J Clin Pharmacol (2016) 82 683–695
59. Marcoux S, Drouin S, Laverdiere C, Alos N, Andelfinger GU, Bertout L, et al. The PETALE study: Late adverse effects and biomarkers in childhood acute lymphoblastic leukemia survivors. Pediatr Blood Cancer. 2017;64(6)
60. Kashyap V, Laursen KB, Brenet F, Viale AJ, Scandura JM, Gudas LJ. RARgamma is essential for retinoic acid induced chromatin remodeling and transcriptional activation in embryonic stem cells. J Cell Sci 2013; 126: 999–1008.
61. Krajinovic M, Elbared J, Drouin S, Bertout L, Rezgui A, Ansari M, et al. Polymorphisms of ABCC5 and NOS3 genes influence doxorubicin cardiotoxicity in survivors of childhood acute lymphoblastic leukemia. Pharmacogenomics J 2015.
62. Semsei AF, Erdelyi DJ, Ungvari I, Csagoly E, Hegyi MZ, Kiszel PS, et al. ABCC1 polymorphisms in anthracycline- induced cardiotoxicity in childhood acute lymphoblastic leukaemia. Cell Biol Int. 2012;36(1):79-86.
63. Wojnowski L, Kulle B, Schirmer M, Schlüter G, Schmidt A, Rosenberger A, Vonhof S, Bickeböller H, Toliat MR, Suk EK, Tzvetkov M, Kruger A, Seifert S, Kloess M, Hahn H, Loeffler M, Nürnberg P, Pfreundschuh M, Trümper L, Brockmöller J, Hasenfuss G. NAD(P)H oxidase and multidrug resistance protein genetic polymorphisms are associated with doxorubicin-induced cardiotoxicity. Circulation. 2005 Dec 13;112(24):3754-62. Epub 2005 Dec 5.
64. Vulsteke C, Pfeil AM, Maggen C, Schwenkglenks M, Pettengell R, Szucs TD, et al. Clinical and genetic risk factors for epirubicin-induced cardiac toxicity in early breast cancer patients. Breast cancer research and treatment. 2015;152(1):67-76.
65. Blanco JG, Leisenring WM, Gonzalez-Covarrubias VM, Kawashima TI, Davies SM, Relling MV, et al. Genetic polymorphisms in the carbonyl reductase 3 gene CBR3 and the NAD(P)H: quinone oxidoreductase 1 gene NQO1 in patients who developed anthracycline-related congestive heart failure after childhood cancer. Cancer 2008; 112: 2789–95.
66. Blanco JG, Sun CL, Landier W, et al. Anthracycline-related cardiomyopathy a◉er childhood cancer: role of polymorphisms in carbonyl reductase genes—a report from the Children's Oncology Group. *J Clin Oncol*. 2012; 30:1415-1421

67. Wang X, Sun CL, Quiñones-Lombraña A, et al. CELF4 variant and anthracycline-related cardiomyopathy: a Children's Oncology Group genome-wide associa◉on study. *J Clin Oncol*. 2016; 34:863-870.
68. Vos HI, Coenen MJ, Guchelaar HJ, Te Loo DM. The role of pharmacogenetics in the treatment of osteosarcoma. Drug Discov Today. 2016 Nov;21(11):1775-1786.
69. Leong SL, Chaiyakunapruk N, Lee SW. Candidate Gene Association Studies of Anthracycline-induced Cardiotoxicity: A Systematic Review and Meta-analysis. Sci Rep. 2017 Feb 27;7(1):39.
70. McOwan TN, Craig LA, Tripdayonis A, Karavendzas K, Cheung MM, Porrello ER, Conyers R, Elliott DA. Evaluating anthracycline cardiotoxicity associated single nucleotide polymorphisms in a paediatric cohort with early onset cardiomyopathy. Cardiooncology. 2020 May 21; 6:5.
71. Petrykey K, Andelfinger GU, Laverdière C, Sinnett D, Krajinovic M. Genetic factors in anthracycline-induced cardiotoxicity in patients treated for pediatric cancer. Expert Opin Drug Metab Toxicol. 2020 Oct;16(10):865-883
72. Lipshultz SE, Lipsitz SR, Kutok JL, et al. Impact of hemochromatosis gene mutations on cardiac status in doxorubicin-treated survivors of childhood high-risk leukemia. *Cancer*. 2013;119(19):3555-3562.
73. Oatmen, Olga H Toro-Salazar, Kristine Hauser, Kia N Zellars, Kathryn C Mason, Kan Hor, Eileen Gillan, Caroline J Zeiss, Daniel M Gatti, Francis G Spinale, Identification of a Novel microRNA Profile in Pediatric Patients with Cancer Treated with Anthracycline Chemotherapy, Am J Physiol Heart Circ Physiol, 2018 Nov 1;315(5):H1443-H1452.
74. Oliveira-Carvalho V, Ferreira LRP, Bocchi EA (2015) Circulating mir-208a fails as a biomarker of doxorubicin-induced cardiotoxicity in breast cancer patients. J Appl Toxicol 35(9):1071–1072.
75. Oikonomou E1, Siasos G, Tousoulis D, Kokkou E, Genimata V, Zisimos K, Latsios G, Stefanadis C. Diagnostic, and therapeutic potentials of microRNAs in heart failure, Curr Top Med Chem. 2013;13(13):1548-58
76. Papageorgiou N1, Tousoulis D, Androulakis E, Siasos G, Briasoulis A, Vogiatzi G, Kampoli AM, Tsiamis E, Tentolouris C, Stefanadis C, The role of microRNAs in cardiovascular disease. Curr Med Chem. 2012;19(16):2605-10
77. Ruggeri C, Gioffre S, Achilli F, Colombo GI, D'Alessandra Y (2018) Role of microRNAs in doxorubicin-induced cardiotoxicity: an overview of preclinical models and cancer patients. Heart Fail Rev 23(1):109–122.
78. Holmgren G, Synnergren J, Andersson CX, Lindahl A, Sartipy P (2016) MicroRNAs as potential biomarkers for doxorubicininduced cardiotoxicity. Toxicol In Vitro 34:26–34.
79. Ludwig N, Leidinger P, Becker K et al (2016) Distribution of miRNA expression across human tissues. Nucleic Acids Res 44(8):3865– 3877.
80. Skála Mikulas, · Barbora Hanousková · Lenka Skálová · Petra Matoušková
81. MicroRNAs in the diagnosis and prevention of drug-induced Cardiotoxicity, Archives of Toxicology, November 2018
82. Shan-shan ZHOU, Jing-peng JIN Ji-qun WANG, Zhi-guo ZHANG1, Jonathan H FREEDMAN4, Yang ZHENG1, Lu CAI, MiRNAS in cardiovascular diseases: potential biomarkers, therapeutic targets and challenges, Acta Pharmacol Sin. 2018 Jul;39(7):1073-1084.Epub 2018 Jun 7
83. Pellegrini L, Sileno S, D'Agostino M, Foglio E, Florio MC, Guzzanti V, Russo MA, Limana F, Magenta A. MicroRNAs in Cancer Treatment-Induced Cardiotoxicity. Cancers (Basel). 2020 Mar 17;12(3):704.
84. Kuwabara Y, Ono K, Horie T et al (2011) Increased MicroRNA-1 and MicroRNA-133a levels in serum of patients with cardiovascular disease indicate myocardial damage. Circ Cardiovasc Genet 4(4):446–454.
85. Rigaud VOC, Ferreira LRP, Ayub-Ferreira SM et al (2017) Circulating miR-1 as a potential biomarker of doxorubicin-induced cardiotoxicity in breast cancer patients. Oncotarget 8(4):6994–7002.
86. Leger KJ, Leonard D, Nielson D, de Lemos JA, Mammen PP, Winick NJ. Circulating microRNAs: Potential Markers of Cardiotoxicity in Children and Young Adults Treated with Anthracycline Chemotherapy. J Am Heart Assoc. 2017 Apr 528 4;6(4).
87. Suresh N Kumar 1, Eugene A Konorev, Deepika Aggarwal, Balaraman Kalyanaraman, Analysis of Proteome Changes in Doxorubicin-Treated Adult Rat Cardiomyocyte, J Proteomics 2011 May 1;74(5):683-97. Epub 2011 Feb 19.
88. Creemers EE, Tijsen AJ, Pinto YM. Circulating microRNAs: novel biomarkers and extracellular communicators in cardiovascular disease? Circ Res. 2012 Feb 3;110(3):483-95.

89. Todorova VK, Makhoul I, Wei JN, Klimberg VS (2017) Circulating miRNA profiles of doxorubicin-induced cardiotoxicity in breast cancer patients. Ann Clin Lab Sci 47(2):115–119
90. Zhao ZY, He J, Zhang J et al (2014) Dysregulated miR1254 and miR579 for cardiotoxicity in patients treated with bevacizumab in colorectal cancer. Tumor Biol 35(6):5227–5235.
91. Emanuel F Petricoin, Vinodh Rajapaske, Eugene H Herman, Ali M Arekani, Sally Ross, Donald Johann, Alan Knapton, J Zhang, Ben A Hitt, Thomas P Conrads, Timothy D Veenstra, Lance A Liotta, Frank D Sistare, Toxicoproteomics: Serum Proteomic Pattern Diagnostics for Early Detection of Drug Induced Cardiac Toxicities and Cardioprotection, Toxicol Pathol, Mar-Apr 2004;32 Suppl 1:122-30.
92. Kaname Ohyama, Mari Tomonari, Tomoko Ichibangase, Hideto To, Naoya Kishikawa, Kenichiro Nakashima, Kazuhiro Imai, Naotaka Kuroda, A Toxicoproteomic Study on Cardioprotective Effects of Pre-Administration of Docetaxel in a Mouse Model of Adriamycin-Induced Cardiotoxicity, Biochem Pharmacol. 2010 Aug 15;80(4):540-7. Epub 2010 May 10.
93. Varsha G Desai, Taewon Lee, Carrie L Moland, Vikrant Vijay, Tao Han, Sherry M Lewis, Eugene H Herman, James C Fuscoe, Candidate Early Predictive Plasma Protein Markers of Doxorubicin-Induced Chronic Cardiotoxicity in B6C3F 1 Mice, Toxicol Appl Pharmacol. 2019 Jan 15; 363:164-173. Epub 2018 Dec 2.
94. Chontida Yarana, Dustin Carroll, Jing Chen, Luksana Chaiswing, Yanming Zhao, Teresa Noel, Michael Alstott, Younsoo Bae, Emily V Dressler, Jeffrey A Moscow, D Allan Butterfield, Haining Zhu, Daret K St Clair, Extracellular Vesicles Released by Cardiomyocytes in a Doxorubicin-Induced Cardiac Injury Mouse Model Contain Protein Biomarkers of Early Cardiac Injury, Clin Cancer Res ,2018 Apr 1;24(7):1644-1653. Epub 2017 Oct 25.
95. Wederson M. Claudino a, Priscila H. Goncalves b, Angelo di Leo c, Philip A. Philip b, Fazlul H. Sarkar d, Metabolomics in cancer: A bench-to-bedside intersection Critical Reviews in Oncology/Hematology 84 (2012) 1–7
96. Johnson CH, Ivanisevic J, Siuzdak G. Metabolomics: beyond biomarkers and towards mechanisms. Nat Rev Mol Cell Biol. 2016 Jul;17(7):451-9. doi: 10.1038/nrm.2016.25. Epub 2016 Mar 16.
97. Lewis GD, Asnani A, Gerszten RE. Application of metabolomics to cardiovascular biomarker and pathway discovery. J Am Coll Cardiol. 2008 Jul 8;52(2):117-23.
98. Deidda M, Mercurio V, Cuomo A, Noto A, Mercuro G, Cadeddu Dessalvi C. Metabolomic Perspectives in Antiblastic Cardiotoxicity and Cardioprotection. Int J Mol Sci. 2019 Oct 4;20(19):4928.
99. Saro H. Armenian, Sarah K. Gelehrter, Tabitha Vase, Rajkumar Venkatramani, Wendy Landier, Karla D. Wilson, Claudia Herrera, Leah Reichman, John-David Menteer, Leo Mascarenhas, David R. Freyer, Kalyanasundaram Venkataraman, Smita Bhatia, Carnitine and cardiac dysfunction in childhood cancer survivors treated with anthracyclines Cancer Epidemiol Biomarkers Prev. 2014 Jun; 23(6): 1109–1114.
100. Yubo Li, Liang Ju, Zhiguo Hou, Haoyue Deng, Zhenzhu Zhang, Lei Wang, Zhen Yang, Jia Yin, and Yanjun Zhang, Screening, Verification, and Optimization of Biomarkers for Early Prediction of Cardiotoxicity Based on Metabolomics, J Proteome Res, 2015 Jun 5;14(6):2437-45.
101. Schnackenberg L.K., Pence L., Vijay V., Moland C.L., George N., Cao Z., Yu L.R., Fuscoe J.C., Beger R.D., Desai V.G J. Early metabolomics changes in heart and plasma during chronic doxorubicin treatment in B6C3F1mice. Appl. Toxicol. 2016; 36:1486–1495.
102. Guangguo Tan, Ziyang Lou, Wenting Liao, Zhenyu Zhu, Xin Dong, Wei Zhang, Wuhong LiYifeng Chai, Potential Biomarkers in Mouse Myocardium of Doxorubicin-Induced Cardiomyopathy: A Metabonomic Method and Its Application. PLoS One 6(11): e27683.
103. Andreadou I, Papaefthimiou M, Constantinou M, Sigala F, Skaltsounis AL, Tsantili-Kakoulidou A, Iliodromitis EK, Kremastinos DT, Mikros E., Metabolomic identification of novel biomarkers in doxorubicin cardiotoxicity and protective eggect of the natural antioxidant oleuropein, NMR Biomed, 2009 Jul;22(6):585-92.
104. Andreadou I., Mikros E., Ioannidis K., Sigala F., Naka K., Kostidis S., Farmakis D., Tenta R., Kavantzas N., Bibli S.I., et al. J. Mol. Cell. Oleuropein prevents doxorubicin-induced cardiomyopathy interfering with signaling molecules and cardiomyocyte metabolism. Cardiol. 2014; 69:4–16.
105. Geng C, Cui C, Wang C, et al. Systematic Evaluations of Doxorubicin-Induced Toxicity in Rats Based on Metabolomics. *ACS Omega*. 2020;6(1):358-366. Published 2020 Dec 28.

106. Tantawy M, Chekka LM, Huang Y, et al. Lactate Dehydrogenase B and Pyruvate Oxidation Pathway Associated with Carfilzomib-Related Cardiotoxicity in Multiple Myeloma Patients: Result of a Multi-Omics Integrative Analysis. *Front Cardiovasc Med*. 2021; 8:645122.
107. Yin J., Xie J., Guo X., Ju L., Li Y., Zhang Y. J. Chromatogr. B Anal. Plasma metabolic profiling analysis of cyclophosphamide-induced cardiotoxicity using metabolomics coupled with UPLC/Q-TOF-MS and ROC curve. Technol. Biomed. Life Sci. 2016; 1033:428–435.
108. Jensen BC, Parry TL, Huang W, Ilaiwy A, Bain JR, Muehlbauer MJ, O'Neal SK, Patterson C, Johnson GL, Willis MS. Non-Targeted Metabolomics Analysis of the Effects of Tyrosine Kinase Inhibitors Sunitinib and Erlotinib on Heart, Muscle, Liver and Serum Metabolism In Vivo, Metabolites. 2017 Jun 22;7(3). pii: E31.
109. Jensen B.C., Parry T.L., Huang W., Beak J.Y., Ilaiwy A., Bain J.R., Newgard C.B., Muehlbauer M.J., Patterson C., Johnson G.L., et al Br. J Effects of the kinase inhibitor sorafenib on heart, muscle, liver, and plasma metabolism in vivo using non-targeted metabolomics analysis. Pharmacol. 2017; 174:4797–4811.
110. Umesh Chaudhari, James K. Ellis, Vilas Wagh, Harshal Nemade, Jürgen Hescheler, Hector C. Keun, Agapios Sachinidis, Metabolite signatures of doxorubicin induced toxicity in human induced pluripotent stem cell-derived cardiomyocytes, Amino Acids (2017) 49:1955–1963
111. Wang X, Sun CL, Quiñones-Lombraña A, Singh P, Landier W, Hageman L, Mather M, Rotter JI, Taylor KD, Chen YD, Armenian SH, Winick N, Ginsberg JP, Neglia JP, Oeffinger KC, Castellino SM, Dreyer ZE, Hudson MM, Robison LL, Blanco JG, Bhatia S. CELF4 Variant and Anthracycline-Related Cardiomyopathy: A Children's Oncology Group Genome-Wide Association Study. J Clin Oncol. 2016 Mar 10;34(8):863-70. Epub 2016 Jan 25.
112. Park B, Sim SH, Lee KS, Kim HJ, Park IH. Genome-wide association study of genetic variants related to anthracycline-induced cardiotoxicity in early breast cancer. Cancer Sci. 2020 Jul;111(7):2579-2587 Epub 2020 Jun 20
113. Rochette L, Guenancia C, Gudjoncik A, Hachet O, Zeller M, Cottin Y, Vergely C. Anthracyclines/trastuzumab: new aspects of cardiotoxicity and molecular mechanisms. Trends Pharmacol Sci. 2015 Jun;36(6):326-48