University of Alberta

Cardiotoxicity of Novel Cancer Therapies

by

Edith Pituskin

A thesis submitted to the Faculty of Graduate Studies and Research
in partial fulfillment of the requirements for the degree of

Doctor of Philosophy

in

Rehabilitation Science

Faculty of Rehabilitation Medicine

Fall 2013
Edmonton, Alberta

Dedication

This work is dedicated to the participants of these studies, whose generosity and resilience during a most difficult time makes their contribution to this and other research projects that much more admirable.

Abstract

Breast cancer is the most common malignancy among females, and the second leading cause of cancer death among Canadian women. Despite improved survival, both short and long term detrimental effects have been observed with both novel and conventional anti-cancer therapies. In particular, the effects of cancer therapy on the cardiovascular system, or 'cardiotoxicity', are increasingly recognized in terms of morbidity and mortality. Subtle or 'sub-clinical' early effects of anti-cancer therapies on cardiac structures may remain undetected, conveying potentially devastating longer-term effects. The overarching theme of this thesis is to examine the short and long-term effects of a novel anti-cancer therapy, trastuzumab, on left ventricular (LV) morphology and function in breast cancer patients.

Two studies were undertaken; study #1 was a cross-sectional study of 17 female breast cancer survivors exposed to trastuzumab-based chemotherapy, examining LV remodeling and exercise capacity. We found that early breast cancer patients had significant alterations in left ventricular geometry and impairment of cardiorespiratory function four years following exposure to trastuzumab-based chemotherapy. The second study was performed to examine the effects of three months of standard heart failure therapy, consisting of angiotensin-converting enzyme inhibitor or beta-blocker vs placebo, on LV remodeling in breast cancer patients receiving trastuzumab-based therapy. The major finding was that that standard heart failure pharmacotherapy appeared to be effective in attenuating LV dysfunction associated with trastuzumab-based

therapy. When compared to patients randomized to the pharmacotherapy group, significant decline in left ventricular ejection fraction occurred in the placebo group.

Acknowledgements

I am incredibly indebted to my supervisor Dr Mark Haykowsky, for his energy, his curiosity, his inspiration and his guidance. Mark's enthusiasm was instrumental in my decision to pursue doctoral work in Rehabilitation Medicine, and he facilitated multiple unique opportunities to enhance my learning along the way. Many many thanks, Mark!

I owe enormous gratitude to my committee members Drs Ian Paterson, John Mackey, Richard Thompson and Margie McNeely, not only for their time commitment and support of this work, but the important clinical and research collaborations that will undoubtedly improve patient care in the future. And special appreciation to Dr Kristen Campbell, for her insightful review of this thesis and her willingness to contribute in this final step of my program.

Also thanks to Dr Lee Jones for being Mark's co-conspirator in influencing me to pursue this degree, and for the learning opportunities afforded during my experience at Duke University. And to my friends, family and colleagues, many thanks for your support during what, at times, seemed a solitary journey. Your kindness and caring meant more than words can say.

The studies in this thesis were supported in part by awards from the Canadian Institutes of Health Research (CIHR) Strategic Training Program in TORCH (Tomorrow's Research Cardiovascular Health Professionals) and Alberta Cancer Research Institute (ACRI) Graduate Studentship to Edith Pituskin; CIHR Operating Grant, Alberta Cancer Foundation High Risk for High Return Grant,

Alberta Cancer Research Institute, and Alberta Cancer Foundation/Roche Canada Investigator-Initiated Research Program to Ian Paterson.

Table of Contents

List of Tables

List of Figures

Symbols, nomenclature, or abbreviations

ADH – patients who adhered to at least 80% of prescribed exercise sessions

ACEI – angiotensin-converting enzyme inhibitor

ARB - Angiotensin-receptor blocking agent

BB – beta-blocker

BMI – body mass index

BNP – brain natriuretic peptide

BP – blood pressure

CCI – Cross Cancer Institute, Edmonton Alberta CANADA

cMRI – cardiac MRI

CKO – conditional knock-out

CTCAE - Common Terminology Criteria for Adverse Events

DBP – diastolic blood pressure

EBC – early breast cancer

ECHO – trans-thoracic echocardiogram

EF – ejection fraction

EPICORE - Epidemiology Coordinating and Research Centre, University of Alberta

erbB2 – see HER2

FS – fractional shortening

GFR – glomerular filtration rate

HER2, HER2*neu*, erbB2 – human epidermal growth factor receptor 2, a plasma membrane-bound receptor tyrosine kinase, that interacts with many intracellular signaling molecules.

HF – heart failure

KPS – Karnofsky Performance Status

LV – left ventricle

LVEDV – left ventricular end-diastolic volume

LVEF – left ventricular ejection fraction

LVESV – left ventricular end-systolic volume

MI – myocardial infarction

MACE – major adverse cardiac event

MAHI – Mazankowski Alberta Heart Institute, Edmonton Alberta CANADA

MANTICORE – Multidisciplinary Approach to Novel Therapies In Cardiology Oncology
Research study

MRI – magnetic resonance imaging

MUGA – radionuclide ventriculography

NAD – patients who did not adhere to at least 80% of prescribed exercise sessions

PI3K/Akt (phosphatidylinositol 3 kinase-Akt) - an intracellular signaling pathway that plays a key role in transmitting proliferative signals from membrane bound receptors

PO – power output

PWth – posterior wall thickness of the left ventricle

Ras/Raf/MEK/MAPK – an intracellular signaling pathway that plays a key role in transmitting proliferative signals from membrane bound receptors

RPE - rate of perceived exertion

SEPth – septal wall thickness of the left ventricle

SBP – systolic blood pressure

RCT – randomized controlled trial

TITAN - Multidisciplinary Team IntervenTion in CArdio-ONcology study

VEGF – a signaling protein produced by cells that stimulates new blood vessel development

VO_2 - oxygen consumption measured during cardiopulmonary exercise testing

VO_{2peak} – peak cardiopulmonary exercise testing

WT – wild-type

Chapter 1

Introduction and Purpose

1.1 Introduction

Forty-five percent of men and 39% of women will be diagnosed with a malignancy at some point in their life(1). Given improvements in the understanding of cancer biology, improved detection and the use of novel adjuvant therapies, increasing numbers of Canadians survive a cancer diagnosis. Notably, the number of cancer survivors is increasing at twice the rate of new cancer diagnoses(2). These statistics confirm that cancer may be a 'chronic' disease that many Canadians will experience and survive, however a chronic condition that often requires complex medical management and care coordination over many years.

Breast cancer is the most common malignancy among females, and the second leading cause of cancer death among Canadian women(1). Research in tumor growth-signaling pathways has led to the discovery of human epidermal growth factor receptor 2 (HER2 or, HER2*neu*), a member of a trans-membrane tyrosine kinase receptor family including HER1, HER3 and HER4. These receptors have multiple activities including cell growth and survival regulation, adhesion, migration and differentiation(3). With the exception of HER3, each receptor has an extracellular binding domain and an intracellular tyrosine kinase domain(4). With activation, the signaling cascades of these receptors promote cellular proliferation and survival via transcription factor nuclear factor –kappa

beta and the phosphatidylinositol 3 kinase-Akt (PI3K-Akt) kinase signaling pathway. “Over-expression” of HER2 is observed in approximately 20% of all breast malignancies; rather than two copies of the gene per cell, as many as 50 – 100 gene copies per cell may be present. As a result, kinase activity is greatly enhanced, resulting in greatly increased cell signaling, proliferation and potential to metastasize to other regions of the body(5). Accordingly, tumor HER2 over-expression is understood as a predictor of disease relapse and shorter survival(6); as such, HER2 inhibition has presented an attractive target for cancer drug therapy.

Consequently, trastuzumab (Herceptin®) a humanized monoclonal antibody was specifically developed. In combination with chemotherapy, women receiving first-line treatment for metastatic breast cancer experienced significantly longer time to disease progression, higher rate of response, longer duration of response, and improved overall survival(7). This was the pivotal study demonstrating the effectiveness of trastuzumab, and also the first clinical experience revealing the potential for significant cardiotoxicity associated with trastuzumab therapy. Clinical signs and symptoms of heart failure (HF) occurred in 27% of the group given anthracycline, cyclophosphamide, and trastuzumab and 13 percent of the group given paclitaxel and trastuzumab. As a result, activities of cardiac HER2 attracted attention once these deleterious effects became evident. Notably, the signaling proteins and pathways affected by trastuzumab (transcription factor nuclear factor –κB and PI3K-Akt) are now understood to protect myocardial cells during stress by inhibiting apoptosis(8, 9) Overall, the

anti-cancer activities of HER2 pathway inhibition, including promotion of angiogenesis, cell apoptosis and inhibition of metastases, are the same vital pathways in the myocardium influencing cell survival in the presence of stress, ischemia, hypertension or advancing age(8).

Nonetheless, in patients with HER2 overexpressing (HER2+) breast cancer, trastuzumab-based chemotherapy is standard of care and a vital treatment. In multivariate analysis of HER2 status, treatment and survival among 2091 women with stage IV breast cancer, receiving trastuzumab was shown to decrease risk of death by 44% among HER2+ women compared to HER2 normal (HER2-)(10), thus altering high-risk disease into lower risk. Meta-analysis of 5 randomized, controlled trials of women with early breast cancer (EBC) receiving trastuzumab-based chemotherapy regimens showed significant reductions in mortality, disease recurrence, rates of metastases and secondary tumors when compared to non-trastuzumab regimens(11). Notably, the likelihood of cardiotoxicity was 2.5-fold higher in trastuzumab treatment arms, compared to non-trastuzumab regimens. The Cochrane group conducted a review of the trastuzumab randomized controlled trials (RCT) in the adjuvant setting, which included 11, 991 patients participating in 8 trials. Hazard ratios for overall survival and disease-free survival favored trastuzumab vs non-trastuzumab regimens (hazard ratio 0.66, $p < 0.00001$; hazard ratio 0.60, $p < 0.00001$, respectively)(12). However, there was a fivefold increase in the relative risk of heart failure (HF) and near doubling risk of left ventricular ejection fraction (LVEF) decline. Importantly, the risks may be considerably higher in the

community, among patients whom may not have met the clinical trial criteria. Bowles *et al* performed a population-based study of 12,500 breast cancer patients and incidence of HF and/or cardiomyopathy, observing a hazard ratio of 4.12 for trastuzumab therapy alone, and 7.19 for anthracycline plus trastuzumab five years following treatment initiation(13). Taken together, trastuzumab-related cardiomyopathy in both the short and long-term survivorship course presents a major clinical issue.

1.2 HER2 signaling and cardiac remodeling/cardiotoxicity

The target presented by HER2 pathway inhibition is one of the same vital pathways in the myocardium influencing cell survival in the presence of stress, ischemia, hypertension or advancing age(8). After infancy, the human heart develops by enlargement of cardiac myocytes rather than replication. The adult heart will change shape of myocyte arrangement, or **'**remodel**'** in response to physiologic or pathologic stimuli. LV remodeling has been traditionally described as 'concentric' or 'eccentric'. Concentric hypertrophy represents an increase in LV wall thickness and mass while eccentric hypertrophy is associated with an increase in LV chamber volume and mass. Both remodeling patterns occur in response to pressure or volume loading, respectively, in health(i.e. athletes heart) or disease states such as HF(14). Physiologic LV remodeling occurs in response to increased systemic demand (i.e., regular exercise or pregnancy). In response to activities creating a pressure load (i.e., wrestling), myocyte cell thickness increases by addition of sarcomeres in parallel, resulting in increased LV wall thickness and mass. With increased volume loading LV chamber enlargement

may occur, or ‘eccentric’ hypertrophy, further enhancing cardiac pump efficiency. In this setting, the LV is enlarged by sarcomeres added in series, increasing myocyte cell thickness. Additionally, chronic exercise promotes adaptation of the collagen network supporting the myocytes, enhancing LV function by facilitating efficient myocyte shortening. Physiologic remodeling is also reversible, for after pregnancy or prolonged withdrawal from exercise habits, cardiac size and function returns to the baseline state without negative consequences. Taken together, physiologic remodeling, both eccentric and concentric, contributes to proportional LV chamber development and enhanced cardiac pump function (Figure 1.1).

Pathologic LV remodeling is an established indicator of cardiac insult, which progresses to worsening cardiac function and overt HF(15-17). Four major contributors to remodeling include negative changes to cardiac myocyte structures, proliferation of fibroblasts, degradation of collagen and pathologic apoptosis. In response to insult, cardiac myocyte numbers decrease (necrosis and apoptosis) while remaining myocytes become elongated as new sarcomeres are added in sequence resulting in eccentric remodeling. With lengthening and weakening of the LV wall, increased wall stress occurs, increasing oxygen demand and ischemia. In response to ischemia and myocyte loss, fibroblasts are stimulated, promoting collagen synthesis and the development of LV fibrosis. With excess collagen, a stiff, fibrotic LV results, with impaired conduction and reduced capillary density. Stiffening of the LV results also in impaired relaxation, with decreased filling (diastolic dysfunction) and impaired contractility (systolic

dysfunction). With progressive myocyte loss, increasing fibrosis and reduced capillarity, oxygen transport is eventually impaired, leading to cardiac ischemia. Finally, overall cardiac pump performance is sufficiently compromised for the patient to become symptomatic with transition to HF(18).

Elegant preclinical studies have demonstrated the essential role of HER2 in cardiomyocyte survival and stress adaptation, with mutation or gene deletion leading to dilated cardiomyopathy (increased LV cavity size and decreased LVEF, Figure 1.2)(19, 20). The best-described signaling pathway mediating physiological and pathological cardiac remodeling is the PI3K/Akt pathway, in part activated by extracellular HER2 signaling. The importance of the PI3K enzyme family is increasingly recognized, with roles in cell growth, survival, differentiation and proliferation(14). Of the 3 classes, Class 1 PI3K subtypes (p110alpha, p110beta and p100gamma) are highly expressed in the heart. Importantly, McMullen *et al* highlighted the vital and distinct activities of PI3K, showing in preclinical studies that p110alpha is implicated in physiologic exercise-induced cardiac growth, but not pathologic hypertrophy(21). The Akt family is known to be involved in various cellular processes, including survival/anti-apoptosis, cell cycle activities, metabolism and protein synthesis(22). In blocking the PI3K/Akt pathway, trastuzumab may negatively impact cardiomyocyte survival, additionally the Ras/Raf/MEK/MAPK/VEGF pathway also effected by trastuzumab blocks normal cellular proliferation activities and angiogenesis(23) (Figure 1.3). Also, roles are emerging for HER2 dependent activation of kinases implicated in cell-cell contact, suggesting

potential activities in the maintenance /repair of cardiac electrical and mechanical coupling(3).

Loss of cardiomyocytes may initiate LV remodeling and negative adaptation, including activation of the renin-angiotensin-aldosterone system and fibrosis(24). Pathologic cardiac remodeling precedes systolic dysfunction and in response to chronic loading LV mass initially increases to normalize wall stress, however if wall stress is not reduced, compensatory LV dilation occurs as well as a concurrent decrease in contractile function leading to overt HF(15, 16, 25) In the general population, HF is a lethal condition, with an approximately 50% five-year mortality rate(26-28). Overall, the activities of this pathway play a critical role in cardiac remodeling, hypertrophy and cardiac morbidity/mortality in humans.

1.3 Issues in detecting cardiotoxicity: clinical detection and imaging

1.3.1 Clinical detection of cardiotoxicity

Routine approaches in detecting cardiac toxicity associated with breast cancer treatment include clinical examination and diagnostic imaging. However, the cardinal symptoms of HF (dyspnea, fatigue and edema) are also common in cancer patients(29, 30), and prove challenging for the practitioner to distinguish from cardiac causes. Further, regular symptom inquiry(31, 32), routine vital sign measurements, and identification or management of cardiac risk factors(33, 34) are not routinely attended to in the oncology outpatient clinic. Patients' reports of fatigue and dyspnea are frequently overlooked (38% and 77% of the time, respectively) by health care professionals(32). Basch *et al* conducted a paired

study of patients and physicians reporting of common symptoms on the Common Terminology Criteria for Adverse Events (CTCAE), finding agreement was higher for symptoms that could be observed directly, such as vomiting and diarrhea, than for more subjective symptoms, such as fatigue and dyspnea(35). Therefore, clinical and sub-clinical cardiotoxicity remains undetected in the majority of patients.

1.3.2 Diagnostic imaging to detect cardiotoxicity

In the oncology clinic, determination of cardiotoxicity (and decision-making regarding initiation or ongoing therapy) is guided by resting LVEF, with the lower limit of normal generally accepted at 50%(36, 37). In the clinical practice setting, asymptomatic cardiac dysfunction is observed in 20 - 25% of patients during one-year of trastuzumab therapy. Chia *et al* reported that in unselected EBC patients receiving sequential chemotherapy with trastuzumab, 21.6% of women experienced a cardiac event, requiring temporary or permanent discontinuation of trastuzumab(38). Jassal *et al* observed similar dysfunction rates with 10/42 women (25%) developing trastuzumab-related cardiotoxicity, requiring discontinuation of treatment(39). Given that trastuzumab is prescribed in the EBC setting with the aim of improving long-term survival, dose reductions, delay or discontinuation due to cardiotoxicity are potentially life-threatening events, from the competing risks of cancer and/or cardiac mortality(40).

Multiple-gated acquisition (MUGA) or echocardiography are clinically available imaging modalities for baseline screening and ongoing monitoring of cardiotoxic effects of anti-cancer therapy, with LVEF the sole metric routinely

considered in the oncology clinic. Given that LVEF is load and rate dependent, marked changes in LV size (cavity dilation) can occur to compensate for a decline in contractile function, resulting in no absolute change in LVEF. Further, MUGA cannot determine evolving LV remodeling, a particular challenge in patients with diastolic dysfunction, frequently represented by older females with cardiovascular risk factors ie breast cancer patients(41). Diastolic HF, also termed 'heart failure with preserved ejection fraction' accounts for 50% of HF cases, with outcomes equally dismal as that of systolic HF(42). Reduced LV systolic function is now recognized as a late effect of cardiotoxic drug exposure and not detectable by the usual imaging modalities until significant damage has already occurred(43). Unfortunately, MUGA and echocardiography are limited by poor accuracy, reproducibility and resolution(44) and are often insensitive to early negative changes in cardiac function and remodeling. Cardiac magnetic resonance imaging (cMRI) is considered the "gold standard" for quantifying LV volumes and LVEF(45-47). cMRI is considered a powerful research tool, as the high accuracy and reproducibility of cardiac measures allows significant reduction of sample size in the population of interest(46, 48). Importantly, patterns of remodeling can be readily determined by cMRI. Given the established limitations of other imaging modalities, cMRI offers the capacity to better detect subtle abnormalities and evolution of heart dysfunction related to cancer therapy; further, cMRI is the reference standard to which enhancements to other modalities should be compared.

Taken together, relying on LVEF decline by conventional imaging or overt clinical symptoms limits detection of cardiotoxicity to the late stage of disease process, if detected at all, and increases the likelihood of irreversible injury. Unfortunately, improvement in cardiac dysfunction is not always possible. Cardinale *et al* showed a four-fold decrease in the chance of complete recovery from cardiac dysfunction for each doubling in time-to-HF treatment(49), emphasizing the urgency for early monitoring and intervention, as well as the need for research into preventive strategies.

1.3.3 Population and long-term impacts of trastuzumab exposure

Another major issue is apparent regarding long-term effects of trastuzumab-based therapy. Health Canada provided Notice of Compliance for trastuzumab in the adjuvant treatment of breast cancer on 18 October 2006(50). Since then, approximately 5000 Canadians per year have been exposed to trastuzumab-based therapy. Some investigators postulate trastuzumab-associated cardiotoxicity as muscle 'stunning', similar to myocardial infarction and reversible(51, 52). However, this assertion is based on retrospective reviews of small numbers of select cases, and as such remains unproven. To date, no prospective studies with sufficiently sensitive testing ie cMRI to accurately distinguish long-term cardiotoxicity have been undertaken. Accordingly, the true prevalence of clinical and subclinical cardiotoxicity in treated survivors is unknown. Furthermore, interventions that may prevent cardiotoxicity during trastuzumab-based breast cancer therapy, such as exercise or pharmacotherapy, warrant study and form the basis of this thesis. Against this background, the

overarching theme of this thesis is to examine the short and long-term effects of trastuzumab-based therapy on LV morphology and function in breast cancer patients.

1.4 Study 1: Long term effects of trastuzumab on left ventricular function and aerobic capacity in women with HER2 overexpressing breast cancer.

Guidelines recommend lifestyle modifications as primary approaches in treating stage A HF, defined as patients at high risk but without structural heart disease or HF symptoms(53). Moreover, aerobic training has been shown to attenuate LV remodeling in clinically stable HF patients(54). We previously assessed the effects of four months of aerobic training during trastuzumab-based chemotherapy in EBC patients(55). Following baseline cMRI and cardiopulmonary (VO_{2peak}) exercise testing, a supervised exercise training program was provided to participants, with re-evaluation with cMRI and VO_{2peak} at the 4 month time point. Participants continued their adjuvant trastuzumab-based chemotherapy during the study period according to routine clinical care. The primary finding was that 4 months of trastuzumab therapy was associated with an increase in LV volumes and decreased LVEF in spite of the exercise intervention (55). As a logical extension of this work, in the first study of this thesis, LV morphology and function and aerobic capacity were assessed 4 years following completion of trastuzumab-based therapy.

1.5 Study 2: A prospective evaluation of LV remodeling among breast cancer patients receiving trastuzumab-based chemotherapy and

randomized to receive conventional heart failure pharmacotherapy or placebo.

Importantly, the work of our group and others has shown that trastuzumab-related LV remodeling and/or cardiotoxicity occurs early(39, 55, 56) and potentially conveys long-term negative consequences, highlighting the need for preventive maneuvers. National evidence-based guidelines recommend HF pharmacotherapy in patients with LV remodeling(53, 57), and therapies aimed at reversing this process have been associated with clinical benefit(58). Angiotensin-converting enzyme inhibitors (ACEI) slow progress of LV dilation, reducing preload and afterload, reducing sympathetic activation and preventing angiotensin II from exacerbating further remodeling(59). Angiotensin-receptor blocking agents (ARB) are considered second-line therapy for patients unable to tolerate ACEI, usually due to bothersome cough. Beta-blocker (BB) cardio-inhibitory effects (chronotropy and inotropy) have been shown to improve LV function and reduce mortality in HF patients; while the mechanism is not understood, blockade of excessive and chronic sympathetic influences may alleviate further exacerbation/remodeling(59). Accordingly, in the second study of this thesis, the effects of that standard HF pharmacotherapy compared to placebo in preventing LV remodeling during the first three months of trastuzumab-based therapy was studied.

Figure 1.1: Patterns and characteristics of cardiac remodeling, adapted from Bernardo(60).

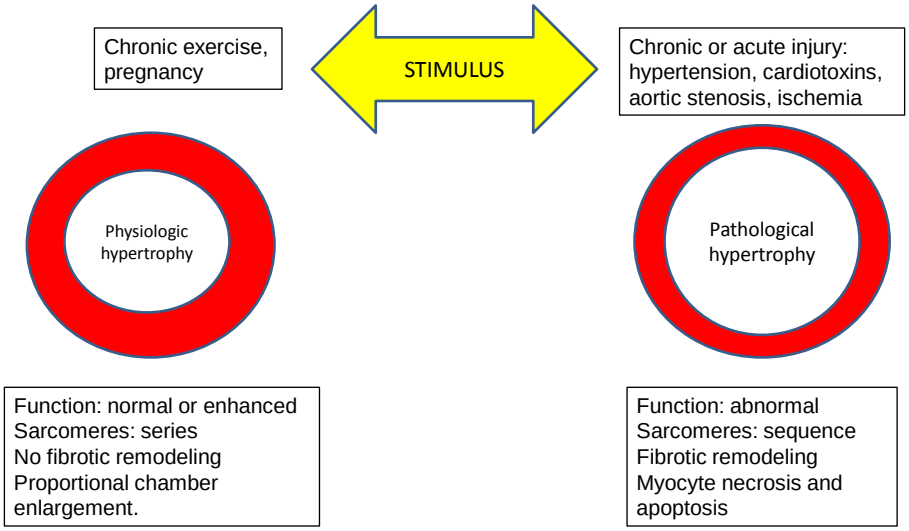
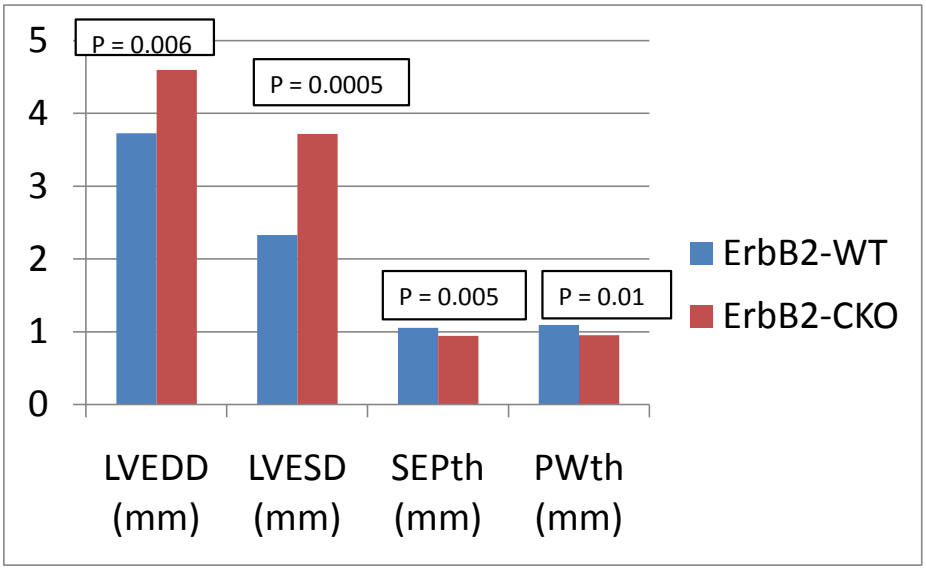
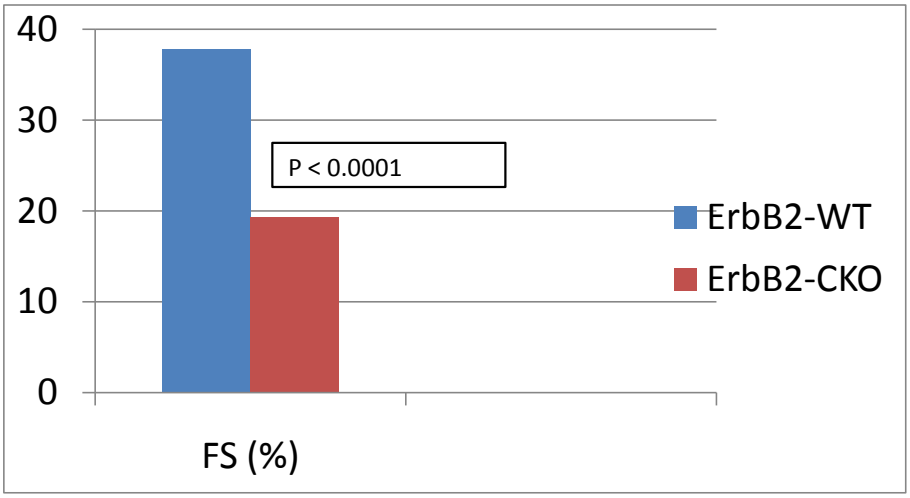


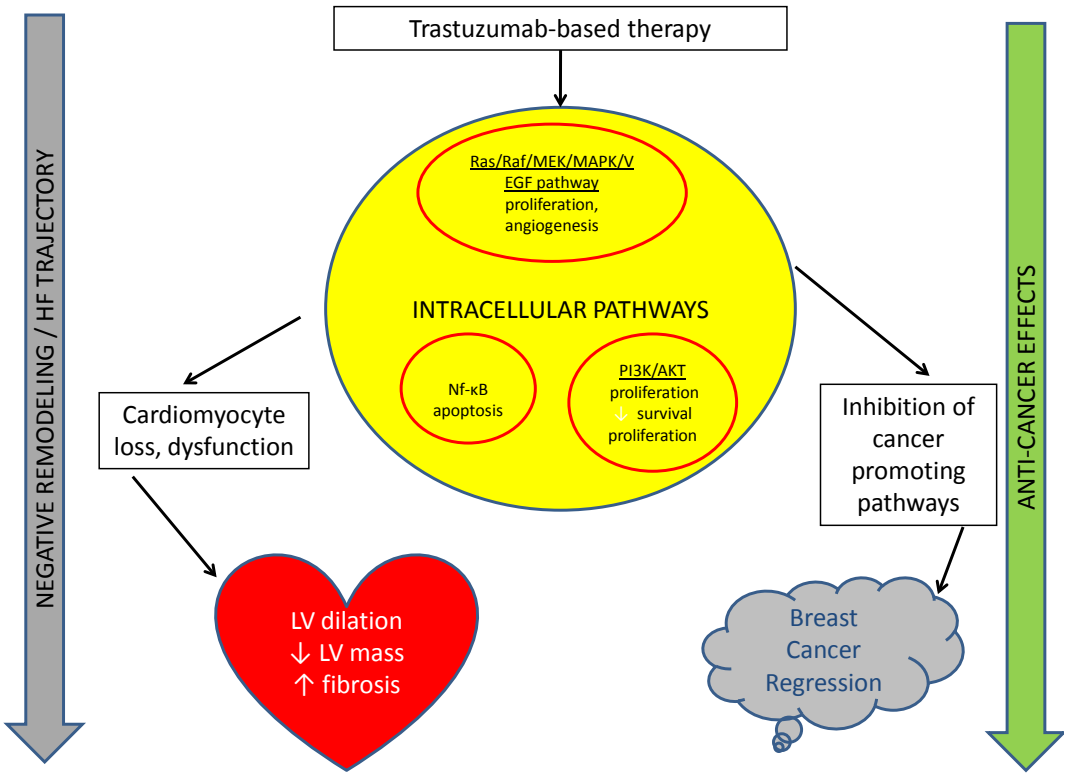
Figure 1.2: Left ventricular morphology and function in erbB2 wild-type and conditional knock-out mice. Adapted from Crone (19).





LVEDD, LV internal diameter in diastole; LVESD, LV internal diameter in systole; FS, fractional shortening; SEPth, septal wall thickness; PWth, posterior wall thickness.

Figure 1.3: Potential effects of trastuzumab on the heart



1.6 References

1. Canadian Cancer Statistics. Toronto, Canada2010.

2. de Moor JS, Mariotto AB, Parry C, Alfano CM, Padgett L, Kent EE, et al. Cancer Survivors in the United States: Prevalence across the Survivorship Trajectory and Implications for Care. Cancer Epidemiology Biomarkers & Prevention. 2013 April 1, 2013;22(4):561-70.

3. Lemmens K, Doggen K, De Keulenaer GW. Role of Neuregulin-1/ErbB Signaling in Cardiovascular Physiology and Disease. Circulation. 2007 August 21, 2007;116(8):954-60.

4. Chen MH, Kerkela R, Force T. Mechanisms of Cardiac Dysfunction Associated With Tyrosine Kinase Inhibitor Cancer Therapeutics. Circulation. 2008 July 1, 2008;118(1):84-95.

5. Nabholtz J-M, Breast Cancer International Research Group. Breast cancer management : application of evidence to patient care. London: Martin Dunitz; 2000. xix, 569 p. p.

6. Slamon DJ, Clark GM, Wong SG, Levin WJ, Ullrich A, McGuire WL. Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. Science. 1987 Jan 9;235(4785):177-82. PubMed PMID: 3798106. Epub 1987/01/09. eng.

7. Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, Bajamonde A, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. The New England journal of medicine. 2001 Mar 15;344(11):783-92. PubMed PMID: 11248153. Epub 2001/03/15. eng.

8. Feldman AM, Koch WJ, Force TL. Developing Strategies to Link Basic Cardiovascular Sciences with Clinical Drug Development: Another Opportunity for Translational Sciences. Clin Pharmacol Ther. 2007 03/28/online;81(6):887-92.

9. Telli ML, Hunt SA, Carlson RW, Guardino AE. Trastuzumab-Related Cardiotoxicity: Calling Into Question the Concept of Reversibility. J Clin Oncol. 2007 August 10, 2007;25(23):3525-33.

10. Dawood S, Broglio K, Buzdar AU, Hortobagyi GN, Giordano SH. Prognosis of women with metastatic breast cancer by HER2 status and trastuzumab treatment: an institutional-based review. J Clin Oncol. Jan 1;28(1):92-8. PubMed PMID: 19933921. Epub 2009/11/26. eng.

11. Viani GA, Afonso SL, Stefano EJ, De Fendi LI, Soares FV. Adjuvant trastuzumab in the treatment of her-2-positive early breast cancer: a meta-analysis of published randomized trials. BMC Cancer. 2007;7:153. PubMed PMID: 17686164. Epub 2007/08/10. eng.

12. Moja L, Tagliabue L, Balduzzi S, Parmelli E, Pistotti V, Guarneri V, et al. Trastuzumab containing regimens for early breast cancer. Cochrane Database Syst Rev. 2012;4:CD006243. PubMed PMID: 22513938.

13. Bowles EJ, Wellman R, Feigelson HS, Onitilo AA, Freedman AN, Delate T, et al. Risk of heart failure in breast cancer patients after anthracycline and trastuzumab treatment: a retrospective cohort study. J Natl Cancer Inst. 2012 Sep 5;104(17):1293-305. PubMed PMID: 22949432. Pubmed Central PMCID: 3433392.

14. Walker J, Bhullar N, Fallah-Rad N, Lytwyn M, Golian M, Fang T, et al. Role of Three-Dimensional Echocardiography in Breast Cancer: Comparison With Two-Dimensional Echocardiography, Multiple-Gated Acquisition Scans, and Cardiac Magnetic Resonance Imaging. Journal of Clinical Oncology. 2010 July 20, 2010;28(21):3429-36.

15. Cohn JN, Ferrari R, Sharpe N. Cardiac remodeling--concepts and clinical implications: a consensus paper from an international forum on cardiac remodeling. Behalf of an International Forum on Cardiac Remodeling. J Am Coll Cardiol. 2000 Mar 1;35(3):569-82. PubMed PMID: 10716457. Epub 2000/03/15. eng.

16. Mann DL, Bristow MR. Mechanisms and Models in Heart Failure: The Biomechanical Model and Beyond. Circulation. 2005 May 31, 2005;111(21):2837-49.

17. Vasan RS, Larson MG, Benjamin EJ, Evans JC, Levy D. Left ventricular dilatation and the risk of congestive heart failure in people without myocardial infarction. N Engl J Med. 1997 May 8;336(19):1350-5. PubMed PMID: 9134875. Epub 1997/05/08. eng.

18. Zile MR, Brutsaert DL. New Concepts in Diastolic Dysfunction and Diastolic Heart Failure: Part I: Diagnosis, Prognosis, and Measurements of Diastolic Function. Circulation. 2002 March 19, 2002;105(11):1387-93.

19. Crone SA, Zhao YY, Fan L, Gu Y, Minamisawa S, Liu Y, et al. ErbB2 is essential in the prevention of dilated cardiomyopathy. Nat Med. 2002 May;8(5):459-65. PubMed PMID: 11984589. Epub 2002/05/02. eng.

20. Ozcelik C, Erdmann B, Pilz B, Wettschureck N, Britsch S, Hubner N, et al. Conditional mutation of the ErbB2 (HER2) receptor in cardiomyocytes leads to dilated cardiomyopathy. Proceedings of the National Academy of Sciences of the United States of America. 2002 Jun 25;99(13):8880-5. PubMed PMID: 12072561. Pubmed Central PMCID: 124392.

21. Tanaka H, Seals DR. Invited Review: Dynamic exercise performance in Masters athletes: insight into the effects of primary human aging on physiological functional capacity. Journal of Applied Physiology. 2003 November 1, 2003;95(5):2152-62.

22. McGuire DK, Levine BD, Williamson JW, Snell PG, Blomqvist CG, Saltin B, et al. A 30-Year Follow-Up of the Dallas Bed Rest and Training Study. Circulation. 2001 September 18, 2001;104(12):1358-66.

23. Hudis CA. Trastuzumab--mechanism of action and use in clinical practice. N Engl J Med. 2007 Jul 5;357(1):39-51. PubMed PMID: 17611206.

24. Chien KR. Myocyte survival pathways and cardiomyopathy: implications for trastuzumab cardiotoxicity. Seminars in oncology. 2000 Dec;27(6 Suppl 11):9-14; discussion 92-100. PubMed PMID: 11236034.

25. Haykowsky MJ, Liang Y, Pechter D, Jones LW, McAlister FA, Clark AM. A meta-analysis of the effect of exercise training on left ventricular remodeling in heart failure patients: the benefit depends on the type of training performed. J Am Coll Cardiol. 2007 Jun 19;49(24):2329-36. PubMed PMID: 17572248. Epub 2007/06/19. eng.

26. Sanderson JE. Heart failure with a normal ejection fraction. Heart. 2007 February 1, 2007;93(2):155-8.

27. Gaudron P, Eilles C, Kugler I, Ertl G. Progressive left ventricular dysfunction and remodeling after myocardial infarction. Potential mechanisms and early predictors. Circulation. 1993 Mar;87(3):755-63. PubMed PMID: 8443896. Epub 1993/03/01. eng.

28. Greenberg B, Quinones MA, Koilpillai C, Limacher M, Shindler D, Benedict C, et al. Effects of long-term enalapril therapy on cardiac structure and function in patients with left ventricular dysfunction. Results of the SOLVD echocardiography substudy. Circulation. 1995 May 15;91(10):2573-81. PubMed PMID: 7743619. Epub 1995/05/15. eng.

29. Chu TF, Rupnick MA, Kerkela R, Dallabrida SM, Zurakowski D, Nguyen L, et al. Cardiotoxicity associated with tyrosine kinase inhibitor sunitinib. Lancet. 2007 Dec 15;370(9604):2011-9. PubMed PMID: 18083403. Epub 2007/12/18. eng.

30. Force T, Krause DS, Van Etten RA. Molecular mechanisms of cardiotoxicity of tyrosine kinase inhibition. Nat Rev Cancer. 2007 May;7(5):332-44. PubMed PMID: 17457301. Epub 2007/04/26. eng.

31. Janjan NA, Cleeland CS. Pain and suffering during cancer therapy: continued sins of omission. Int J Radiat Oncol Biol Phys. 2008 Sep 1;72(1):6-8. PubMed PMID: 18722257. Epub 2008/08/30. eng.

32. Fromme EK, Eilers KM, Mori M, Hsieh YC, Beer TM. How accurate is clinician reporting of chemotherapy adverse effects? A comparison with patient-

reported symptoms from the Quality-of-Life Questionnaire C30. J Clin Oncol. 2004 Sep 1;22(17):3485-90. PubMed PMID: 15337796. Epub 2004/09/01. eng.

33. Wilson PW, D'Agostino RB, Levy D, Belanger AM, Silbershatz H, Kannel WB. Prediction of coronary heart disease using risk factor categories. Circulation. 1998 May 12;97(18):1837-47. PubMed PMID: 9603539. Epub 1998/05/29. eng.

34. Lenihan DJ. Tyrosine Kinase Inhibitors: Can Promising New Therapy Associated With Cardiac Toxicity Strengthen the Concept of Teamwork? J Clin Oncol. 2008 November 10, 2008;26(32):5154-5.

35. Basch E, Iasonos A, McDonough T, Barz A, Culkin A, Kris MG, et al. Patient versus clinician symptom reporting using the National Cancer Institute Common Terminology Criteria for Adverse Events: results of a questionnaire-based study. The Lancet Oncology. 2006;7(11):903-9.

36. Seidman A, Hudis C, Pierri MK, Shak S, Paton V, Ashby M, et al. Cardiac dysfunction in the trastuzumab clinical trials experience. J Clin Oncol. 2002 Mar 1;20(5):1215-21. PubMed PMID: 11870163.

37. Mackey JR, Clemons M, Cote MA, Delgado D, Dent S, Paterson A, et al. Cardiac management during adjuvant trastuzumab therapy: recommendations of the Canadian Trastuzumab Working Group. Curr Oncol. 2008 Feb;15(1):24-35. PubMed PMID: 18317582. Epub 2008/03/05. eng.

38. McArthur HL, Chia S. Cardiotoxicity of trastuzumab in clinical practice. The New England journal of medicine. 2007 Jul 5;357(1):94-5. PubMed PMID: 17611218. Epub 2007/07/06. eng.

39. Fallah-Rad N, Walker JR, Wassef A, Lytwyn M, Bohonis S, Fang T, et al. The utility of cardiac biomarkers, tissue velocity and strain imaging, and cardiac magnetic resonance imaging in predicting early left ventricular dysfunction in patients with human epidermal growth factor receptor II-positive breast cancer treated with adjuvant trastuzumab therapy. J Am Coll Cardiol. 2011 May 31;57(22):2263-70. PubMed PMID: 21616287.

40. Patnaik JL, Byers T, DiGuiseppi C, Dabelea D, Denberg TD. Cardiovascular disease competes with breast cancer as the leading cause of death for older females diagnosed with breast cancer: a retrospective cohort study. Breast cancer research : BCR. 2011;13(3):R64. PubMed PMID: 21689398. Pubmed Central PMCID: 3218953.

41. Kitzman DW, Gardin JM, Gottdiener JS, Arnold A, Boineau R, Aurigemma G, et al. Importance of heart failure with preserved systolic function in patients > or = 65 years of age. CHS Research Group. Cardiovascular Health Study. Am J Cardiol. 2001 Feb 15;87(4):413-9. PubMed PMID: 11179524. Epub 2001/02/17. eng.

42. Maeder MT, Kaye DM. Heart failure with normal left ventricular ejection fraction. J Am Coll Cardiol. 2009 Mar 17;53(11):905-18. PubMed PMID: 19281919. Epub 2009/03/14. eng.

43. Zuppinger C, Timolati F, Suter TM. Pathophysiology and diagnosis of cancer drug induced cardiomyopathy. Cardiovasc Toxicol. 2007;7(2):61-6. PubMed PMID: 17652805. Epub 2007/07/27. eng.

44. Bellenger NG, Burgess MI, Ray SG, Lahiri A, Coats AJ, Cleland JG, et al. Comparison of left ventricular ejection fraction and volumes in heart failure by echocardiography, radionuclide ventriculography and cardiovascular magnetic resonance; are they interchangeable? Eur Heart J. 2000 Aug;21(16):1387-96. PubMed PMID: 10952828. Epub 2000/08/23. eng.

45. Bellenger N, Pennell D. Magnetic resonance imaging in cardiology. J R Coll Physicians Lond. 1999 Jan-Feb;33(1):12-8. PubMed PMID: 10192063. Epub 1999/04/07. eng.

46. Bellenger NG, Davies LC, Francis JM, Coats AJ, Pennell DJ. Reduction in sample size for studies of remodeling in heart failure by the use of cardiovascular magnetic resonance. Journal of cardiovascular magnetic resonance : official journal of the Society for Cardiovascular Magnetic Resonance. 2000;2(4):271-8. PubMed PMID: 11545126. Epub 2001/09/08. eng.

47. Wolfe RR. Skeletal muscle protein metabolism and resistance exercise. J Nutr. 2006 Feb;136(2):525S-8S. PubMed PMID: 16424140. Epub 2006/01/21. eng.

48. Strohm O, Schulz-Menger J, Pilz B, Osterziel KJ, Dietz R, Friedrich MG. Measurement of left ventricular dimensions and function in patients with dilated cardiomyopathy. J Magn Reson Imaging. 2001 Mar;13(3):367-71. PubMed PMID: 11241808. Epub 2001/03/10. eng.

49. Cardinale D, Colombo A, Lamantia G, Colombo N, Civelli M, De Giacomi G, et al. Anthracycline-induced cardiomyopathy: clinical relevance and

response to pharmacologic therapy. J Am Coll Cardiol. Jan 19;55(3):213-20. PubMed PMID: 20117401. Epub 2010/02/02. eng.

50. Report Card on Cancer in Canada.: Cancer Advocacy Coalition of Canada.; 2010.

51. Ewer MS, Lippman SM. Type II Chemotherapy-Related Cardiac Dysfunction: Time to Recognize a New Entity. Journal of Clinical Oncology. 2005 May 1, 2005;23(13):2900-2.

52. Ewer MS, Vooletich MT, Durand J-B, Woods ML, Davis JR, Valero V, et al. Reversibility of Trastuzumab-Related Cardiotoxicity: New Insights Based on Clinical Course and Response to Medical Treatment. J Clin Oncol. 2005 November 1, 2005;23(31):7820-6.

53. Hunt SA, Abraham WT, Chin MH, Feldman AM, Francis GS, Ganiats TG, et al. ACC/AHA 2005 Guideline Update for the Diagnosis and Management of Chronic Heart Failure in the Adult: A Report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Writing Committee to Update the 2001 Guidelines for the Evaluation and Management of Heart Failure): Developed in Collaboration With the American College of Chest Physicians and the International Society for Heart and Lung Transplantation: Endorsed by the Heart Rhythm Society. Circulation. 2005 September 20, 2005;112(12):e154-235.

54. Haykowsky M, Brubaker P, Kitzman D. Role of Physical Training in Heart Failure with Preserved Ejection Fraction. Current heart failure reports. 2012 2012/06/01;9(2):101-6. English.

55. Haykowsky MJ, Mackey JR, Thompson RB, Jones LW, Paterson DI. Adjuvant trastuzumab induces ventricular remodeling despite aerobic exercise training. Clin Cancer Res. 2009 Aug 1;15(15):4963-7. PubMed PMID: 19622583.

56. Wadhwa D, Fallah-Rad N, Grenier D, Krahn M, Fang T, Ahmadie R, et al. Trastuzumab mediated cardiotoxicity in the setting of adjuvant chemotherapy for breast cancer: a retrospective study. Breast Cancer Res Treat. 2009 Sep;117(2):357-64. PubMed PMID: 19082707.

57. Malcom J, Arnold O, Howlett JG, Ducharme A, Ezekowitz JA, Gardner MJ, et al. Canadian Cardiovascular Society Consensus Conference guidelines on heart failure--2008 update: best practices for the transition of care of heart failure patients, and the recognition, investigation and treatment of cardiomyopathies. Can J Cardiol. 2008 Jan;24(1):21-40. PubMed PMID: 18209766. Epub 2008/01/23. eng.

58. Kramer DG, Trikalinos TA, Kent DM, Antonopoulos GV, Konstam MA, Udelson JE. Quantitative evaluation of drug or device effects on ventricular remodeling as predictors of therapeutic effects on mortality in patients with heart failure and reduced ejection fraction: a meta-analytic approach. J Am Coll Cardiol. 2010 Jul 27;56(5):392-406. PubMed PMID: 20650361.

59. Katzung BG. Basic and Clinical Pharmacology. 11 ed: McGraw-Hill; 2009.

60. Bernardo BC, Weeks KL, Pretorius L, McMullen JR. Molecular distinction between physiological and pathological cardiac hypertrophy:

experimental findings and therapeutic strategies. Pharmacology & therapeutics. 2010 Oct;128(1):191-227. PubMed PMID: 20438756.

Chapter 2

Long term effects of trastuzumab on left ventricular function and aerobic capacity in women with HER2 overexpressing breast cancer

---

2.1 Introduction

Trastuzumab is a proven therapy for HER2+ breast cancer, improving overall survival compared to non-trastuzumab containing chemotherapy regimens(1, 2). As a result of these and other highly effective adjuvant therapies, five-year survival rates approach 88% among EBC patients(3). Another consequence of this favorable response is that cardiovascular disease is now the major cause of death in EBC survivors(4), therefore, a need for improved understanding of the increasing risks and rates of late-onset effects are warranted. Short-term effects of trastuzumab on LV remodeling (LV dilation and decline in LVEF) may, over time, progress to further LV dysfunction, reduced exercise tolerance and overt HF(5-7), highlighting the need for long-term study of these metrics.

Our group previously evaluated the effects of four months of aerobic training on LV remodeling during trastuzumab-based chemotherapy in EBC patients(8). In brief, following baseline cMRI and cardiopulmonary (VO_{2peak}) exercise testing, a supervised aerobic exercise training program was provided to participants, with re-evaluation with cMRI and VO_{2peak} at the 4 month time point. Participants continued their adjuvant trastuzumab-based chemotherapy during the study period according to routine clinical care. The primary finding was that 4

months of trastuzumab therapy and aerobic training was associated with an increase in LV volumes and decreased LVEF(8).

Currently, the long-term sequelae of trastuzumab-related LV remodeling remain unknown. As an extension of this prior work, a follow-up study was performed to determine the long-term effects of trastuzumab-based chemotherapy, examining LV remodeling, and aerobic capacity. Given decision-making in the oncology clinic is guided by LVEF, our primary outcome of interest was change in LVEF from baseline, while secondary outcomes included patient reported symptoms and cardiovascular risk profile. Further, as aerobic training has been shown to be an effective intervention in improving peak VO_2 in clinically stable HF patients that adhere to prescribed training sessions,(9) a sub-analysis comparing those who adhered (ADH) to the exercise intervention during the original study (defined as 80% or greater attendance to the exercise program) vs those who did not adhere to training (NAD) was performed(9).

2.2 Methods

2.2.1 Participants

Seventeen women with histologically confirmed HER2 overexpressing early breast cancer who participated in the 2009 study were contacted by telephone to explain the project and establish interest in participation. Exclusion criteria included: recurrent malignancy, contraindication to MRI (i.e., metal implants) or unwillingness or inability to undergo study-related testing. Upon arrival at the research center, study procedures were reviewed in detail and written informed consent was obtained. Vital signs, height and weight, medication history and

functional status(10) were evaluated. Ethics approvals were secured from the two relevant Boards.

2.2.2 Cardiac MRI LV morphology and function.

Assessment of LV volumes, mass, and LVEF was performed with a 1.5 Tesla Magnetom Sonata scanner (Siemens Medical Solutions). Steady-state free precession cine techniques acquired parallel, 8-mm-thick short-axis slices (plus a 2-mm gap) throughout the LV. Standard post-processing software (Syngo MR;Siemens) was used to calculate LV volumes, mass and function from baseline, 4 month and 4 year scans. Heart rate and blood pressure were monitored throughout the examination.

2.2.3 Cardiopulmonary (VO_{2peak}) exercise testing.

This test was performed on an electrically braked cycle ergometer; power output was initiated at 15 W and increased by 15 W every 2 min. HR, BP, rate of perceived exertion (RPE), and expired gas analysis was measured. The highest oxygen consumed over a 1-min period determined the VO_{2peak} score.

2.2.4 Statistical considerations

Descriptive statistics were obtained for all study variables. Mean and standard deviations were reported for continuous variables, frequency and proportions were reported for categorical variables. One way analysis of variance was used to compare the means of the continuous variables between two or more groups. For post hoc comparison between the groups Tukey's tests were used. Independent t-tests were used to compare the mean difference between two

groups. All analysis was conducted using SPSS version 19 (IBM SPSS Statistics). A p-value ≤ 0.05 was considered for all statistical tests.

2.3 Results

2.3.1 Participant characteristics

From March 2012 to June 2012, 16 of the 17 eligible women agreed to participate in the study and attended the clinic for study-related testing (Table 2.1). All 16 women underwent cardiac MRI and VO_{2peak} testing, with no adverse events.

The subjects' mean age was 58 ± 7 years, with nearly four years elapsing since completion of trastuzumab therapy. Eight participants had received anthracycline-containing regimens. Three patients did not complete the entire year of trastuzumab therapy due to LVEF decline, two in the NAD group and one in the ADH group. Two participants were receiving their final year of tamoxifen. All reported full functional status (KPS(10)) and denied any symptoms limiting their usual activities of daily living. None reported an adverse cardiac event (MI or stroke) nor clinical diagnosis of HF since their anti-cancer therapy. Two participants were current smokers, 4 were ex-smokers with the remainder never-smokers. One participant had succeeded in quitting smoking since the original study. The mean BMI was 26.5 ± 8 compared to 26.4 ± 9 at baseline, p = 0.9. Antihypertensive therapies (ACEI, beta-blocker or ARB) were prescribed for 5 participants, compared to 3 at baseline. Two were receiving treatment for type 2 diabetes (one at baseline), two received lipid-lowering agents (one at baseline) and one received both antihypertensive and lipid-lowering therapy.

The ADH group consisted of six of the 16 participants who attended at least 80% of the prescribed sessions; of these, three women received anthracycline-based regimens and two received left breast/chest wall radiotherapy. The ten patients in the NAD group attended an average of 50% of the exercise sessions. Of these, four patients received anthracycline-containing therapy and three received left breast/chest wall radiotherapy.

2.3.2 Cardiac MRI LV morphology and function

Left ventricular ejection fraction significantly decreased from baseline to 4 months, and returned to baseline at the 4-year timepoint. Left ventricular end-systolic volume significantly increased from baseline to 4 months (54 ± 11 versus. 68 ± 12, p = 0.02) followed by a 15 mL reduction after 4 years compared to 4 months (68 ± 12 versus 52 ± 15, p = 0.003). Left ventricular end-diastolic volume was significantly lower when comparing 4 month and 4-year measures, with an absolute volume difference of approximately 25mL (153 ± 20 versus 128 ± 27, p = 0.002). A 10% difference in longitudinal strain was found at 4 months compared to baseline (-17.4 ± 2.2 versus -15.7 ± 2.1, p = 0.05), returning to baseline values at 4 year testing. LV mass was significantly reduced at 4 years versus baseline. No significant difference was found in LVSV or LV mass/volume ratio across timepoints (Table 2.2).

Examining the results by exercise adherence, LVEF was not significantly different when comparing 4-year testing with baseline and 4 month observations for either group (Tables 2.3 and 2.4). Left ventricular end diastolic volume decreased in the NAD group by 26mL from 4 months to 4-years (p = 0.06) while

LVESV was significantly different from baseline and 4 months and 4 months to 4 years (+13mL and -15mL, p = 0.03 and p = 0.02, respectively). Lastly, a 13% decrease in longitudinal strain was observed from baseline to 4 months in the NAD group, that persisted at the 4 year evaluation.

2.3.3 Cardiopulmonary exercise and hemodynamics

No statistically significant differences were observed for the entire group for peak VO_2 (L/min or ml/kg/min), power output, heart rate, systolic blood pressure, rate of perceived exeetion, or respiratory exchange ratio (Table 2.5). Diastolic blood pressure increased after 4-years compared to the 4 month (89 ± 9 versus 81 ± 7 mmHg, p = 0.02). Exercise outcomes by adherence revealed a higher peak VO_2 (L/min) in the ADH between baseline and 4 months (p = 0.08, Table 2.6). Peak diastolic blood pressure DBP in the NAD group was significantly different in 2012 versus 4 months, with an absolute difference of 13mmHg (p = 0.01, Table 2.7).

2.4 Discussion

2.4.1 Long-term effects of trastuzumab on LV morphology and function

Currently, the long-term effects of trastuzumab-based therapy on LV remodeling and exercise capacity are not known. We performed a cross-sectional study of 16 female breast cancer survivors to determine the long-term effects of trastuzumab-based chemotherapy, examining LV remodeling and aerobic capacity. Our primary outcome, LVEF assessed by cardiac MRI, declined signficantly by 6% at the 4 month timepoint vs baseline, and returned to baseline values 4 years post trastuzumab therapy. Exercise adherence did not alter this

finding. This result is consistent with longer term observations reported by the investigators of the pivotal breast cancer studies, suggesting that LVEF recovers over time (2, 11, 12). However, LVEF may be an insensitive measure of gradual remodeling of the LV. Rather, assessment of LV volumes is important in the early detection and characterization of LV remodeling(5, 6).

Accordingly, the key findings of this study are the observations of long-term and significant alterations in LV geometry following trastuzumab exposure. Detrimental LV remodeling is established as an early indicator of cardiac injury, which progresses to worsening cardiac function and overt HF(5-7). We observed acute LV dilatation at 4 months, reflected by increases in LVEDV and LVESV (15mL and 14mL, respectively). This was followed by statistically significant decrease of LV volumes after 4-years of cessation of trastuzumab therapy, with LVEDV decreasing by 25mL and LVESV by 16mL (Figures 2.1, 2.2). Changes in LV mass also occurred, with a 5g increase from baseline at 4 months, followed by a statistically significant 14g decrease at 4 years (Figure 2.3). These alterations may be explained by two processes. Cardiomyocytes represent the approximately 75% of cardiac tissue volume(13). ErbB2 inhibition blocks the Akt pathway, an established cardiomyocyte growth-promotion and anti-apoptotic regulator(14). Furthermore, PI3K is intimately associated with Akt activities, and has an established role in physiologic cardiac hypertrophy(15) (16). Inhibition of these pathways could potentially result in cardiomyocyte apoptosis, contributing to decreasing cardiac mass. As a result, loss of mass with thinning LV walls could potentially lead to eccentric hypertrophy and LV dilation over time.

Secondly, alterations in LV mass and volumes could be explained by the effect of chronic physical inactivity over time. Dorfman *et al* used cardiac MRI to examine cardiac mass and volumes in 24 healthy females before and after 60 days of head-down-tilt bedrest(17). Subjects were randomized to no intervention, supine treadmill and resistance training or protein supplementation. In the subjects randomized to the exercise intervention, LV mass was preserved and negative remodeling was prevented, however, in the control group significant decreases in LV mass and LV volumes were observed. Sedentary lifestyle is highly prevalent in breast cancer patients. In a long-term study of 631 breast cancer survivors, Mason *et al* observed that before diagnosis, 34 percent of the women met U.S. physical activity guidelines, dropping to 21.4 percent at 10 years(18). Similar negative lifestyle patterns could explain the findings observed here. Indeed, peak VO_2 remained unaltered in the non-adherent subjects at all testing points.

Aside from 4-year LVESV in the nonadherence group, we did not observe significant interactions of exercise adherence on LV geometry. McMullen's preclinical work showed that p110alpha is implicated in physiologic exercise-induced cardiac growth(19), suggesting a theoretical opportunity to offset trastuzumab's inhibition of this important pathway(20). It is also possible that exercise habit could up-regulate VEGF, enhance cardiac capillarity, and attenuate, to some degree, subclinical fibrotic remodeling(21). However, preclinical observations may not apply in humans, specifically breast cancer patients receiving chemotherapy, additionally complicated by the multiple toxic therapies

administered concurrently with exercise. Further, even with adherence to exercise, intensity of effort may not be sufficient to offset direct and indirect cardiotoxic effects. Finally, it is likely, given the redundancy of cellular signaling pathways (and the importance of PI3K/Akt) that escape pathways exist, hampering remediation of toxicity by any single intervention.

Reduced LV mass has been reported in long-term imaging studies of adult survivors of pediatric malignancy. In 156 survivors at a median 11 years, echocardiography revealed decreased LV mass and LV wall thickness(22). Armstrong *et al* performed cMRI and echocardiography in 114 survivors a median 28 years after diagnosis, observing 48% with reduced cardiac mass(23). These findings were even more pronounced in those patients who received anthracyclines, even at conservative doses of 150mg/m^2 or less. Here, reduced LVEF was observed in 27% and reduced cardiac mass in 56%. Importantly, in both studies cardiac abnormalities were also observed in survivors who did not undergo systemic or radiation therapy, suggesting a complex interplay between malignancy, cardiovascular disease and effects of lifestyle factors (Figure 2.4). Preclinical work is necessary to elucidate these phenomenon, for current understanding suggests the same gene pathways are activated, regardless of whether the heart hypertrophies or atrophies(24).

Loss of LV mass may also represent cardiac muscle wasting (atrophy) or accelerated cardiac aging. Loss of skeletal muscle mass (termed cachexia or sarcopenia) has been documented in the elderly, HF patients(25) and across the cancer continuum(26, 27), however little attention has been paid to cardiac

muscle atrophy. In an elegant murine model study, Cosper *et al* showed that presence of malignancy causes a loss of cardiac mass, associated with decreased cardiac myocyte size(28). Further, fibrosis was significantly increased in both male (50%) and females (65%). Notably, in spite of extensive cardiac muscle loss, no change in ejection fraction was observed in M-mode echocardiography(28). These observations not only confirm the effects of cancer on both skeletal and cardiac muscle, but that LVEF drop does not occur until significant impairment has occurred.

The potential contribution of aging in this population also warrants consideration. Aging is associated with progressive fibrosis occurring in multiple organ systems(29) which, even in healthy older individuals, is associated with gradual structural and functional alterations. Typically, increased LV mass is observed with age, attributed to fibrotic remodeling and response to peripheral vascular stiffening and increased hemodynamic load(30). However, this increase in mass is accompanied by age-related senesence of functional cardiomyocytes. Potential interactions may influence the evolution of LV mass in cancer survivors, such as baseline individual risk factors, the type of anti-cancer therapy received, gender and lifestyle before, during and after therapy. Large scale studies of these factors are necessary to elucidate these interactions.

Longitudinal strain decreased from baseline to 4 months, and persisted 4 years later in the nonadherence group. Novel measures such as longitudinal strain may be early indicators of cardiac dysfunction. In serial echocardiography in 35 breast cancer patients receiving trastuzumab-based chemotherapy, a decrease in

longitudinal strain rate was observed in 18 (51%) patients; 5 of these patients experienced LVEF reduction concurrently or within 20 months follow-up(31). In 43 breast cancer patients receiving anthracycline and trastuzumab-based chemotherapy, Sawaya *et al* showed that early decrease in myocardial strain (baseline to 3 month assessment) was an independent predictor of later cardiotoxicity (32). Here, a decrease of longitudinal strain by >10% at 3 months predicted those patients who developed a significant decrease in LVEF at 6 months. We observed a greater than 10% decrease in strain from baseline to 4 months, which persisted in 2012 in the nonadherence group. It is possible that the exercise intervention, even with less than optimal participation, provided some degree of initial cardioprotection. Further study of this particular metric is warranted.

2.4.2 Long-term effects of trastuzumab on cardiopulmonary function

Another key finding is that, on the background of normal LVEF, we showed that significant impairment of cardiopulmonary function remains an average of four years after adjuvant trastuzumab-based chemotherapy. The average VO_{2peak} of 22.4 ml/kg/min in this group of women, mean age 58, is equivalent to that of a 72 year old sedentary female(33). These findings are similar to our previous observations in breast cancer populations, where VO_{2peak} is consistently and significantly impaired, regardless of the timing of assessment (before or after definitive therapy, survivorship period)(34). In a pooled study of 248 breast cancer patients across the disease continuum, we observed that approximately one third of breast cancer patients have a VO_{2peak} lower than 15.4

ml/kg/ml (threshold for independent functioning) and reach a predicted age-related VO_{2peak} approximately 20 to 30 years earlier than healthy women. We also performed a meta-analysis of the published literature on RCTs of exercise interventions in cancer patients, with VO_{2peak} as the gold standard measure. Here, exercise training was associated with a variety of healthful benefits, and a modest but statistically significant VO_{2peak} (weighted mean difference = +2.90 mL kg$^{/}$min relative to sedentary control group subjects(35).

Notably, we observed that adherence to exercise resulted in higher VO_{2peak} measures at 4 months and 4 years, with a 4.1 mL/kg/min difference between ADH and NAD. The clinical importance of this difference in this specific population is unknown, however, Gulati found mortality risk decreases by 17% for every 1-MET (3.5 mL/kg/min) difference in aerobic capacity in healthy females(36). Further, this difference in VO_2peak was observed on a background of stable HR and lower LVEF, implying a non-cardiac contribution i.e. skeletal muscle. Similar findings have been observed in elderly patients with heart failure and preserved ejection fraction. Haykowsky *et al* studied 40 elderly patients after 4 months of supervised endurance training compared to controls, observing improved peak VO_2 in trained vs controls (16.3 +/- 2.6 vs 13.1 +/- 3.4 ml/kg/min)(37). Importantly, this was observed on a background of improved peak A-VO_2 diff but no significant improvements in peak LVEDV, stroke volume or cardiac output. This key finding suggests improved skeletal muscle functioning as a result of exercise training, and that rather than cardiovascular improvements, adaptations to other components of the oxygen cascade may be more important. Future work

should focus on better understanding of the contribution of both cardiovascular and systemic effects of exercise interventions.

2.4.3 Strengths and limitations

This study is the first to examine the long-term effects of trastuzumab using cardiac MRI and aerobic capacity. While access to both of these modalities may present challenges at some centers, both represent ‘gold standard’ measures of LV remodeling and cardiorespiratory function in this and other clinical settings. Further, cardiac MRI is established as a powerful research tool, given the high reproducibility of cardiac measures allowing a significant reduction in the sample size(38, 39).

The main limitation of this project is the lack of healthy, age-matched controls with serial scanning performed at similar timepoints. This hampers our ability to elucidate the relative contributions of exercise, chemotherapy agents and aging. Further, late gadolinium contrast imaging was not available at the time of the original study, and given the lack of a baseline comparator, was not peformed in this follow-up project. Accordingly, our ability to examine other characteristics such as acute inflammation, scarring or evolving fibrosis was limited. Given our findings, future projects should include this additional imaging technique.

We did not administer a self-reported exercise measurement tool as none were administered as part of the original project. Further, given the design of this project no true control group was possible. Future projects would benefit by including a validated patient-reported outcomes tool completed at various timepoints, and in the years of survivorship to further examine the longer-term

effects of toxic cancer therapies. Additionally, prospective examination with randomized controlled trials would support the findings of such projects.

2.4.4 Conclusions

In summary, we found that, on a background of normal LVEF, long-term and significant alterations in LV geometry and significant impairment of cardiorespiratory function are present in EBC patients four years following exposure to trastuzumab-based chemotherapy. Adherence to regular aerobic exercise may attenuate these deleterious effects.

Table 2.1: Participant characteristics

Median age, range (years)	58 (47 – 76)
Median time since last trastuzumab infusion, range (years)	3.9 (3.0 – 5.3)
Mean chemotherapy dose, range (mg)	Fluoruracil: 3887 (2850 – 5100) Epirubicin: 778 (564 – 1020) Doxorubicin: 462 (388 – 534) Cyclophosphamide: 4060 (1520 – 5900) Carboplatin: 3819 (2981 – 5443) Docetaxel: 650 (180 – 965) Trastuzumab: 6721 (1560 – 11,320)
Mean left breast or chest wall radiotherapy dose, range (cGy), n = 4	4875 (4500 – 6000)
Current smoking status	10 = never, 4 = ex-smokers, 2 = current
Karnofsky performance status	100%
Mean hemoglobin, range (g/L)	140.0 (125 – 158)

n=16

Table 2.2: LV morphology and function in breast cancer patients prior, 4 months and 4 years after trastuzumab therapy

	Baseline	4 months	4 years	Baseline vs 4 months p-value	Baseline vs 4 years p-value	4 months vs 4 years p-value
LVEF (%)	61 ± 5	55 ± 4	60 ± 6	0.04	0.88	0.12
LVEDV (mL)	138 ± 27	153 ± 20	128 ± 27	0.21	0.47	0.02
LVESV (mL)	54 ± 11	68 ± 12	52 ± 15	0.02	0.89	0.003
LVSV (mL)	84 ± 20	85 ± 13	76 ± 16	0.97	0.35	0.25
LV Mass (g)	78 ± 16	83 ± 14	69 ± 14	0.65	0.22	0.03
M/V ratio	0.57 ± 0.06	0.54 ± 0.09	0.54 ± 0.05	0.63	0.59	0.99
Long strain	-17.4 ± 2.2	-15.7 ± 2.1	-16.3 ± 1.5	0.05	0.29	0.66

n=16; Values are mean and standard deviation

Abbreviations: LVEF, left ventricular ejection fraction; LVEDV, left ventricular end-diastolic volume; LVESV, left ventricular end-systolic volume; LVSV, left ventricular stroke volume; LV mass, left ventricular mass; M/V ratio, mass to volume ratio.

Table 2.3: LV morphology and function in exercise adherent breast cancer patients prior, 4 months and 4 years after trastuzumab therapy

	Baseline	4 months	4 years	Baseline vs 4 months p-value	Baseline vs 4 years p-value	4 months vs 4 years p-value
LVEF (%)	58 ± 4	54 ± 5	59 ± 7	0.52	0.94	0.34
LVEDV (mL)	125 ± 29	147 ± 22	123 ± 21	0.29	0.99	0.24
LVESV (mL)	53 ± 17	68 ± 18	50 ± 14	0.30	0.96	0.20
LVSV (mL)	72 ± 13	79 ± 6	73 ± 14	0.52	0.99	0.60
LV Mass (g)	72 ± 11	79 ± 12	65 ± 11	0.52	0.52	0.09
M/V ratio	0.59 ± 0.08	0.54 ± 0.06	0.53 ± 0.06	0.50	0.32	0.94
Long strain	-17.0 ± 1.9	-16.2 ± 2.9	-17.0 ± 2.0	0.83	1.0	0.82

n=6; Values are mean and standard deviation

Abbreviations: LVEF, left ventricular ejection fraction; LVEDV, left ventricular end-diastolic volume; LVESV, left ventricular end-systolic volume; LVSV, left ventricular stroke volume; LV mass, left ventricular mass; M/V ratio, mass to volume ratio.

Table 2.4: LV morphology and function in exercise non-adherent breast cancer patients prior, 4 months and 4 years after trastuzumab therapy

	Baseline	4 months	4 years	Baseline vs 4 months p-value	Baseline vs 4 years p-value	4 months vs 4 years p-value
LVEF (%)	62 ± 5	56 ± 4	60 ± 6.0	0.06	0.63	0.32
LVEDV (mL)	146 ± 23	157 ± 19	131 ± 30	0.59	0.35	0.06
LVESV (mL)	55 ± 7	68 ± 7	53 ± 16	0.03	0.93	0.02
LVSV (mL)	91 ± 20	89 ± 15	78 ± 18	0.96	0.22	0.34
LV Mass (g)	82 ± 18	85 ± 15	72 ± 16	0.90	0.39	0.20
M/V ratio	0.55 ± 0.05	0.55 ± 0.11	0.55 ± 0.04	0.96	0.99	0.98
Long strain	-17.6 ± 2.5	-15.4 ± 1.5	-15.9± 1.04	0.03	0.09	0.82

n=10; Values are mean and standard deviation

Abbreviations: LVEF, left ventricular ejection fraction; LVEDV, left ventricular end-diastolic volume; LVESV, left ventricular end-systolic volume; LVSV, left ventricular stroke volume; LV mass, left ventricular mass; M/V ratio, mass to volume ratio.

Table 2.5: Peak exercise cardiorespiratory capacity and hemodynamics

	Baseline	4 month	4 years	Baseline vs 4 months p-value	Baseline vs 4 years p-value	4 months vs 4 years p-value
HR	166 ± 13	163 ± 16	158 ± 19	0.85	0.34	0.66
SBP (mmHg)	183 ± 24	175 ± 24	172 ± 15	0.53	0.29	0.90
DBP (mmHg)	86 ± 8	81 ± 7	89 ± 9	0.16	0.55	0.02
RER	1.18 ± 0.10	1.20 ± 0.11	1.14 ± 0.08	0.83	0.50	0.21
RPE (10 scale)	8.6 ± 1.7	8.8 ± 1.8	8.4 ± 1.4	0.97	0.81	0.68
VO_2 (L/min)	1.49 ± 0.39	1.58 ± 0.35	1.64 ± 0.30	0.76	0.43	0.85
VO_2 (mL/kg/min)	20.0 ± 4.3	22.0 ± 6.6	22.4 ± 5.0	0.44	0.55	0.98
PO (watts)	103 ± 24	104 ± 22	120 ± 30	0.99	0.17	0.20

n=16

Abbreviations: HR, heart rate; SBP, systolic blood pressure; DBP, diastolic blood pressure; RER, respiratory exchange ratio; RPE, rate of perceived exertion; VO_2, oxygen consumption, PO, power output.

Table 2.6: Peak exercise cardiorespiratory capacity and hemodynamics in breast cancer patients who were adherent with exercise program

	Baseline	4 month	4 years	Baseline vs 4 months p-value	Baseline vs 4 years p-value	4 months vs 4 years p-value
HR	169 ± 13	162 ± 17	160 ± 18	0.76	0.63	0.97
SBP (mmHg)	183 ± 11	182 ± 18	173 ± 10	0.99	0.44	0.50
DBP (mmHg)	87 ± 6	83 ± 4	85 ± 8	0.67	0.90	0.90
RER	1.23 ± 0.09	1.22 ± 0.14	1.14 ± 0.10	0.98	0.40	0.50
RPE (10 scale)	8.8 ± 1.9	9.2 ± 1.2	8.0 ± 1.5	0.93	0.64	0.43
VO2 (L/min)	1.32 ± 0.22	1.70 ± 0.34	1.62 ± 0.26	0.08	0.18	0.89
VO2 (mL/kg/min)	20.9 ± 5.8	26.4 ± 5.9	24.9 ± 6.6	0.28	0.49	0.91
PO (watts)	93 ± 17	100 ± 15	115 ± 29	0.82	0.20	0.47

n=6

Abbreviations: HR, heart rate; SBP, systolic blood pressure; DBP, diastolic blood pressure; RER, respiratory exchange ratio; RPE, rate of perceived exertion; VO_2, oxygen consumption, PO, power output.

Table 2.7: Peak exercise cardiorespiratory capacity and hemodynamics in breast cancer patients who did not adhere with exercise program

	Baseline	4 month	4 years	Baseline vs 4 months p-value	Baseline vs 4 years p-value	4 months vs 4 years p-value
HR	165 ± 13	164 ± 16	157 ± 21	0.99	0.57	0.65
SBP (mmHg)	183 ± 30	171 ± 27	171 ± 17	0.52	0.52	1.00
DBP (mmHg)	86 ± 9	79 ± 9	92 ± 9	0.23	0.31	0.01
RER	1.15 ± 0.10	1.19 ± 0.10	1.15 ± 0.06	0.58	0.97	0.43
RPE (10 scale)	9.0 ± 0.816	9.1 ± 1.215	8.7 ± 1.38	1.0	0.99	0.99
VO_2 (L/min)	1.59 ± 0.44	1.50 ± 0.35	1.66 ± 0.33	0.87	0.92	0.65
VO_2 (mL/kg/min)	19.5 ± 3.8	19.4 ± 5.8	20.8 ± 3.33	0.99	0.77	0.74
PO (watts)	110 ± 26	107 ± 30	123 ± 32	0.97	0.55	0.41

n=10

Abbreviations: HR, heart rate; SBP, systolic blood pressure; DBP, diastolic blood pressure; RER, respiratory exchange ratio; RPE, rate of perceived exertion; VO_2, oxygen consumption, PO, power output.

Figure 2.1: Effect of exercise adherence on resting LVESV (mL)

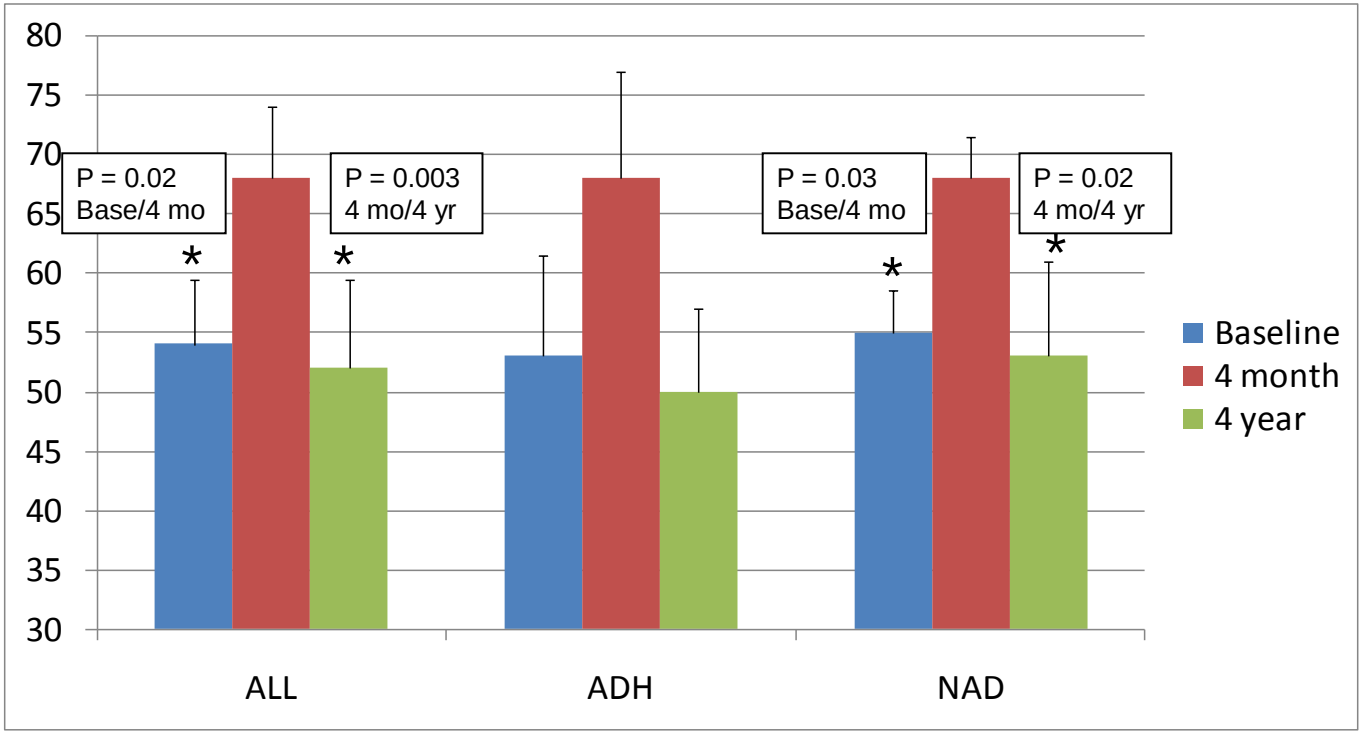


Figure 2.2: Effect of exercise adherence on resting LVEDV (mL)

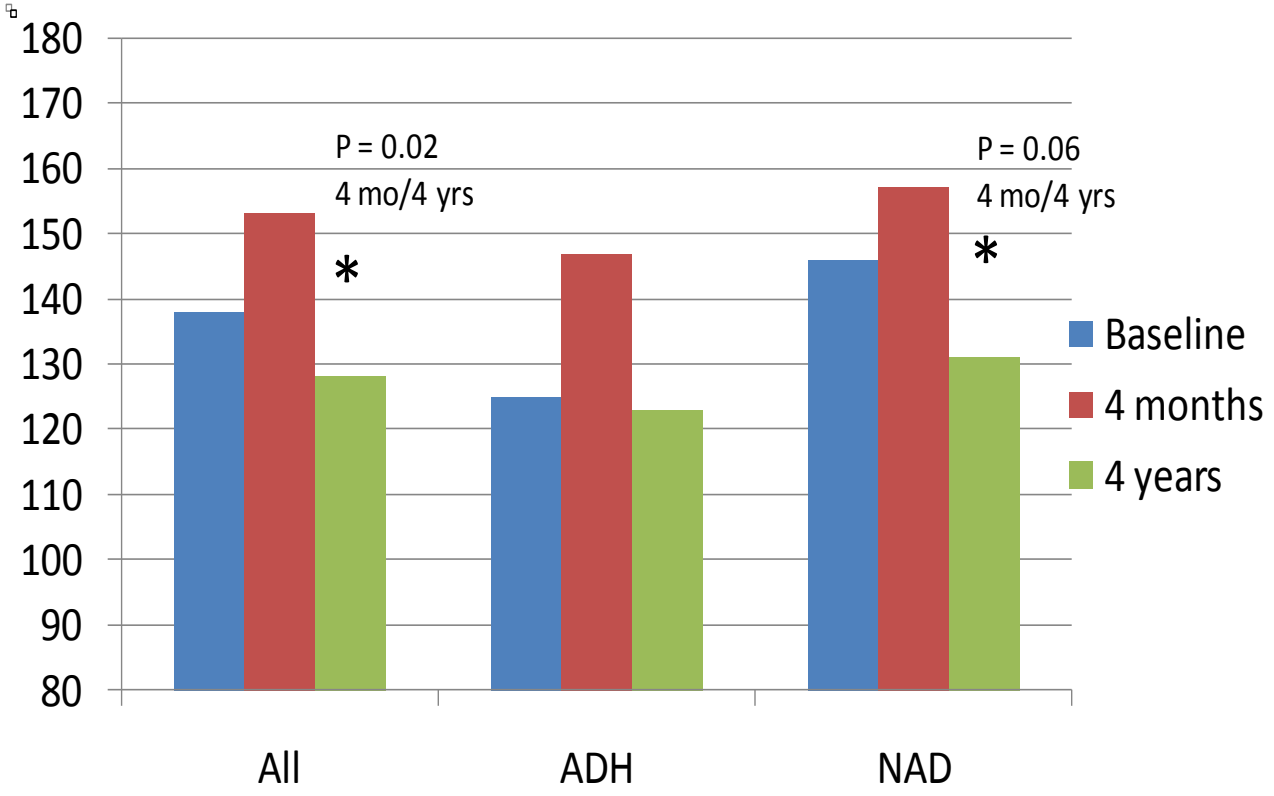


Figure 2.3: Effect of exercise adherence on LV mass (g)

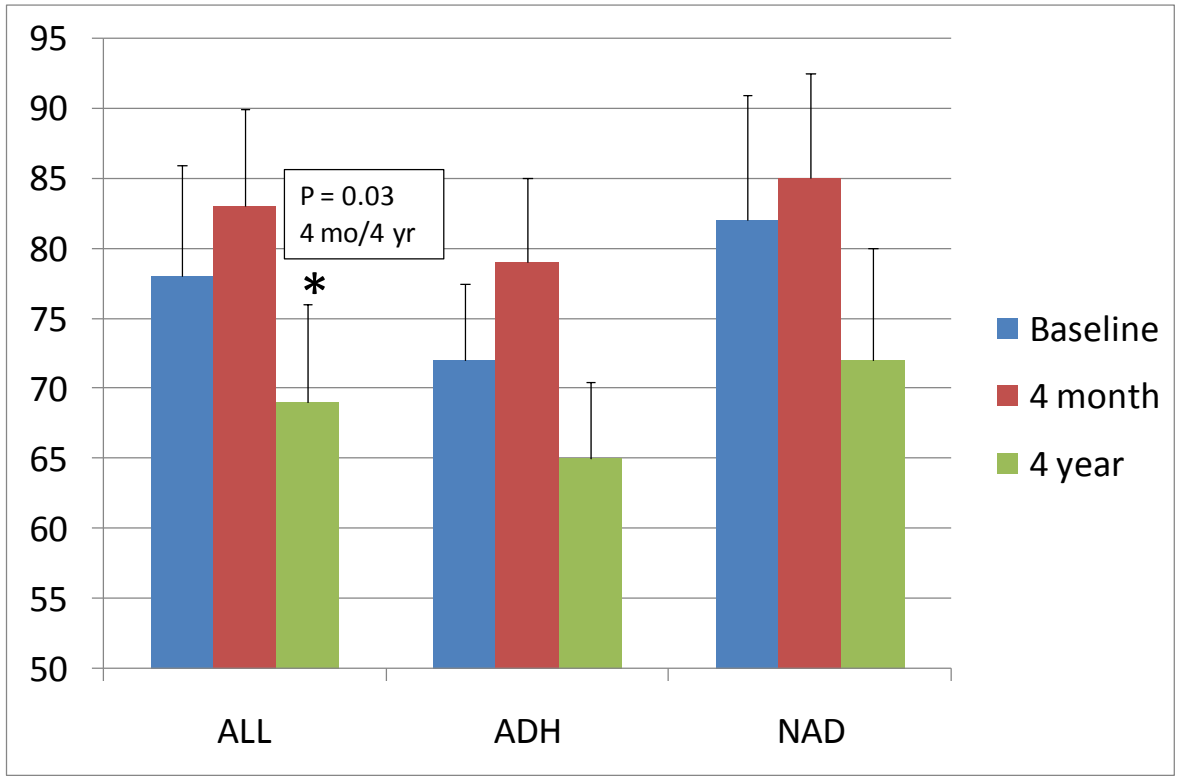
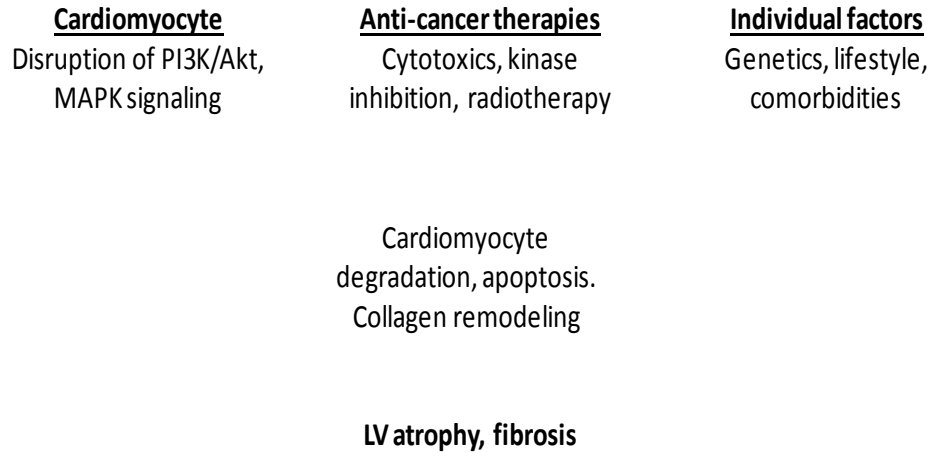


Figure 2.4: Potential factors involved in reduced LV mass in breast cancer patients



2.5 References

1. Romond EH, Perez EA, Bryant J, Suman VJ, Geyer CE, Jr., Davidson NE, et al. Trastuzumab plus adjuvant chemotherapy for operable HER2-positive breast cancer. N Engl J Med. 2005 Oct 20;353(16):1673-84. PubMed PMID: 16236738. Epub 2005/10/21. eng.

2. Slamon D, Eiermann W, Robert N, Pienkowski T, Martin M, Press M, et al. Adjuvant trastuzumab in HER2-positive breast cancer. N Engl J Med. 2011 Oct 6;365(14):1273-83. PubMed PMID: 21991949. Pubmed Central PMCID: 3268553.

3. Canadian Cancer Statistics. Toronto, Canada2010.

4. Patnaik JL, Byers T, DiGuiseppi C, Dabelea D, Denberg TD. Cardiovascular disease competes with breast cancer as the leading cause of death for older females diagnosed with breast cancer: a retrospective cohort study. Breast cancer research : BCR. 2011;13(3):R64. PubMed PMID: 21689398. Pubmed Central PMCID: 3218953.

5. Cohn JN, Ferrari R, Sharpe N. Cardiac remodeling--concepts and clinical implications: a consensus paper from an international forum on cardiac remodeling. Behalf of an International Forum on Cardiac Remodeling. J Am Coll Cardiol. 2000 Mar 1;35(3):569-82. PubMed PMID: 10716457. Epub 2000/03/15. eng.

6. Mann DL, Bristow MR. Mechanisms and Models in Heart Failure: The Biomechanical Model and Beyond. Circulation. 2005 May 31, 2005;111(21):2837-49.

7. Vasan RS, Larson MG, Benjamin EJ, Evans JC, Levy D. Left ventricular dilatation and the risk of congestive heart failure in people without myocardial infarction. N Engl J Med. 1997 May 8;336(19):1350-5. PubMed PMID: 9134875. Epub 1997/05/08. eng.

8. Haykowsky MJ, Mackey JR, Thompson RB, Jones LW, Paterson DI. Adjuvant trastuzumab induces ventricular remodeling despite aerobic exercise training. Clin Cancer Res. 2009 Aug 1;15(15):4963-7. PubMed PMID: 19622583.

9. Mandic S, Tymchak W, Kim D, Daub B, Quinney HA, Taylor D, et al. Effects of aerobic or aerobic and resistance training on cardiorespiratory and skeletal muscle function in heart failure: a randomized controlled pilot trial. Clinical rehabilitation. 2009 Mar;23(3):207-16. PubMed PMID: 19218296.

10. Schag CC, Heinrich RL, Ganz PA. Karnofsky performance status revisited: reliability, validity, and guidelines. J Clin Oncol. 1984 Mar;2(3):187-93. PubMed PMID: 6699671.

11. Perez EA, Romond EH, Suman VJ, Jeong JH, Davidson NE, Geyer CE, Jr., et al. Four-year follow-up of trastuzumab plus adjuvant chemotherapy for operable human epidermal growth factor receptor 2-positive breast cancer: joint analysis of data from NCCTG N9831 and NSABP B-31. J Clin Oncol. 2011 Sep 1;29(25):3366-73. PubMed PMID: 21768458. Pubmed Central PMCID: 3164242.

12. Romond EH, Jeong JH, Rastogi P, Swain SM, Geyer CE, Jr., Ewer MS, et al. 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 growth factor receptor 2-positive breast cancer. J Clin Oncol. 2012 Nov 1;30(31):3792-9. PubMed PMID: 22987084. Pubmed Central PMCID: 3478574.

13. Nag AC. Study of non-muscle cells of the adult mammalian heart: a fine structural analysis and distribution. Cytobios. 1980;28(109):41-61. PubMed PMID: 7428441.

14. Lemmens K, Doggen K, De Keulenaer GW. Role of Neuregulin-1/ErbB Signaling in Cardiovascular Physiology and Disease: Implications for Therapy of Heart Failure. Circulation. 2007 August 21, 2007;116(8):954-60.

15. Pretorius L, Owen KL, Jennings GL, McMullen JR. Promoting physiological hypertrophy in the failing heart. Clinical and experimental pharmacology & physiology. 2008 Apr;35(4):438-41. PubMed PMID: 18307737.

16. McMullen JR, Shioi T, Huang WY, Zhang L, Tarnavski O, Bisping E, et al. The insulin-like growth factor 1 receptor induces physiological heart growth via the phosphoinositide 3-kinase(p110alpha) pathway. The Journal of biological chemistry. 2004 Feb 6;279(6):4782-93. PubMed PMID: 14597618.

17. Dorfman TA, Levine BD, Tillery T, Peshock RM, Hastings JL, Schneider SM, et al. Cardiac atrophy in women following bed rest. Journal of Applied Physiology. 2007 July 2007;103(1):8-16.

18. Mason C, Alfano CM, Smith AW, Wang CY, Neuhouser ML, Duggan C, et al. Long-term physical activity trends in breast cancer survivors. Cancer epidemiology, biomarkers & prevention : a publication of the American

Association for Cancer Research, cosponsored by the American Society of Preventive Oncology. 2013 Apr 10. PubMed PMID: 23576689.

19. Tanaka H, Seals DR. Invited Review: Dynamic exercise performance in Masters athletes: insight into the effects of primary human aging on physiological functional capacity. Journal of Applied Physiology. 2003 November 1, 2003;95(5):2152-62.

20. Owen KL, Pretorius L, McMullen JR. The protective effects of exercise and phosphoinositide 3-kinase (p110alpha) in the failing heart. Clin Sci (Lond). 2009 Mar;116(5):365-75. PubMed PMID: 19175355.

21. Scott JM, Lakoski S, Mackey JR, Douglas PS, Haykowsky MJ, Jones LW. The potential role of aerobic exercise to modulate cardiotoxicity of molecularly targeted cancer therapeutics. Oncologist. 2013;18(2):221-31. PubMed PMID: 23335619.

22. Lipshultz SE, Landy DC, Lopez-Mitnik G, Lipsitz SR, Hinkle AS, Constine LS, et al. Cardiovascular status of childhood cancer survivors exposed and unexposed to cardiotoxic therapy. J Clin Oncol. 2012 Apr 1;30(10):1050-7. PubMed PMID: 22393080. Pubmed Central PMCID: 3341149.

23. Armstrong GT, Plana JC, Zhang N, Srivastava D, Green DM, Ness KK, et al. Screening adult survivors of childhood cancer for cardiomyopathy: comparison of echocardiography and cardiac magnetic resonance imaging. J Clin Oncol. 2012 Aug 10;30(23):2876-84. PubMed PMID: 22802310.

24. Baskin KK, Taegtmeyer H. Taking pressure off the heart: the ins and outs of atrophic remodelling. Cardiovascular research. 2011 May 1;90(2):243-50. PubMed PMID: 21354996. Pubmed Central PMCID: 3115281.

25. Oreopoulos A, Padwal R, Kalantar-Zadeh K, Fonarow GC, Norris CM, McAlister FA. Body mass index and mortality in heart failure: a meta-analysis. Am Heart J. 2008 Jul;156(1):13-22. PubMed PMID: 18585492. Epub 2008/07/01. eng.

26. Prado CM, Baracos VE, McCargar LJ, Reiman T, Mourtzakis M, Tonkin K, et al. Sarcopenia as a determinant of chemotherapy toxicity and time to tumor progression in metastatic breast cancer patients receiving capecitabine treatment. Clin Cancer Res. 2009 Apr 15;15(8):2920-6. PubMed PMID: 19351764. Epub 2009/04/09. eng.

27. Prado CMM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L, et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. The Lancet Oncology. 2008;9(7):629-35.

28. Cosper PF, Leinwand LA. Cancer Causes Cardiac Atrophy and Autophagy in a Sexually Dimorphic Manner. Cancer research. 2011 March 1, 2011;71(5):1710-20.

29. Biernacka A, Frangogiannis NG. Aging and Cardiac Fibrosis. Aging Dis. Apr;2(2):158-73. PubMed PMID: 21837283. Epub 2011/08/13. Eng.

30. North BJ, Sinclair DA. The intersection between aging and cardiovascular disease. Circ Res. Apr 13;110(8):1097-108. PubMed PMID: 22499900. Epub 2012/04/14. eng.

31. Hare JL, Brown JK, Leano R, Jenkins C, Woodward N, Marwick TH. Use of myocardial deformation imaging to detect preclinical myocardial dysfunction before conventional measures in patients undergoing breast cancer treatment with trastuzumab. American heart journal. 2009 Aug;158(2):294-301. PubMed PMID: 19619708. Epub 2009/07/22. eng.

32. Sawaya H, Sebag IA, Plana JC, Januzzi JL, Ky B, Cohen V, et al. Early Detection and Prediction of Cardiotoxicity in Chemotherapy-Treated Patients. The American Journal of Cardiology.107(9):1375-80.

33. Fitzgerald MD, Tanaka H, Tran ZV, Seals DR. Age-related declines in maximal aerobic capacity in regularly exercising vs. sedentary women: a meta-analysis. Journal of Applied Physiology. 1997 July 1, 1997;83(1):160-5.

34. Jones LW, Courneya KS, Mackey JR, Muss HB, Pituskin EN, Scott JM, et al. Cardiopulmonary Function and Age-Related Decline Across the Breast Cancer Survivorship Continuum. Journal of Clinical Oncology. 2012 July 10, 2012;30(20):2530-7.

35. Jones LW, Liang Y, Pituskin EN, Battaglini CL, Scott JM, Hornsby WE, et al. Effect of exercise training on peak oxygen consumption in patients with cancer: a meta-analysis. Oncologist.16(1):112-20. PubMed PMID: 21212429. Epub 2011/01/08. eng.

36. Gulati M, Pandey DK, Arnsdorf MF, Lauderdale DS, Thisted RA, Wicklund RH, et al. Exercise capacity and the risk of death in women: the St James Women Take Heart Project. Circulation. 2003 Sep 30;108(13):1554-9. PubMed PMID: 12975254.

37. Haykowsky MJ, Brubaker PH, Stewart KP, Morgan TM, Eggebeen J, Kitzman DW. Effect of Endurance Training on the Determinants of Peak Exercise Oxygen Consumption in Elderly Patients With Stable Compensated Heart Failure and Preserved Ejection Fraction. Journal of the American College of Cardiology. 2012 7/10/;60(2):120-8.

38. Bellenger N, Pennell D. Magnetic resonance imaging in cardiology. J R Coll Physicians Lond. 1999 Jan-Feb;33(1):12-8. PubMed PMID: 10192063. Epub 1999/04/07. eng.

39. Bellenger NG, Davies LC, Francis JM, Coats AJ, Pennell DJ. Reduction in sample size for studies of remodeling in heart failure by the use of cardiovascular magnetic resonance. Journal of cardiovascular magnetic resonance : official journal of the Society for Cardiovascular Magnetic Resonance. 2000;2(4):271-8. PubMed PMID: 11545126. Epub 2001/09/08. eng.

Chapter 3

A prospective evaluation of LV remodeling in breast cancer patients receiving trastuzumab-based chemotherapy and randomized to receive conventional heart failure pharmacotherapy or placebo.

3.1 Introduction

Trastuzumab is a proven therapy for HER2+ breast cancer, improving overall survival compared to non-trastuzumab containing chemotherapy regimens(1, 2). As a result of these and other highly effective adjuvant therapies, five-year survival rates approach 88% among EBC patients(3). While trastuzumab is a vital anti-cancer therapy, significant cardiac safety concerns have been observed. In population-based studies, longer-term observations indicate fourfold increase of HF or cardiomyopathy in women who received trastuzumab alone, and sevenfold for anthracycline plus trastuzumab over time(4). Accordingly, detection and prevention of such negative sequelae are of great clinical interest. However, conventional imaging modalities routinely available in the oncology clinic are hampered by poor accuracy, reproducibility and resolution(5) and are insensitive to early negative changes in cardiac function and remodeling. Cardiac MRI is established as the gold standard for quantifying LV volumes and patterns of remodeling(6-8). Detrimental LV remodeling is established as an early indicator of cardiac injury, progressing to worsening cardiac function and overt HF in the general population(9-11). In Study One, we observed LV remodeling

occurs as early as four months of trastuzumab therapy, in spite of lifestyle modifications i.e. aerobic exercise training(12).

Overall, the work of our group and others has convincingly shown that trastuzumab-related LV remodeling and/or cardiotoxicity occurs early and may be permanent(12-14), therefore, definitive preventive approaches are necessary. National evidence-based guidelines recommend pharmacotherapy in HF patients (15-17). Angiotensin-converting enzyme inhibitors (ACEI) are first-line therapy, slowing progress of LV dilation, reducing preload and afterload, reducing sympathetic activation and preventing angiotensin II from exacerbating further remodeling(18). Beta-blockers have been shown to improve cardiac function and reduce mortality in HF patients; while the mechanism is not fully understood, blockade of excessive and chronic sympathetic influences may alleviate further exacerbation/remodeling.

Heart failure therapy guidelines have been extrapolated to cancer patients with cardiotoxicity(19) with data gradually accumulating to suggest that such therapy may be beneficial in anthracycline-induced cardiomyopathy(20-22). Carvedilol has been shown to be an effective single-agent therapy in anthracycline-induced cardiomyopathy in various malignancies(23). Cardinale *et al* demonstrated that that ACEI can prevent a decline in LVEF and cardiac events in heterogeneous cancer patient groups receiving high dose anthracyclines(24). However, to date, no investigators have specifically examined the effect of ACEI or BB in preventing cardiotoxicity in the setting of trastuzumab-based therapy. Accordingly, the purpose of this randomized study was to examine the effects of 3

months of ACEI or BB vs placebo on LV remodeling in breast cancer patients receiving trastuzumab-based therapy. Our hypothesis was that standard HF pharmacotherapy would prevent trastuzumab mediated LV remodeling compared to placebo.

3.2 Methods

This substudy examined baseline and 3 month clinical and MRI data in patients participating in MANTICORE (Multidisciplinary Approach to Novel Therapies in Cardiology Oncology Research), a randomized trial to determine if conventional HF pharmacotherapy (perindopril or bisoprolol versus placebo) can prevent trastuzumab-mediated LV remodeling, as measured with cardiac MRI, among patients with HER2+ early breast cancer.

3.2.1 Participants

3.2.1.1 *Inclusion criteria:* patients with HER-2 positive EBC from the outpatient clinic of the Cross Cancer Institute (CCI) scheduled to receive 1 year of trastuzumab; age > 18 years; able to give informed consent; no contraindication to MRI; estimated glomerular filtration rate (GFR) > 30ml/min due to risk of nephrogenic systemic fibrosis from MRI contrast.

3.2.1.2 *Exclusion criteria:* known contraindication to ACEI therapy (i.e., baseline potassium > 5.3mmol/L, glomerular filtration rate < 30 mls/min); known contraindication to BB therapy (i.e., heart rate < 50/min, severe asthma); current treatment with ACEI or BB therapy for other indication; history of HF, cardiomyopathy, or baseline LVEF < 50%; history of uncontrolled hypertension or myocardial infarction.

Following written informed consent, the participant was scheduled for the following baseline assessments:

3.2.3 Clinical assessment

Participants underwent clinical examinations including weight, blood pressure, pulse, respiratory rate, comorbidities and concurrent medications. Specific inquiry regarding smoking status and alcohol use was undertaken. Review of current malignancy and cardiac disease history was performed.

3.2.4 Cardiac MRI LV function and morphology

Assessment of LV volumes, mass, and LVEF was performed with a 1.5 Tesla Magnetom Sonata scanner (Siemens Medical Solutions). Steady-state free precession cine techniques acquired parallel, 8-mm-thick short-axis slices (plus a 2-mm gap) throughout the LV. Standard post-processing software (Syngo MR;Siemens) was used to calculate LV volumes, mass and function. Heart rate and blood pressure were monitored throughout the examination.

3.2.5 Randomization

Participants were randomized in a 1:1:1 ratio to ACEI, BB or placebo using a secure internet randomization service provided by EPICORE (Epidemiology Coordinating and Research Centre). For this study and analysis, the ACEI and BB groups were combined as a 'pharmacotherapy' group. There was no run-in period. Study medication was taken daily *per os* consisting of either placebo, perindopril 2 mg or bisoprolol 2.5 mg. Weekly up-titration occurred following clinical assessments (blood pressure, heart rate and symptom report) and laboratory (creatinine and electrolytes) to a potential maximum dose

of perindopril 8mg or bisoprolol 10mg. All personnel were blinded to drug allocation. At the three month time point, participants underwent the same assessments as performed at baseline (Figure 3.1).

3.2.6 Statistical analysis

Data were collated by EPICORE as of July 31, 2012, drawn from case report forms of the first 31 study participants. Data were anonymized to maintain ongoing blinding of study personnel involved in the main MANTICORE study. For each of the outcome variables, a random effects model was fitted to the data, and between and within comparison were made. Analysis was performed using STATISTICA 7 (StatSoft, Tulsa, OK) software. $P < 0.05$ was considered significant for all statistical measures.

3.3 Results

3.3.1 Participant characteristics

Thirty-one participants were recruited between September 23, 2010 and October 27, 2011 (Figure 3.2). All participants completed baseline and 3 month assessments, with 20 assigned to pharmacotherapy and 11 to placebo. No adverse events were reported, and all participants continued to take the supplied capsules through the study period. Both pharmacotherapy and placebo groups were similar in terms of clinical demographics (Table 3.1). Mean age was 50.1 ± 8.8 years in the pharmacotherapy group versus 48.3 ± 6.1in placebo. Similar mean height and weight were observed in each group, with similar mean cumulative dose of trastuzumab delivered (1381mg pharmacotherapy versus 1305 mg placebo). Chemotherapy doses were not available at the time of this analysis. Fifteen of the

20 pharmacotherapy and 9 of the 11 placebo participants had undergone definitive surgery; the remaining patients received neoadjuvant therapy with surgical intervention planned upon completion of chemotherapy. In terms of cardiovascular risk factors, the majority of patients were never-smokers, and only 2 patients in each group reported a current smoking habit. Vital signs (heart rate, systolic and diastolic blood pressure) at baseline were equivalent between groups.

3.3.2 Cardiac MRI LV function and morphology

Baseline comparisons of the pharmacotherapy and placebo groups reveal similar LVEF (62.1 ± 1.1 mL vs 61.6 ± 1.4 mL), LV mass (91 ± 4.2 g versus 94.8 ± 5.6 g) and LVESV (42.7 ± 3.1 mL versus 52.3 ± 4.1 mL). However, the baseline LVEDV was lower in the pharmacotherapy group compared to the placebo (112.0 ± 6.7 versus 135.8 ± 8.8 mL, $p = 0.031$).

Examining within-group changes over time (Table 3.2), an absolute 8% decline in LVEF was found in the placebo group from baseline to 3 months ($p < 0.001$) secondary to an increase in LVESV of 13mL ($p = <0.001$). By comparison, LVEF in the pharmacotherapy group declined by 3.4% from baseline to 3 months ($p = 0.002$), while LVEDV and LVESV increased (14mL and 9.3mL, $p = 0.006$ and <0.001, respectively).

At three months examining between group changes, a 5% difference in LVEF was found in the placebo compared to the pharmacotherapy group ($p = 0.004$). Furthermore, both LV end-diastolic (+14 mL) and LV end-systolic volume (+13 mL) increased. No significant differences in HR, SBP, DBP or LV mass were observed (Table 3.3).

3.4 Discussion

3.4.1 Effects of heart failure pharmacotherapy on LV function

This randomized controlled study examined the effects of 3 months of ACEI or BB versus placebo on LV remodeling in breast cancer patients receiving trastuzumab-based therapy. The major finding was that compared to the pharmacotherapy group, a significant decline in LVEF occurred in the placebo group secondary to LV cavity dilation (Figure 3.2). Similar to the observations in Study One, LVEF decline occurs early, within 3 months exposure to trastuzumab-based therapy. Importantly, the 8% difference in LVEF observed here approaches the critical (10%) cut-point for stopping and re-evaluating trastuzumab therapy. The significance of early LVEF decline is not well described, however Wadhwa *et al* reported that among patients who developed cardiomyopathy, within 3 months of trastuzumab exposure LVEF drop was observed and was progressive(13). In these patients, the average duration of trastuzumab was 6 ± 3 months compared to those without cardiomyopathy who received the full 12 month planned doses. The long-term consequences of receiving less than planned dose of trastuzumab are unknown, but potentially convey negative long-term impacts to survival. Additionally, a decline in LVEF is now increasingly understood as a risk factor for late cardiomyopathy, and even with optimal pharmacotherapy and medical support, LV function may not recover. Among patients referred for LVEF decline secondary to anthracyclines, Cardinale *et al* observed that early intervention predicts long term recovery(25). In 36 patients with trastuzumab-related cardiomyopathy, with optimal pharmacologic support 4

patients were able to complete full therapy; the remaining 32 showed no improvement or further LVEF decline(13). Taken together, the importance of early recognition of treatment-related cardiomyopathy cannot be understated, and hence the need for sensitive diagnostic imaging approaches, such as cMRI.

The underlying mechanisms contributing to trastuzumab-related LVEF decline are not well known in humans. Preclinical studies have shown the important role of HER2/erbB2 in maintaining cardiac function. Specifically, ventricular restricted deletion of the erbB2 receptor has been shown to result in dilated cardiomyopathy (increased LV cavity size and decreased LVEF, Figure 1.2) in mice(26). Furthermore, isolated murine cardiomyocytes were more susceptible to anthracycline mediated toxicity. As this cardiotoxic interaction was also observed in humans(27), clinical adjuvant protocols require that anthracyclines and trastuzumab to be given sequentially rather than co-administered, however, other chemotherapeutic agents are co-administered with trastuzumab, including taxanes and platinum salts, with varying degrees of toxic effects. Debilitating side effects such as poor appetite, nausea, vomiting and diarrhea may contribute to episodic dehydration and decreased preload, while fluid retention associated with taxane-induced capillary leak syndrome could increase peripheral vascular resistance and afterload conditions.

3.4.2 Effects of heart failure pharmacotherapy on LV morphology

Examining LVESV reveals a 13mL dilation comparing pharmacotherapy to placebo ($p = 0.009$), which may have ultimately negatively influenced LV systolic function and LVEF (Figure 3.4). These observations are consistent with

our earlier work, where we observed LV cavity dilation and decreased systolic function with four months of exposure to trastuzumab(12). In the current study, interpretations of LVEDV results are hampered by an imbalance at baseline between the pharmacotherapy group versus placebo. Reasons for this are unknown, but could include the small sample size included, influence of prior exposure to anthracyclines in sequential therapy, or a yet undiscovered raw data error or outlier. Nonetheless, given the significant differences in LV volumes at baseline between the pharmacotherapy and placebo groups, the three month observations of this study should be interpreted cautiously.

In this study, pharmacotherapy appears effective in attenuating LV dysfunction associated with trastuzumab-based therapy. International evidence-based guidelines recommend HF pharmacotherapy as a proven approach in treating LV dysfunction in various patient groups(15-17) and have been extrapolated to the treatment of cancer patients with cardiotoxicity(19). In blocking the PI3K/Akt pathway, trastuzumab negatively impacts cardiomyocyte survival; the parallel Ras/Raf/MEK/MAPK/VEGF pathway also effected by trastuzumab blocks normal cellular proliferation activities and angiogenesis(28). Loss of cardiomyocytes may initiate negative LV remodeling and negative adaptation, including activation of the renin-angiotensin-aldosterone system and fibrosis(29). As a result, the LV may dilate, diastolic performance may be reduced with increasing fibrosis, and LV mass decline with reduced cardiomyocytes (Figure 3.5). Negative remodeling may ultimately evolve to declining pump performance, and a HF spectrum characterized by overlapping systolic and

diastolic dysfunction(30). In preventing or attenuating trastuzumab-related cardiac dysfunction, ACEI may reduce sympathetic activation, reduce preload/afterload and slow the rate of LV remodeling(18, 31). Beta-blockers may also be beneficial in this setting by enhancing cardiac perfusion by rate reduction. We did not observe significant differences in HR within or between groups over time, an effect that may have been diluted as a result of combining two different therapies into one group for this analysis. Also, SBP decreased by 5mmHg over 3 months with pharmacotherapy accompanied by an approximate 14mL difference in 3 month LVEDV measures between pharmacotherapy and placebo groups, suggesting that LVEDV systolic wall stress may be attenuated with pharmacotherapy. Future work should also focus on combination therapy, given the proven effectiveness of this approach in the HF population.

Given the interest in preventive pharmacotherapeutic approaches, other groups have attempted various interventions in the setting of other chemotherapy regimens. In 114 cancer patients receiving various high-dose chemotherapy regimens, if troponin elevations were observed patients were randomized in an open-label fashion to placebo or ACEI (enalapril). Patients on the treatment arm maintained LVEF over the study observation period, compared to declines observed in the non-treatment group(32). Kalay *et al* randomized a heterogeneous group of cancer patients (n = 50) initiating anthracycline-based therapy to carvedilol or control, again observing a protective effect on LVEF of ACEI versus no therapy(23). The current findings extend this work, confirming that standard

HF pharmacotherapy such as ACEI may be efficacious in preventing trastuzumab-related cardiomyopathy.

No significant changes were observed in LV mass across or within the groups, which is in contrast with the findings of our study examining the effects of a four-month exercise intervention in serial unselected patients receiving trastuzumab-based therapy(12). Again, further study of larger groups is warranted to better understand the influence of pharmacotherapy on potentially sensitive imaging indices.

3.4.3 Strengths and limitations

The MANTICORE study will provide the first high-level evidence given the randomized, double-blind design, compared to the existing literature of open-label studies. Additional strength lies in cardiac MRI as the imaging modality of choice, allowing for accurate and reproducible metrics. Limitations of this sub-analysis include the interim analysis approach, with a small sample size available. Further, to allow ongoing blinding of the study team, the treatment arms were combined to create one treatment group, twice as large as the placebo group. As a result, the therapeutic effect of one treatment arm may have been potentially diluted by the other; furthermore, this approach may have inadvertently created group imbalance hampering analysis.

A major limitation of this sub-analysis of a large study actively recruiting patients is limited access to potentially important data points. Specifically, data regarding cumulative chemotherapy dosing in patients receiving sequential therapy including anthracyclines (administered prior to randomization to the

MANTICORE study) were not available at the time of this analysis. Given what is known of the potential cardiotoxic effects of anthracyclines and trastuzumab, these data may have influenced our findings. With future analysis of the full MANTICORE dataset, we expect to clarify this interaction.

3.4.4 Conclusions

In summary, we found that that standard heart failure pharmacotherapy appeared to be effective in attenuating LV dysfunction associated with trastuzumab-based therapy. When compared to patients randomized to the pharmacotherapy group, significant decline in LVEF occurred in the placebo group secondary to LV cavity dilation.

Table 3.1: Subject characteristics

	pharmacotherapy	placebo
Number	20	11
Age (years)	50 ± 8.8	48 ± 6.1
Height (cm)	162.1 ± 6.7	165.3 ± 9.2
Weight (kg)	72.0 ± 17.5	73.5 ± 14.4
Cumulative trastuzumab dose (mg)	1381 ± 107	1306 ± 139
Cancer surgery		
- Surgery performed	15	9
- Segmental resection	3	6
- Mastectomy	12	3
Stage		
- T1	7	4
- T2	7	5
- T3	1	0
- N0	7	8
- N1	5	0
- N2	2	1
Smoking status		
- Never	13	6
- Past smoker	5	3
- Current smoker	2	2
Alcohol use		
- None	0	2
- Occasional	15	7
- 1 – 2 beverages/day	1	1
- > 2 beverages/day	0	1
- History of alcohol abuse	0	1
Family history premature heart disease	0	1
Comorbid diseases		
- Chronic renal failure	0	0
- Diabetes type 1	1	0
- Diabetes type 2	1	0
- Dyslipidemia	0	0
- Hypertension	1	1

Table 3.2: LV ventricular morphology and function in breast cancer patients receiving trastuzumab-based therapy: placebo versus pharmacotherapy groups

	Placebo N = 11					Pharmacotherapy N = 20			
	Baseline	3 months	Delta	p-value		Baseline	3 months	Delta	p-value
SBP (mmHg)	121 ± 3	122 ± 3	1 ± 4.2	0.9		121 ± 3	116 ± 2	-5 ± 3.6	0.05
DBP (mmHg)	78 ± 3	74 ± 3	-4 ± 4.2	0.3		74 ± 2	70 ± 2	-4 ± 2.8	0.08
HR (bpm)	74 ± 3	76 ± 3	2 ± 4.2	0.7		70 ± 2	73 ± 2	3 ± 2.8	0.13
LVEF (%)	61.6 ± 1.4	53.6 ± 1.4	-8.0 ± 2.0	<0.001		62.1 ± 1.1	58.7 ± 1.1	-3.4 ± 1.6	0.002
LVEDV (ml)	135.8 ± 8.8	140.3 ± 8.8	4.5 ± 12.4	0.5		112.0 ± 6.7	126.4 ± 6.6	14.4 ± 9.4	0.006
LVESV (ml)	52.3 ± 4.1	65.4 ± 4.1	13.1 ± 5.8	<0.001		42.7 ± 3.1	52.0 ± 3.0	9.3 ± 4.3	<0.001
LV Mass (g)	94.8 ± 5.6	97.9 ± 5.6	3.1 ± 7.9	0.6		90.6 ± 4.2	93.9 ± 4.2	3.3 ± 5.9	0.45

Data are within-group comparisons. Values are mean and standard error.
Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; HR, heart rate; LVEF, left ventricular ejection fraction; LVEDV, left ventricular end-diastolic volume; LVESV, left ventricular end-systolic volume; LV mass, left ventricular mass.

Table 3.3: LV morphology and function in breast cancer patients receiving trastuzumab-based therapy: placebo versus pharmacotherapy groups

	Baseline					Three months			
	N = 20	N = 11				N = 20	N = 11		
	Pharmaco therapy	Placebo	Delta	p-value		Pharmaco therapy	Placebo	Delta	p-value
SBP (mmHg)	121 ± 3	121 ± 3	0 ± 4.2	0.961		116 ± 2	122 ± 3	6 ± 2.8	0.164
DBP (mmHg)	74 ± 2	78 ± 3	4 ± 2.8	0.317		70 ± 2	74 ± 3	4 ± 2.8	0.195
HR (bpm)	70 ± 2	74 ± 3	4 ± 2.8	0.239		73 ± 2	76 ± 3	3 ± 2.8	0.506
LVEF (%)	62.1 ± 1.1	61.6 ± 1.4	-0.5 ± 1.7	0.778		58.7 ± 1.1	53.6 ± 1.4	-5.1 ± 1.7	0.004
LVEDV (ml)	112.0 ± 6.7	135.8 ± 8.8	23.8 ± 11.1	0.031		126.4 ± 6.6	140.3 ± 8.8	13.9 ± 11.0	0.206
LVESV (ml)	42.7 ± 3.1	52.3 ± 4.1	9.6 ± 5.1	0.061		52 ± 3.0	65.4 ± 4.1	13.4 ± 5.1	0.009
LV Mass (g)	90.6 ± 4.2	94.8 ± 5.6	4.2 ± 7.0	0.549		93.9 ± 4.2	97.9 ± 5.6	4.0 ± 7.0	0.571

Data are between-group comparisons. Values are mean and standard error.
Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; HR, heart rate; LVEF, left ventricular ejection fraction; LVEDV, left ventricular end-diastolic volume; LVESV, left ventricular end-systolic volume; LV mass, left ventricular mass.

Figure 3.1: Study Flow Diagram

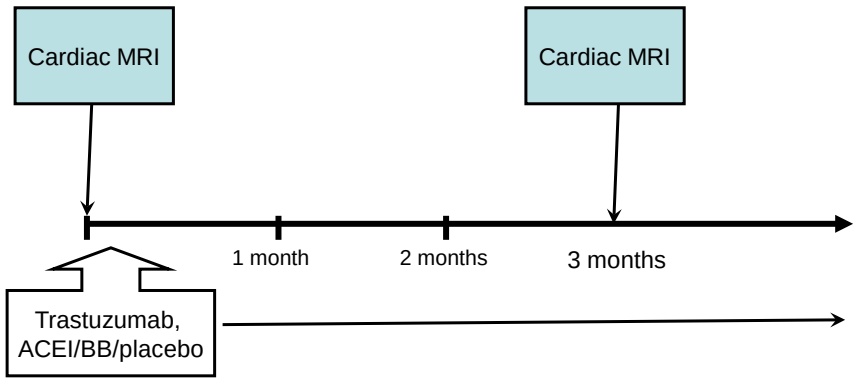


Figure 3.2: MANTICORE Recruitment (September 23, 2010 – October 27 2011)

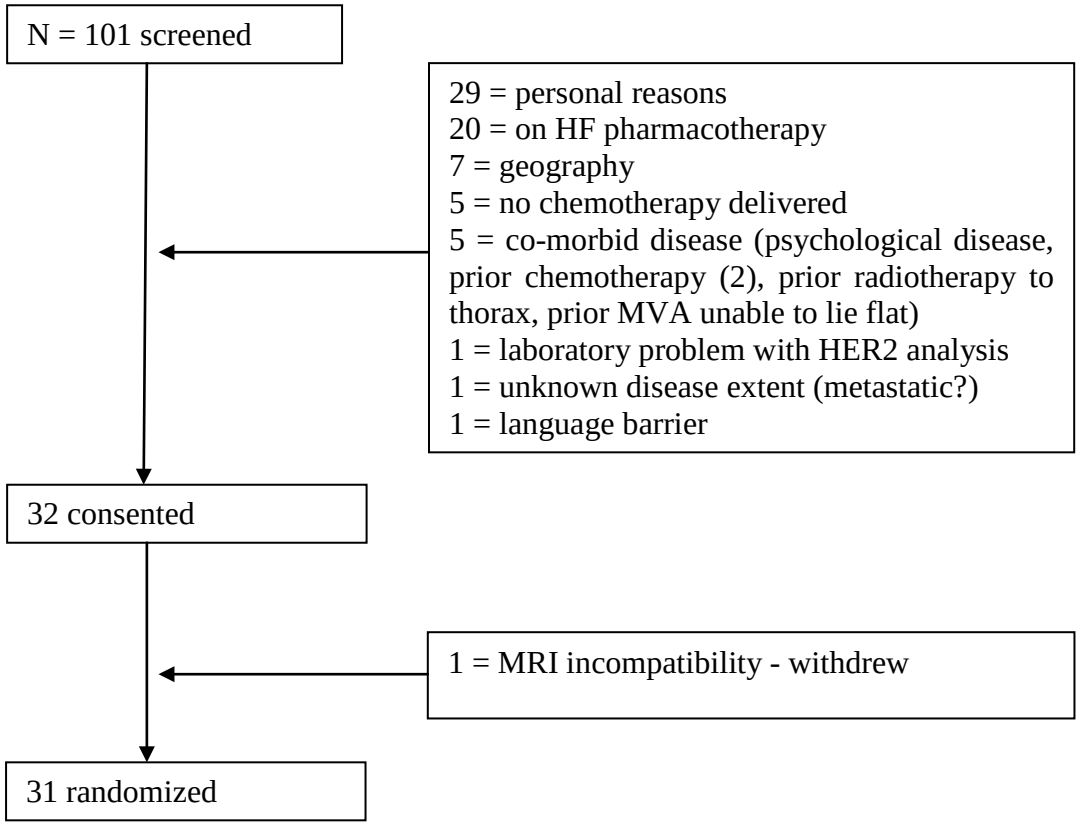


Figure 3.3: Effect of 3 months of pharmacotherapy on LVEF

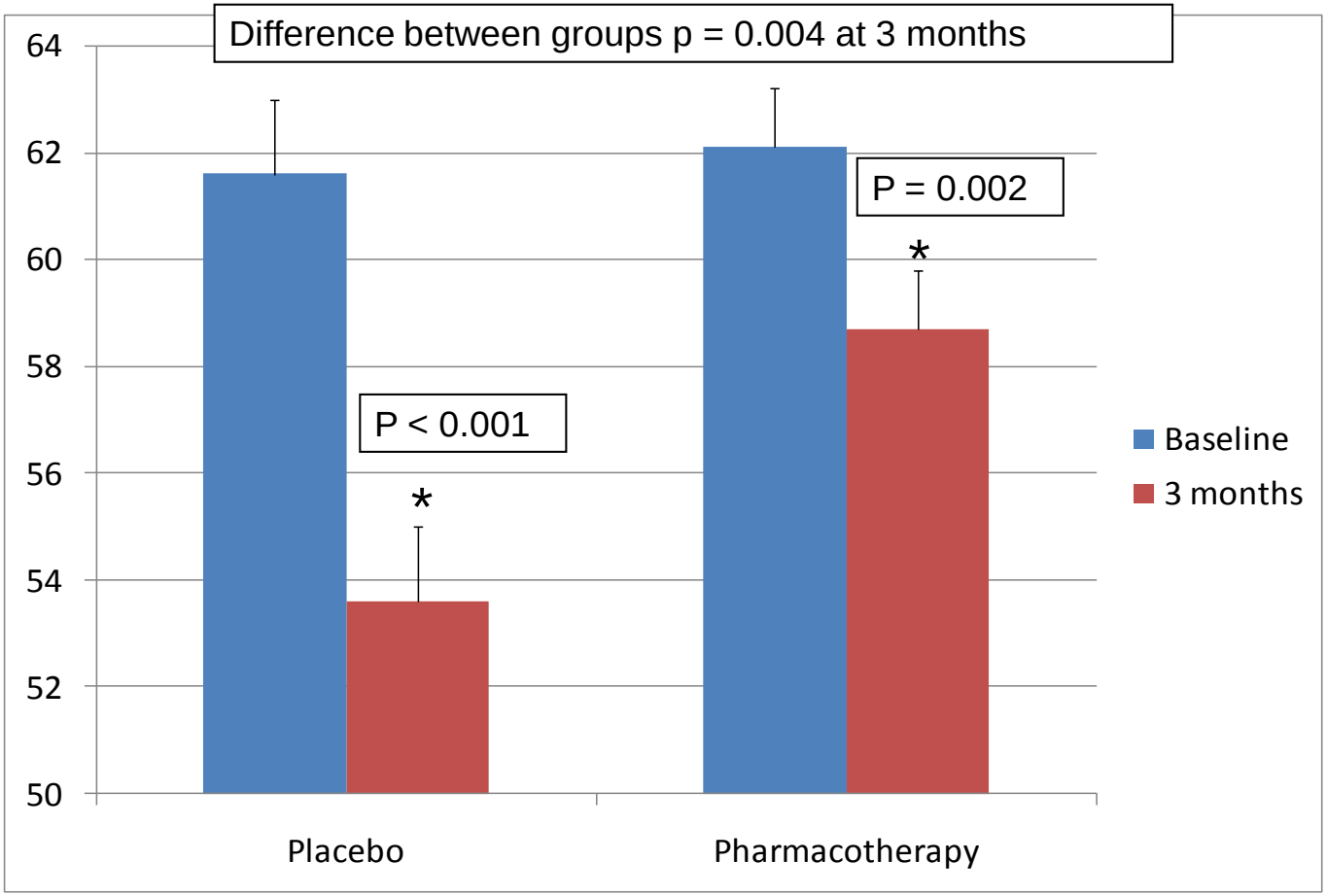


Figure 3.4: Effect of 3 months of pharmacotherapy on LVESV

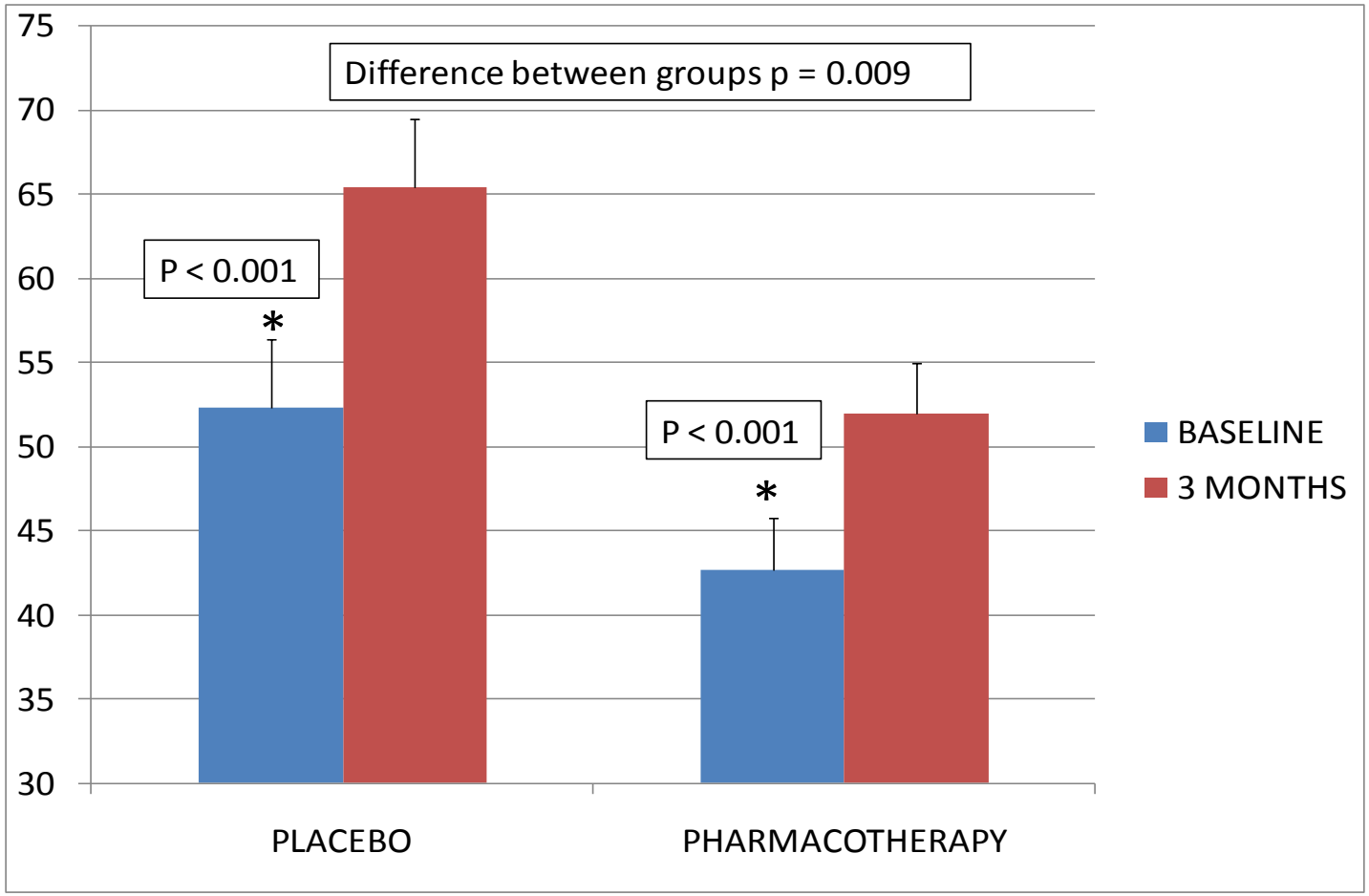
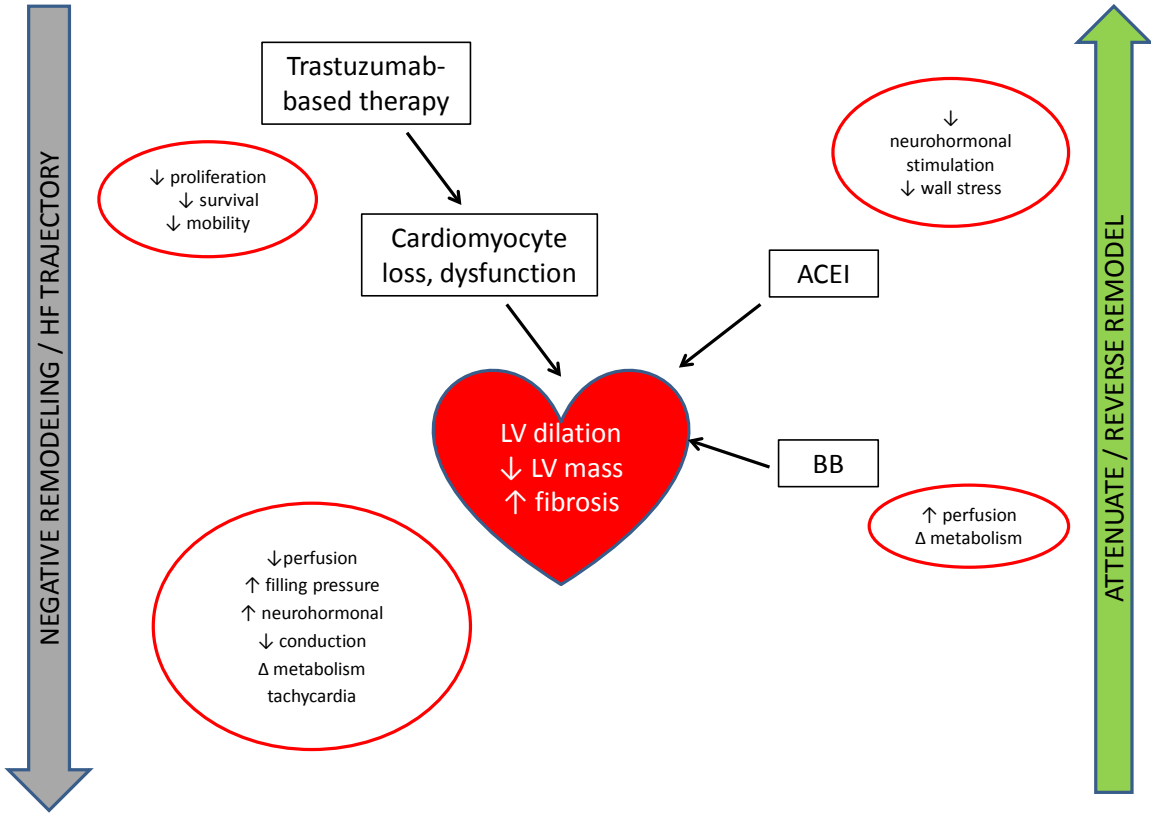


Figure 3.5: Potential cardiac remodeling effects of trastuzumab and pharmacotherapy



3.5 References

1. Romond EH, Perez EA, Bryant J, Suman VJ, Geyer CE, Jr., Davidson NE, et al. Trastuzumab plus adjuvant chemotherapy for operable HER2-positive breast cancer. N Engl J Med. 2005 Oct 20;353(16):1673-84. PubMed PMID: 16236738. Epub 2005/10/21. eng.

2. Slamon D, Eiermann W, Robert N, Pienkowski T, Martin M, Press M, et al. Adjuvant trastuzumab in HER2-positive breast cancer. N Engl J Med. 2011 Oct 6;365(14):1273-83. PubMed PMID: 21991949. Pubmed Central PMCID: 3268553.

3. Canadian Cancer Statistics. Toronto, Canada2010.

4. Bowles EJ, Wellman R, Feigelson HS, Onitilo AA, Freedman AN, Delate T, et al. Risk of heart failure in breast cancer patients after anthracycline and trastuzumab treatment: a retrospective cohort study. J Natl Cancer Inst. 2012 Sep 5;104(17):1293-305. PubMed PMID: 22949432. Pubmed Central PMCID: 3433392.

5. Bellenger NG, Burgess MI, Ray SG, Lahiri A, Coats AJ, Cleland JG, et al. Comparison of left ventricular ejection fraction and volumes in heart failure by echocardiography, radionuclide ventriculography and cardiovascular magnetic resonance; are they interchangeable? Eur Heart J. 2000 Aug;21(16):1387-96. PubMed PMID: 10952828. Epub 2000/08/23. eng.

6. Bellenger N, Pennell D. Magnetic resonance imaging in cardiology. J R Coll Physicians Lond. 1999 Jan-Feb;33(1):12-8. PubMed PMID: 10192063. Epub 1999/04/07. eng.

7. Bellenger NG, Davies LC, Francis JM, Coats AJ, Pennell DJ. Reduction in sample size for studies of remodeling in heart failure by the use of cardiovascular magnetic resonance. Journal of cardiovascular magnetic resonance : official journal of the Society for Cardiovascular Magnetic Resonance. 2000;2(4):271-8. PubMed PMID: 11545126. Epub 2001/09/08. eng.

8. Wolfe RR. Skeletal muscle protein metabolism and resistance exercise. J Nutr. 2006 Feb;136(2):525S-8S. PubMed PMID: 16424140. Epub 2006/01/21. eng.

9. Cohn JN, Ferrari R, Sharpe N. Cardiac remodeling--concepts and clinical implications: a consensus paper from an international forum on cardiac remodeling. Behalf of an International Forum on Cardiac Remodeling. J Am Coll Cardiol. 2000 Mar 1;35(3):569-82. PubMed PMID: 10716457. Epub 2000/03/15. eng.

10. Mann DL, Bristow MR. Mechanisms and Models in Heart Failure: The Biomechanical Model and Beyond. Circulation. 2005 May 31, 2005;111(21):2837-49.

11. Vasan RS, Larson MG, Benjamin EJ, Evans JC, Levy D. Left ventricular dilatation and the risk of congestive heart failure in people without myocardial infarction. N Engl J Med. 1997 May 8;336(19):1350-5. PubMed PMID: 9134875. Epub 1997/05/08. eng.

12. Haykowsky MJ, Mackey JR, Thompson RB, Jones LW, Paterson DI. Adjuvant trastuzumab induces ventricular remodeling despite aerobic exercise training. Clin Cancer Res. 2009 Aug 1;15(15):4963-7. PubMed PMID: 19622583.

13. Wadhwa D, Fallah-Rad N, Grenier D, Krahn M, Fang T, Ahmadie R, et al. Trastuzumab mediated cardiotoxicity in the setting of adjuvant chemotherapy for breast cancer: a retrospective study. Breast Cancer Res Treat. 2009 Sep;117(2):357-64. PubMed PMID: 19082707.

14. Fallah-Rad N, Walker JR, Wassef A, Lytwyn M, Bohonis S, Fang T, et al. The utility of cardiac biomarkers, tissue velocity and strain imaging, and cardiac magnetic resonance imaging in predicting early left ventricular dysfunction in patients with human epidermal growth factor receptor II-positive breast cancer treated with adjuvant trastuzumab therapy. J Am Coll Cardiol. 2011 May 31;57(22):2263-70. PubMed PMID: 21616287.

15. Hunt SA, Abraham WT, Chin MH, Feldman AM, Francis GS, Ganiats TG, et al. ACC/AHA 2005 Guideline Update for the Diagnosis and Management of Chronic Heart Failure in the Adult: A Report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Writing Committee to Update the 2001 Guidelines for the Evaluation and Management of Heart Failure): Developed in Collaboration With the American College of Chest Physicians and the International Society for Heart and Lung Transplantation: Endorsed by the Heart Rhythm Society. Circulation. 2005 September 20, 2005;112(12):e154-235.

16. Malcom J, Arnold O, Howlett JG, Ducharme A, Ezekowitz JA, Gardner MJ, et al. Canadian Cardiovascular Society Consensus Conference guidelines on heart failure--2008 update: best practices for the transition of care of heart failure patients, and the recognition, investigation and treatment of cardiomyopathies. Can J Cardiol. 2008 Jan;24(1):21-40. PubMed PMID: 18209766. Epub 2008/01/23. eng.

17. Kramer DG, Trikalinos TA, Kent DM, Antonopoulos GV, Konstam MA, Udelson JE. Quantitative evaluation of drug or device effects on ventricular remodeling as predictors of therapeutic effects on mortality in patients with heart failure and reduced ejection fraction: a meta-analytic approach. J Am Coll Cardiol. 2010 Jul 27;56(5):392-406. PubMed PMID: 20650361.

18. Katzung BG. Basic and Clinical Pharmacology. 11 ed: McGraw-Hill; 2009.

19. Mackey JR, Clemons M, Cote MA, Delgado D, Dent S, Paterson A, et al. Cardiac management during adjuvant trastuzumab therapy: recommendations of the Canadian Trastuzumab Working Group. Curr Oncol. 2008 Feb;15(1):24-35. PubMed PMID: 18317582. Epub 2008/03/05. eng.

20. Kalay N, Basar E, Ozdogru I, Er O, Cetinkaya Y, Dogan A, et al. Protective Effects of Carvedilol Against Anthracycline-Induced Cardiomyopathy. Journal of the American College of Cardiology. 2006;48(11):2258-62.

21. Cardinale D, Lamantia G, Cipolla CM. Troponin I and cardiovascular risk stratification in patients with testicular cancer. J Clin Oncol. 2006 Jul

20;24(21):3508; author reply -9. PubMed PMID: 16849772. Epub 2006/07/20. eng.

22. Lipshultz SE, Lipsitz SR, Sallan SE, Simbre VC, 2nd, Shaikh SL, Mone SM, et al. Long-term enalapril therapy for left ventricular dysfunction in doxorubicin-treated survivors of childhood cancer. J Clin Oncol. 2002 Dec 1;20(23):4517-22. PubMed PMID: 12454107. Epub 2002/11/28. eng.

23. Kalay N, Basar E, Ozdogru I, Er O, Cetinkaya Y, Dogan A, et al. Protective effects of carvedilol against anthracycline-induced cardiomyopathy. J Am Coll Cardiol. 2006 Dec 5;48(11):2258-62. PubMed PMID: 17161256. Epub 2006/12/13. eng.

24. Cardinale D, Sandri MT, Martinoni A, Tricca A, Civelli M, Lamantia G, et al. Left ventricular dysfunction predicted by early troponin I release after high-dose chemotherapy. J Am Coll Cardiol. 2000 Aug;36(2):517-22. PubMed PMID: 10933366. Epub 2000/08/10. eng.

25. Cardinale D, Colombo A, Lamantia G, Colombo N, Civelli M, De Giacomi G, et al. Anthracycline-induced cardiomyopathy: clinical relevance and response to pharmacologic therapy. J Am Coll Cardiol. Jan 19;55(3):213-20. PubMed PMID: 20117401. Epub 2010/02/02. eng.

26. Crone SA, Zhao YY, Fan L, Gu Y, Minamisawa S, Liu Y, et al. ErbB2 is essential in the prevention of dilated cardiomyopathy. Nat Med. 2002 May;8(5):459-65. PubMed PMID: 11984589. Epub 2002/05/02. eng.

27. Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, Bajamonde A, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic

breast cancer that overexpresses HER2. N Engl J Med. 2001 Mar 15;344(11):783-92. PubMed PMID: 11248153. Epub 2001/03/15. eng.

28. Hudis CA. Trastuzumab--mechanism of action and use in clinical practice. N Engl J Med. 2007 Jul 5;357(1):39-51. PubMed PMID: 17611206.

29. Chien KR. Myocyte survival pathways and cardiomyopathy: implications for trastuzumab cardiotoxicity. Seminars in oncology. 2000 Dec;27(6 Suppl 11):9-14; discussion 92-100. PubMed PMID: 11236034.

30. De Keulenaer GW, Brutsaert DL. Systolic and Diastolic Heart Failure Are Overlapping Phenotypes Within the Heart Failure Spectrum / Response to De Keulenaer and Brutsaert. Circulation. May 10, 2011;123(18):1996-2005.

31. Frigerio M, Roubina E. Drugs for left ventricular remodeling in heart failure. Am J Cardiol. 2005 Dec 19;96(12A):10L-8L. PubMed PMID: 16399088.

32. Cardinale D, Colombo A, Sandri MT, Lamantia G, Colombo N, Civelli M, et al. Prevention of high-dose chemotherapy-induced cardiotoxicity in high-risk patients by angiotensin-converting enzyme inhibition. Circulation. 2006 Dec 5;114(23):2474-81. PubMed PMID: 17101852. Epub 2006/11/15. eng.

Chapter 4

Summary of work completed and future directions

4.1 Summary

Given the successes of anti-cancer therapies, clinicians are faced with a rapidly increasing population of cancer survivors. North American statistics project that in the next decade, the number of people surviving 5 years or more after their cancer diagnosis to increase to approximately 37%, to 11.9 million(1). As an example, five-year survival rates approach 88% for early stage breast cancer(2); however, treatment-related sequelae are sufficiently significant that cardiovascular disease has replaced cancer as the predominant cause of mortality(3).

A major advancement in the treatment of breast cancer is represented by trastuzumab, a humanized monoclonal antibody was specifically developed to target HER2. Meta-analysis of EBC patients receiving trastuzumab-based chemotherapy regimens shows significant reductions in mortality, disease recurrence, rates of metastases and secondary tumors when compared to non-trastuzumab regimens(4). However, a fivefold increase in the relative risk of HF and near doubling risk of LVEF decline was observed. Accordingly, the potentially devastating short and long-term effects of this and other novel anti-cancer therapies warrant careful study. The overarching theme of this thesis was to examine the short and long-term effects of trastuzumab on LV morphology and function in breast cancer patients.

Two studies were undertaken; the first a cross-sectional study of 16 female breast cancer survivors exposed to trastuzumab-based chemotherapy, examining LV remodeling and exercise capacity four years after completion of treatment. We found that early breast cancer patients had significant alterations in left ventricular geometry and impairment of cardiorespiratory function four years following exposure to trastuzumab-based chemotherapy. We observed acute LV dilatation at 4 months, reflected by increases in LV volumes, followed by statistically significant decrease of LV volumes after 4-years of cessation of trastuzumab therapy. Changes in LV mass also occurred, with a 5g increase from baseline at 4 months, followed by a statistically significant 14g decrease at 4 years. LVEF returned to baseline values after 4 years. We also observed that adherence to exercise prescription during the four-month intervention was associated with higher VO_{2peak} at 4 years, with a clinically relevant 4.1 mL/kg/min difference between patients who attended at least 80% of sessions versus those who did not.

The second study was performed to examine the effects of three months of standard heart failure pharmacotherapy, consisting of angiotensin-converting enzyme inhibitor or beta-blocker versus placebo, in preventing LV remodeling in EBC patients receiving trastuzumab-based therapy. The major finding was that that pharmacotherapy appeared to be effective in attenuating early LV dysfunction, for when compared to patients randomized to the pharmacotherapy group, significant decline in LVEF occurred in the placebo group.

4.2 Future directions

Taken together, the findings of these studies highlight the potential contributions of cardiovascular risk reduction and supportive care measures in reducing short and long term cardiovascular sequelae of cancer therapy. Overweight/obesity, low fruit and vegetable intake coupled with high fat diet, physical inactivity, smoking and alcohol use are firmly established cancer-promoting, modifiable behaviors(5). Furthermore, disability associated with cancer therapy is exacerbated in survivors in the presence of these risk factors, termed the 'multiple-hit' effect (6). One potential approach is multidisciplinary team intervention, as provision of such care has been shown to be effective in obesity management(7) as well as reducing mortality and hospitalizations among HF patients(8-10). Accordingly, we are undertaking a randomized controlled trial of intensive multidisciplinary care in early breast cancer and lymphoma patients (Figure 4.1), delivered by a team of exercise physiologists, pharmacists, dieticians, nurses and physicians with the aim of improving health and avoiding cardiac dysfunction(11). We expect our results to inform comprehensive, holistic care for patients at risk for negative cancer therapy mediated sequelae.

Given the cardiac MRI observations in Study One, the potential contribution of aging among cancer survivors also warrants consideration. Aging is associated with progressive fibrosis occurring in multiple organ systems(12) which, even in healthy older individuals, is associated with gradual structural and functional alterations. Typically, increased LV mass is observed with age, attributed to fibrotic remodeling and response to peripheral vascular stiffening and increased hemodynamic load(13). However, this increase in mass is generally

accompanied by senesence of functional cardiomyocytes. Potential interactions may influence the evolution of LV mass, and eventually LV function in cancer survivors, such as pre-existing risk factors at the time of diagnosis, the type of anti-cancer therapy received, gender and lifestyle before, during and after therapy. Large scale studies of these factors over the long-term cancer trajectory are necessary to elucidate these interactions.

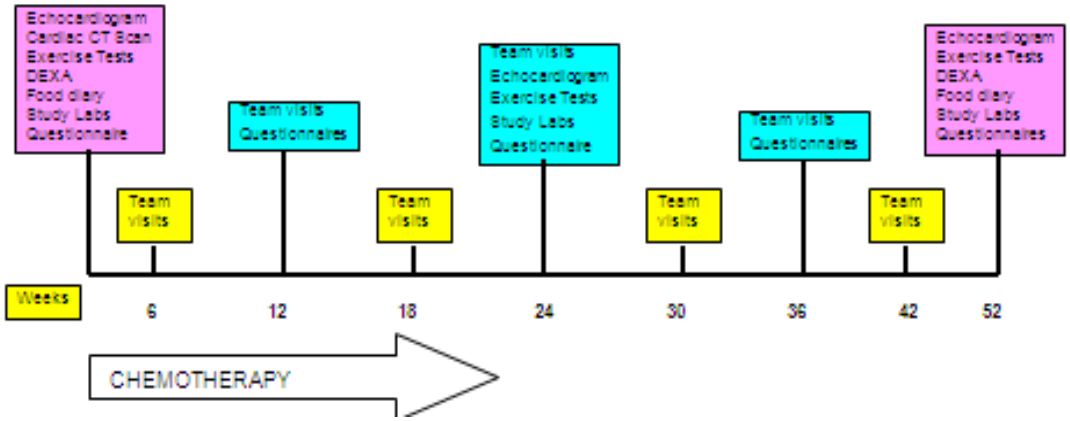
As evidence accumulates, a risk 'algorithm' for a specific anti-cancer therapy could potentially be developed, including established cardiovascular risk factors and emerging risks, genetic influences, biomarkers or clinical investigations. The addition of cardiopulmonary exercise testing to standard imaging and clinical assessments may be informative, given the findings of our meta-analysis(14). Furthermore, given limited healthcare resources, this algorithm would potentially identify those high-risk patients requiring additional monitoring and interventions versus those at low risk, minimize unnecessary or painful/invasive tests, and offer a 'personalized medicine' approach to treatment and survivorship.

The findings of this thesis suggest that additional questions remain regarding the direct and indirect effects of trastuzumab (and other cancer therapies) on not only the heart, but other body compartments. A bench to bedside to bench approach is required to better understand the negative alterations observed, and approaches to offset these. Questions also remain regarding the role of cardiopulmonary exercise, in terms of the optimal timing, optimal training and potentially augmentation of exercise with pharmacotherapy or other experimental

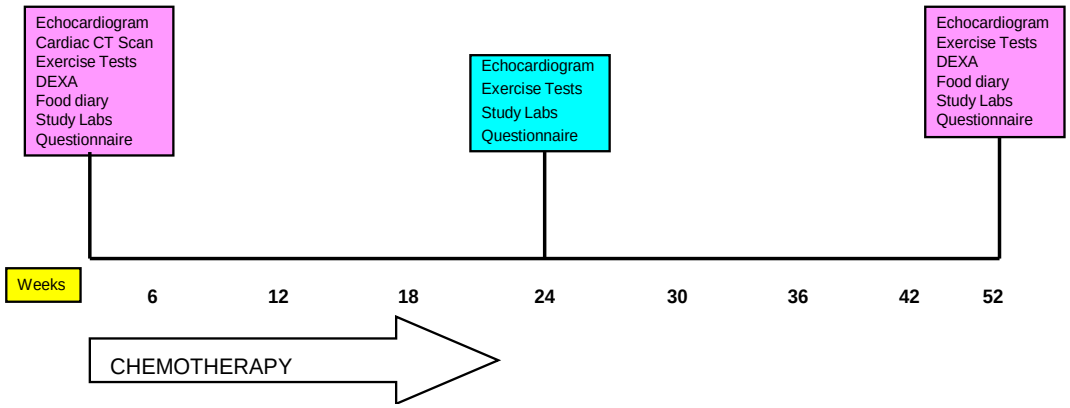
agents(15). Here too, basic science efforts are required to elucidate the molecular pathways influenced negatively or positively by the various interventions. In cancer patient populations with impaired exercise tolerance, different training approaches may be more tolerable i.e. high-intensity intervals; well-designed trials are currently underway(16). Randomized controlled trials of combination therapy, either combined pharmacotherapy and/or combined exercise intervention are warranted.

Figure 4.1 – Schematic of TITAN study randomization arms

A. Multidisciplinary care arm



B. Usual care arm



4.5 References

1. de Moor JS, Mariotto AB, Parry C, Alfano CM, Padgett L, Kent EE, et al. Cancer Survivors in the United States: Prevalence across the Survivorship Trajectory and Implications for Care. Cancer Epidemiology Biomarkers & Prevention. 2013 April 1, 2013;22(4):561-70.

2. Canadian Cancer Statistics. Toronto, Canada2010.

3. Patnaik JL, Byers T, DiGuiseppi C, Dabelea D, Denberg TD. Cardiovascular disease competes with breast cancer as the leading cause of death for older females diagnosed with breast cancer: a retrospective cohort study. Breast cancer research : BCR. 2011;13(3):R64. PubMed PMID: 21689398. Pubmed Central PMCID: 3218953.

4. Viani GA, Afonso SL, Stefano EJ, De Fendi LI, Soares FV. Adjuvant trastuzumab in the treatment of her-2-positive early breast cancer: a meta-analysis of published randomized trials. BMC Cancer. 2007;7:153. PubMed PMID: 17686164. Epub 2007/08/10. eng.

5. Anand SS, Yusuf S. Stemming the global tsunami of cardiovascular disease. The Lancet.377(9765):529-32. PubMed PMID: S0140-6736(10)62346-X.

6. Jones LW, Haykowsky MJ, Swartz JJ, Douglas PS, Mackey JR. Early breast cancer therapy and cardiovascular injury. J Am Coll Cardiol. 2007 Oct 9;50(15):1435-41. PubMed PMID: 17919562. Epub 2007/10/09. eng.

7. Wharton S, Vanderlelie S, Sharma AM, Sharma S, Kuk JL. Feasibility of an interdisciplinary program for obesity management in Canada. Can Fam Physician. Jan;58(1):e32-8. PubMed PMID: 22267637. Epub 2012/01/24. eng.

8. McAlister FA, Stewart S, Ferrua S, McMurray JJ. Multidisciplinary strategies for the management of heart failure patients at high risk for admission: a systematic review of randomized trials. J Am Coll Cardiol. 2004 Aug 18;44(4):810-9. PubMed PMID: 15312864. Epub 2004/08/18. eng.

9. Holland R, Battersby J, Harvey I, Lenaghan E, Smith J, Hay L. Systematic review of multidisciplinary interventions in heart failure. Heart. 2005 July 1, 2005;91(7):899-906.

10. Inglis SC, Pearson S, Treen S, Gallasch T, Horowitz JD, Stewart S. Extending the Horizon in Chronic Heart Failure: Effects of Multidisciplinary, Home-Based Intervention Relative to Usual Care. Circulation. 2006 December 5, 2006;114(23):2466-73.

11. Clinicaltrials.gov. Multidisciplinary Team IntervenTion in CArdio-ONcology (TITAN Study) 2013 [cited 2013]. Available from: http://clinicaltrials.gov/show/NCT01621659.

12. Biernacka A, Frangogiannis NG. Aging and Cardiac Fibrosis. Aging Dis. Apr;2(2):158-73. PubMed PMID: 21837283. Epub 2011/08/13. Eng.

13. North BJ, Sinclair DA. The intersection between aging and cardiovascular disease. Circ Res. Apr 13;110(8):1097-108. PubMed PMID: 22499900. Epub 2012/04/14. eng.

14. Jones LW, Liang Y, Pituskin EN, Battaglini CL, Scott JM, Hornsby WE, et al. Effect of exercise training on peak oxygen consumption in patients with cancer: a meta-analysis. Oncologist. 2011;16(1):112-20. PubMed PMID: 21212429. Pubmed Central PMCID: 3228052.

15. Dolinsky VW, Jones KE, Sidhu RS, Haykowsky M, Czubryt MP, Gordon T, et al. Improvements in skeletal muscle strength and cardiac function induced by resveratrol during exercise training contribute to enhanced exercise performance in rats. The Journal of physiology. 2012 Jun 1;590(Pt 11):2783-99. PubMed PMID: 22473781. Pubmed Central PMCID: 3424731.

16. Jones L, Douglas P, Eves N, Marcom PK, Kraus W, Herndon J, et al. Rationale and design of the Exercise Intensity Trial (EXCITE): A randomized trial comparing the effects of moderate versus moderate to high-intensity aerobic training in women with operable breast cancer. BMC Cancer.10(1):531. PubMed PMID: doi:10.1186/1471-2407-10-531.

APPENDIX A: Ethics Certificates of Approval and Consent Forms

Health Research Ethics Board

308 Campus Tower
University of Alberta, Edmonton, AB T6G 1K8
p. 780.492.9724 (Biomedical Panel)
p. 780.492.0302 (Health Panel)
p. 780.492.0459
p. 780.492.0839
f. 780.492.9429

Approval Form

Date:	September 6, 2011	
Principal Investigator:	Mark Haykowsky	
Study ID:	Pro00024632	
Study Title:	**LONG TERM EFFECTS OF HERCEPTIN ON HEART FUNCTION AND EXERCISE CAPACITY IN WOMEN WITH HER2 POSITIVE BREAST CANCER**	
Approval Expiry Date:	September 4, 2012	
Date of Informed Consent:	Approval Date 8/31/2011	Approved Document HEREX Extension Consent

Thank you for submitting the above study to the Health Research Ethics Board - Biomedical Panel. Your application has been reviewed and approved on behalf of the committee. The Protocol (undated) and Informed Consent Form, dated 17 Jul 2011, form part of this approval.

The Health Research Ethics Board assessed all matters required by section 50(1)(a) of the Health Information Act. Subject consent for access to identifiable health information is required for the research described in the ethics application, and appropriate procedures for such consent have been approved by the HREB - Biomedical Panel. In order to comply with the Health Information Act, a copy of the approval form is being sent to the Office of the Information and Privacy Commissioner.

A renewal report must be submitted next year prior to the expiry of this approval if your study still requires ethics approval. If you do not renew on or before the renewal expiry date (September 4, 2012), you will have to re-submit an ethics application.

The membership of the Health Research Ethics Board - Biomedical Panel complies with the membership requirements for research ethics boards as defined in Division 5 of the Food and Drug Regulations and the Tri-Council Policy Statement. The HREB - Biomedical Panel carries out its functions in a manner consistent with Good Clinical Practices.

Approval by the Health Research Ethics Board does not encompass authorization to access the patients, staff or resources of Alberta Health Services or other local health care institutions for the purposes of the research. Enquiries regarding Alberta Health administrative approval, and operational approval for areas impacted by the research, should be directed to the Alberta Health Services Research Administration office, #1800 College Plaza, phone (780) 407-6041.

Sincerely,

J. Stephen Bamforth, MD
Associate Chair, HREB Biomedical

Note: This correspondence includes an electronic signature (validation and approval via an online system).

UNIVERSITY OF ALBERTA Alberta Health Services Covenant Health

INFORMATION SHEET

LONG TERM EFFECTS OF HERCEPTIN ON HEART FUNCTION AND EXERCISE CAPACITY IN WOMEN WITH HER2 POSITIVE BREAST CANCER

INVESTIGATORS:

M. Haykowsky, PhD	U of A, Dept. of Physical Therapy	492-5970
E. Pituskin RN PhD(c)	U of A, Dept. of Physical Therapy	432-8518
J. Mackey, MD, FRCPC	Dept. of Medical Oncology, Cross Cancer Institute	432-8221
I. Paterson, MD, FRCPC	U of A, Division of Cardiology	407-1857
R. Thompson, PhD	U of A, Dept. of Biomedical Engineering	492-8665

BACKGROUND: You received a medication called Herceptin as part of the treatment plan for your breast cancer, and took part in a study examining the effect of aerobic exercise training in the short term. Currently, the long term effects of Herceptin therapy on the heart muscle and exercise capacity are unknown.

PURPOSE: You are being asked to participate in a study that will examine the long term effect of Herceptin therapy on heart muscle function and exercise capacity. We will ask the same women who participated in our original study if they would like to take part in this study.

DESCRIPTION OF THE STUDY: If you decide to take part, the total time commitment will be approximately one-half day. We will ask you to undergo the same tests you did in the original study. Participation in this study will not affect/influence any treatments you may still be receiving at the Cross Cancer Institute.

RESEARCH PROCEDURES

Test 1. Blood Sample at a Laboratory near your home. Duration: ½ hour.

We would like to check your blood cell count. This is a routine blood test that may be collected at a laboratory near your home. The study team will help organize this for you.

Test 2: Physical examination, symptom report and health history. Duration: ½ hour.

We will measure your weight, blood pressure, heart rate and breathing rate. We will ask you to complete a symptom assessment form. We will also ask you how your health has been overall, and any medications you have been taking since you took part in the earlier study.

Test 3. Exercise Laboratory, Division of Cardiology. Duration: 1 hour.

Exercise test on a bicycle. Prior to this test, ECG leads (electrical contacts) will be placed on your chest to measure your heart rate. A blood pressure cuff will be placed on your arm to

measure your blood pressure. You will start the test with easy pedalling that will become a little more difficult every two minutes. A special mouthpiece and nose clip will be used to measure your oxygen uptake. Throughout the test, your heart rate and blood pressure will be measured. The exercise test usually lasts 8 to 10 minutes and a specially trained health care worker will supervise the test.

Test 4. University of Alberta *In Vivo* NMR Center. Duration: 1 hour.

Cardiac Magnetic Resonance Imagining (MRI) Assessment of Heart Function

Three ECG leads will be placed on your chest to measure your heart rate and a cuff will be placed on your arm to measure your blood pressure. A special probe that measures the amount of oxygen in your body will be placed on your finger. You will then go into the MRI machine for images of your heart to be taken. The MRI examination itself is painless and involves lying on your back in the machine for about an hour. While in the MRI machine you will hear a banging or knocking sound as the computer opens and closes various switches. We will supply ear plugs and headphones to reduce the noise and allow us to speak with you while you are in the MRI machine. You will also be given a small hand-held alarm so that you can stop the procedure at any time. A physician and registered nurse will supervise this test.

POSSIBLE BENEFITS: This study will determine the long-term effect of Herceptin therapy has on heart muscle function and exercise capacity. You may not personally derive benefit by participating in this study. Some people enjoy exercising, and like to know their exercise capacity.

POSSIBLE RISKS: The exercises that you will perform are generally regarded as very safe. All testing and exercise sessions will be performed under appropriate supervision. Data from individuals with or without heart disease suggests that the likelihood of having a heart attack or dying during a bicycle test is 1 in 10,000 tests. The mouthpiece that is used during the exercise test may make your mouth feel dry. You may also experience temporary muscle soreness after the initial exercise test and during the first weeks of exercise training.

Exposure to the magnetic fields in an MRI machine has no known harmful effects. As part of usual MRI procedure, you will be asked a series of questions to make sure it is safe before you have an MRI scan.

COSTS: You will not have to pay for the consultations that you receive by participating in this study. However, you may be coming to the University of Alberta more often than if you were not participating in this study. As a result, there may be some extra costs for you such as gasoline, child care or meals. Parking will be provided for you while you participate in this study.

COMPENSATION FOR INJURY: If you become ill or injured as a result of participating in this study, necessary medical treatment will be available at no additional cost to you. By signing

this consent form you are not releasing the investigator(s), institution(s) and/or sponsor(s) from their legal and professional responsibilities.

CONTACTS: Please contact the following investigators listed below if you have any questions and concerns. M Haykowsky, PhD (492-5970), E Pituskin RN PhD(c) (432-8518) or J. Mackey, MD, FRCPC (432-8221)

CONFIDENTIALITY: Personal records will be kept confidential. Only the persons listed above will have access to your data. Any report published as a result of this study will not identify you by name.

For this study, the researchers may need to access your personal health records for health information such as past medical history and test results. The health information collected as part of this study will be kept confidential unless release is required by law, and will be used only for the purpose of the research study. By signing the consent form you give permission to the study staff to access any personally identifiable health information which is under the custody of other health care professionals as deemed necessary for the conduct of the research.

In addition to the investigators listed, the Health Research Ethics Board or the Alberta Cancer Research Ethics Board may have access to your personal health records to monitor the research and verify the accuracy of study data.

By signing the consent form you give permission for the collection, use and disclosure of your medical records. In Canada, study information is required to be kept for 5 years. Even if you withdraw from the study, the medical information which is obtained from you for study purposes will not be destroyed. You have a right to check your health records and request changes if your personal information is incorrect.

VOLUNTARY PARTICIPATION: You are free to withdraw from the research study at any time, and your continuing medical care will not be affected in any way. If the study is not undertaken or if it is discontinued at any time, the quality of your medical care will not be affected. If any knowledge gained from this or any other study becomes available which could influence your decision to continue in the study, you will be promptly informed.

CONTACT INFORMATION: If you have any questions or concerns about any aspect this study, you may contact the Health Research Ethics Board at 780 492-9724. This office has no affiliation with the study investigators.

CONSENT FORM

LONG TERM EFFECTS OF HERCEPTIN ON HEART FUNCTION AND EXERCISE CAPACITY IN WOMEN WITH HER2 POSITIVE BREAST CANCER

INVESTIGATORS:

M. Haykowsky, PhD	U of A, Dept. of Physical Therapy	492-5970
E. Pituskin RN PhD(c)	U of A, Dept. of Physical Therapy	432-8518
J. Mackey, MD, FRCPC	Dept. of Medical Oncology, Cross Cancer Institute	432-8221
I. Paterson, MD, FRCPC	U of A, Division of Cardiology	407-1857
R. Thompson, PhD	U of A, Dept. of Biomedical Engineering	492-8665

Please answer the following questions:

	Yes	No
Do you understand that you are being asked to be in a research study?	___	___
Have you read and received a copy of an attached information sheet?	___	___
Do you understand the benefits and risks in taking part in this research study?	___	___
Have you had an opportunity to ask questions and discuss this study?	___	___
Do you understand that you are free to withdraw from the study at any time without having to give a reason and without affecting your future medical care?	___	___
Has the issue of confidentiality been explained to you, and do you understand who will have access to your medical records?	___	___
Do you want the investigators to inform you family doctor that you are participating in this study?	___	___

Who explained this study to you? _____

	Yes	No
I agree to take part in this study:	___	___

_____ _____
Signature of Participant Date

_____
Printed Name of Participant

_____ _____
Signature of Witness Date

I believe that the person signing this form understands what is involved in the study and voluntarily agrees to participate.

_____ _____
Signature of Investigator or Designee Date

Department of Physical Therapy
250 Corbett Hall • University of Alberta • Edmonton • Canada • T6G 2G4
Telephone: (780) 492-5983 • Fax: (780) 492-4429

Health Research Ethics Board

308 Campus Tower
University of Alberta, Edmonton, AB T6G 1K8
p. 780.492.9724 (Biomedical Panel)
p. 780.492.0302 (Health Panel)
p. 780.492.0459
p. 780.492.0839
f. 780.492.9429

ETHICS APPROVAL FORM - DELEGATED REVIEW

Date:	March 16, 2010	
Principal Investigator:	Ian Paterson	
Study ID:	Pro00010851	
Study Title:	Multidisciplinary Approach to Novel Therapies In Cardiology Oncology REsearch	
Protocol Number:	101	
Protocol Date:	01/09/2009	
Approval Expiry Date:	March 15, 2011	
Date of Informed Consent:	Approval Date 16/03/2010	Approved Document HREB MANTICORE CONSENT IP edits Dec 18 2009.doc

Thank you for responding to all of the comments raised by the Health Research Ethics Board (Biomedical Panel) arising from the presentation of this project at the December 2009 meeting. The protocol involved in this project has been found to be acceptable within the limitations of human experimentation. There are no outstanding ethical issues and the study is approved. The protocol, dated September 1, 2009 and informed consent document dated March 16, 2010 have also been approved. We also note that Health Canada approval for this study has been obtained.

This ethics approval is valid for one year. A renewal report must be submitted next year prior to the expiry of this approval if your study still requires ethics approval. You will receive electronic reminders at 45, 30, 15 and 1 day(s) prior to the expiry date. If you do not renew on or before that date, you will have to re-submit another ethics application.

For studies where investigators must obtain informed consent, signed copies of the consent form must be retained, as should all study related documents, and be available to the HREB upon request. As you study falls under Health Canada jurisdiction, these records must be retained for 25 years in accordance with Division 5 of the Food and Drug Regulations.

Approval by the Health Research Ethics Board does not encompass authorization to access the patients, staff or resources of Alberta Health Services. Enquiries regarding AHS administrative approval, and operational approval for areas impacted by research, should be directed to the AHS Research Administration office, #1800 College Plaza (780-407-6041).

Sincerely,

S.K.M. Kimber, MD, FRCPC
Chair, Health Research Ethics Board (Biomedical Panel)

Note: This correspondence includes an electronic signature (validation and approval via an online system).

Health Research Ethics Board

308 Campus Tower
University of Alberta, Edmonton, AB T6G 1K8
p. 780.492.9724 (Biomedical Panel)
p. 780.492.0302 (Health Panel)
p. 780.492.0459
p. 780.492.0839
f. 780.492.9429

Re-Approval Form

Date:	February 23, 2012
Principal Investigator:	Ian Paterson
Study ID:	Pro00010851
Study Title:	Multidisciplinary Approach to Novel Therapies In Cardiology Oncology REsearch
Approval Expiry Date:	March 14, 2013
Protocol Number:	101
Sponsor/Funding Agency:	Alberta Cancer Foundation CIHR - Canadian Institutes for Health Research

The Health Research Ethics Board - Biomedical Panel has reviewed the renewal request and file for this project and found it to be acceptable within the limitations of human experimentation.

The re-approval for the study as presented is valid for another year. It may be extended following completion of the annual renewal request. Beginning 45 days prior to expiration, you will receive notices that the study is about to expire. Once the study has expired you will have to resubmit. Any proposed changes to the study must be submitted to the HREB for approval prior to implementation.

All study-related documents should be retained so as to be available to the HREB on request. They should be kept for the duration of the project and for at least five years following study completion. In the case of clinical trials approved under Division 5 of the Food and Drug regulations of Health Canada, study records must be retained for 25 years.

Sincerely,

S.K.M. Kimber, MD, FRCPC
Chair, HREB Biomedical

Note: This correspondence includes an electronic signature (validation and approval via an online system).

UNIVERSITY OF ALBERTA Alberta Health Services Covenant Health

INFORMATION SHEET

Study Title: Multidisciplinary Approach to Novel Therapies In Cardiology Oncology REsearch (MANTICORE 101)

INVESTIGATORS:

I. Paterson, MD	U of A, Division of Cardiology	407-1857
M. Haykowsky, PhD	U of A, Dept. of Physical Therapy	492-5970
J. Mackey, MD	U of A, Dept. of Oncology	432-8221
J. Ezekowitz. MD	U of A, Division of Cardiology	407-8719
R. Thompson, PhD	U of A, Dept. of Biomedical Engineering	492-8665
S. Koshman. PharmD	U of A, Division of Cardiology	492.6560
G. Oudit, MD	U of A, Division of Cardiology	407-8569
E. Pituskin PhD(c)	U of A, Dept. of Oncology	432-8518

BACKGROUND

You are being asked to take part in this study because trastuzumab (Herceptin®) has been recommended as treatment for your breast cancer. While trastuzumab has been shown to prevent recurrences of breast cancer, some women may also experience damage to their heart muscle (including heart failure) as a result of trastuzumab treatment. Currently, there is no way of knowing who is at more risk of having this side effect, nor are there any proven ways to prevent this heart damage.

Another concern is that current tests (MUGA or multiple gated acquisition scan) do not detect early signs of heart damage. A new kind of scanner called MRI or 'magnetic resonance imaging' has been shown to detect early changes in how the heart muscle works and how blood flow within the heart.

PURPOSE

We hope to learn if standard medications used to treat heart failure can prevent heart damage caused by trastuzumab in women with breast cancer. We would also like to know more about when damage to the heart occurs during treatment, as well as if there are any ways to detect this damage earlier using MRI and blood tests.

DESCRIPTION OF THE STUDY

In this study, you will be randomly chosen (by a computer) to receive one of the treatments described below. Neither you nor the researcher will choose which treatment you will be assigned to. It is like flipping a coin to determine which treatments you will get.

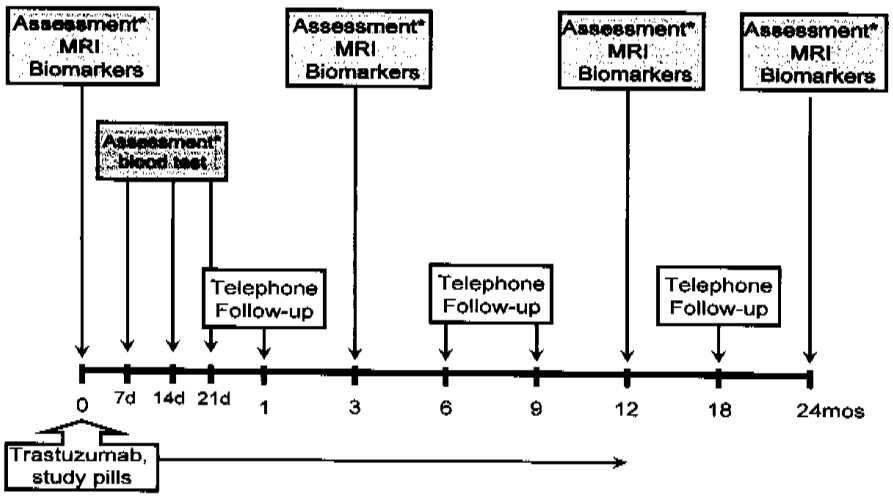
You will have a one in three equal chance of being assigned to:

- Receive a heart medication called perindopril,
- Receive a heart medication called bisoprolol,
- Receive placebo pills.

You will be provided with a supply of pills to take once a day for 1 year during the course of your trastuzumab chemotherapy. The dose of these medications will be gradually increased over the first 3 weeks. You and your doctor will not be told which treatment you are receiving. You will be followed regularly to see what effect the treatment has on your health and especially on your

heart. Participation in this study does not change the chemotherapy medications that have been recommended for you.

Schedule of Study Assessments:



Assessments will include:

- questions about your health history and how the pills are working for you,
- physical examination with blood pressure, pulse, temperature, breathing rate, weight, listening to lungs and heart, and looking for signs of heart problems,
- blood collection will include tests for markers of heart damage as well as monitoring of medications including electrolytes and kidney function measurements. These tests will be done before starting the medications, with increases in medication dose as well as at 3, 12 and 24 months. Extra tests may be done as needed to monitor medications.
- electric heart tracing (ECG) before starting the medications and as required to monitor medications
- cardiac MRI before starting the medications, 3 and 12 months after starting the medications and 1 year after the medications are finished (4 scans in total).

Depending on the treatment and scheduling of trastuzumab recommended by your oncologist, you may require an extra MRI scan. The study coordinator will talk about this, if it applies to you.

Also, depending on where you live, for the first month of this study, you may be eligible to borrow a blood pressure machine and have your blood checked at a laboratory close to your home. If you are eligible, the study coordinator will provide supplies and teaching.

Sample Banking for Future Research

You may also be asked if samples of your blood and urine can be stored for future research in the Alberta Research Tumor Bank. You will be given another consent form asking for your permission.

POSSIBLE RISKS

Cardiac MRI

Exposure to the magnetic fields in an MRI machine has no known harmful effects. As part of usual MRI procedure, you will be asked a series of questions to make sure it is safe before you have an MRI scan. The MRI contrast agent is very safe. Side effects of MRI contrast include temporary feeling of nausea or a cold feeling at the intravenous site (up to 3% of people). Other minor reactions include a rash or a headache. Very rarely people experience a more serious reaction (about 1:400,000) that could be life-threatening, such as wheezing, shortness of breath or decrease in blood pressure. If you experience a reaction during this study, the doctor supervising the MRI scan will treat you immediately. As well, a rare side called 'nephrogenic systemic fibrosis' has been reported in patients who have severe kidney disease and received MRI contrast. Symptoms of NSF include swelling, hardening and tightening of the skin, joint stiffness, reddened or darkened patches on the skin, among others. You will not be able to take part in this study if your kidney function is severely abnormal.

Medications

The following are most common side effects of each drug used in this study. These side effects may or may not be more severe when the drugs are taken together.

Drug	Common	Less Common	Rare
Perindopril	Headache Cough	Increase potassium levels Mild decrease kidney function Dizziness Rash	Severe allergic reaction Low white blood cell counts Kidney failure
Drug	**Common**	**Less Common**	**Rare**
Bisoprolol	Fatigue Diarrhea	Trouble sleeping Slow heart rate Dizziness	Cold extremities

These side effects may be temporary, long term and/or permanent. However, most of the side effects listed above are reversible and will stop or improve once the dose is lowered or the medication is discontinued. These side effects will be monitored closely during the study.

Blood Tests

You may feel some discomfort from the needle when blood drawn. There is also a small risk of fainting, swelling, bruising, bleeding or (rarely) local infections at the site of the needle punctures.

COSTS

You will not have to pay for the consultations that you receive by participating in this study. However, you may be coming to the University of Alberta more often than if you were not

participating in this study. As a result, there may be some extra costs for you such as gasoline, child care or meals.

COMPENSATION FOR INJURY

If you become ill or injured as a result of participating in this study, necessary medical treatment will be available at no additional cost to you. By signing this consent form you are not releasing the investigator(s), institution(s) and/or sponsor(s) from their legal and professional responsibilities.

CONTACTS

If you have any questions or concerns, please contact the investigators listed below:

Dr Ian Paterson at 780-407-1857, Dr John Mackey at 780-432-8221, Dr Sheri Koshman at 780-492-6560, Edith Pituskin PhD(c) at 780-432-8518 or Margo Miller RN at 780-989-5969.

CONFIDENTIALITY

Personal records will be kept confidential. Only the persons listed above will have access to your data. Any report published as a result of this study will not identify you by name.

For this study, the researchers may need to access your personal health records for health information such as past medical history and test results. The health information collected as part of this study will be kept confidential unless release is required by law, and will be used only for the purpose of the research study. By signing the consent form you give permission to the study staff to access any personally identifiable health information which is under the custody of other health care professionals as deemed necessary for the conduct of the research.

In addition to the investigators listed, the Health Research Ethics Board or the Alberta Cancer Research Ethics Board may have access to your personal health records to monitor the research and verify the accuracy of study data.

By signing the consent form you give permission for the collection, use and disclosure of your medical records. In Canada, study information is required to be kept for 5 years. Even if you withdraw from the study, the medical information which is obtained from you for study purposes will not be destroyed. You have a right to check your health records and request changes if your personal information is incorrect.

VOLUNTARY PARTICIPATION

You are free to withdraw from the research study at any time, and your continuing medical care will not be affected in any way. If the study is not undertaken or if it is discontinued at any time, the quality of your medical care will not be affected. If any knowledge gained from this or any other study becomes available which could influence your decision to continue in the study, you will be promptly informed.

CONTACT INFORMATION

If you have any questions or concerns about any aspect this study, you may contact the Health Research Ethics Board at 780 492-9724. This office has no affiliation with the study investigators.

CONSENT FORM

Multidisciplinary Approach to Novel Therapies In Cardiology Oncology REsearch (MANTICORE 101)

I. Paterson, MD	U of A, Division of Cardiology	407-1857
M. Haykowsky, PhD	U of A, Dept. of Physical Therapy	492-5970
J. Mackey, MD	U of A, Dept. of Oncology	432-8221
J. Ezekowitz. MD	U of A, Division of Cardiology	407-8719
R. Thompson, PhD	U of A, Dept. of Biomedical Engineering	492-8665
S. Koshman. PharmD	U of A, Division of Cardiology	492.6560
G. Oudit, MD	U of A, Division of Cardiology	407-8569
E. Pituskin PhD(c)	U of A, Dept. of Oncology	432-8518

Please answer the following questions:

	Yes	No
Do you understand that you are being asked to be in a research study?	___	___
Have you read and received a copy of an attached information sheet?	___	___
Do you understand the benefits and risks in taking part in this research study?	___	___
Have you had an opportunity to ask questions and discuss this study?	___	___
Do you understand that you are free to withdraw from the study at any time without having to give a reason and without affecting your future medical care?	___	___
Has the issue of confidentiality been explained to you, and do you understand who will have access to your medical records?	___	___
Do you want the investigators to inform you family doctor that you are participating in this study?	___	___

Who explained this study to you? _____

I agree to take part in this study: **Yes** ___ **No** ___

_____ _____
Signature of Participant Date

_____
Printed Name of Participant

_____ _____
Signature of Witness Date

I believe that the person signing this form understands what is involved in the study and voluntarily agrees to participate.

_____ _____
Signature of Investigator or Designee Date

APPENDIX B

Additional PhD work

THE ROLE OF EXERCISE INTERVENTIONS IN REDUCING THE RISK OF CARDIOMETABOLIC DISEASE IN CANCER SURVIVORS

(A version of this chapter has been published as: Pituskin E, Paterson I and Haykowsky M.

Current Cardiovascular Risk Reports, August 2012, Volume 6, Issue 4, pp 313-322)

B1.1 Introduction

Cancer is the leading cause of death worldwide, and accounted for 7.6 million deaths in 2008 in developed countries(17). The increased cancer incidence is due, in part to the aging population coupled with adherence to negative lifestyle. Indeed, behavioral and dietary risks account for at least of 30% of cancer deaths. Overweight/obesity, low fruit and vegetable intake coupled with high fat diet, physical inactivity, smoking and alcohol use are firmly established cancer-promoting, modifiable behaviors(5). Importantly, these same factors are well established, causative factors of cardiometabolic syndrome (CMS). CMS is not only associated with increased cancer risk, but more aggressive tumor biology(18, 19). Breast, gynecologic, colorectal, and prostate cancers represent the majority of new cancer diagnoses reported annually, influenced by many components of CMS(20). As such, most cancer patients are already at high baseline risk of cardio-metabolic disease well before anti-cancer therapy is initiated.

Initially coined ‘syndrome X’ (Reaven) CMS is a cluster of interconnected factors promoted by negative lifestyle choices(21). While specific criteria, interrelationships of components and prognostic utility of various CMS definitions is still evolving, common components of CMS across all definitions include: insulin resistance or impaired glucose tolerance; abdominal obesity or increased body-mass index (BMI); hypertension; and lipid profile abnormalities. Ranges and cut-off values for each of the components vary among the published definitions, hampering comparisons across patient groups and study results (Table 1). Further, controversy remains whether CMS represents a specific disorder or

represents an overall effect of combined risk factors(21). Nonetheless, these components are all associated with exercise intolerance, poor diet and ultimately increased risk of cardiovascular disease and death (22, 23).

In the general population, treatment of CMS is aimed primarily at lifestyle changes to ameliorate these factors, such as weight reduction, smoking cessation, dietary modification and increased physical activity; if these changes are insufficient, appropriate pharmacotherapy is initiated. In theory, cancer survivors should benefit even more from lifestyle modifications directed at alleviating components of CMS than the general population. The purpose of this paper is to review the potential role of exercise interventions in reducing the risk for cardio-metabolic disease in survivors of adult-onset malignancies.

B1.2 Significance of CMS for cancer survivors

In the last decade, important gains in cancer survival have been achieved with advances in anti-cancer therapies(2, 24). Unfortunately, these gains do not come without cost, with the 'multiple hits' of definitive surgery, systemic therapy and radiation therapy(6) negatively impacting upon pre-existing individual risk factors and behaviors, ultimately resulting in long-term complications of therapy. As a result, cancer survivors are at enhanced baseline risk of CMS and cardiovascular disease, such that for some, risk of death from cardiovascular disease may exceed that of recurrent cancer(25). For example, long-term follow-up of patients after treatment with anthracyclines demonstrated abnormal cardiac function present in 18% of patients followed up for less than 10 years and 38% of those followed for 10 years or more(26). Subclinical cardiac dysfunction may be

manifested by impaired exercise tolerance and fatigue(27),(28). Given that 70 – 100% of patients report exercise intolerance and fatigue during therapy and persisting years after treatment(29), it is likely that only 'the tip of the iceberg' of cardiovascular dysfunction and long-term morbidity is appreciated.

Given the variability in CMS definitions described in the literature, specific prevalence of CMS in cancer survivors is challenging to determine, but is l present in the majority of survivors(30). Further, certain patient populations may be at significantly heightened risk of CMS by virtue of baseline risk factors, the intensity of anti-cancer therapy administered and known short and long-term toxicities of specific anti-cancer agents. Here, influences of anti-cancer treatment effects on individual components of cardiometabolic disease will be reviewed.

B2.1 Weight gain, body composition and abdominal adiposity

Weight gain is frequently observed in cancer survivors. In spite of side effects commonly associated with receiving chemotherapy (nausea, vomiting, diarrhea, mouth sores, etc) with 50–96% of breast cancer patients reporting increased weight during treatment(31) with progressive gain in the months and years of follow-up(32). Weight gain in these patients is distinguished by a pattern of increasing fat mass but diminishing lean tissue, a phenotype known as sarcopenic obesity(33, 34). Such alterations to body composition are likely more relevant than actual weight gained, as sarcopenia (even in the presence of obesity) in cancer patients has been demonstrated as an independent risk factor for poor survival(35). Similar findings are observed in male cancer patients; in a meta-analysis of the impact of androgen deprivation therapy in prostate cancer (14

cohorts and 2 RCTs, n = 573 patients), Haseen reported adverse effects to body composition, with increased weight, increased fat and decreased lean body mass in treated vs non-treated patients(36). Negative body composition changes were noted to start as early as one month of treatment, with longer therapy associated with greater weight gain over time. Weight gain and loss of lean body mass is also commonly observed in patients surviving high-dose chemotherapy and bone marrow transplant (HDCT/BMT); at a median 5 years post-treatment, 38% of patients do not regain their pre-BMT lean body mass. Further, while the prevalence of overweight/obese (body-mass index [BMI] of <25 kg/m^2) was 15% at 6 months post-BMT, this increased to 50% of patients at 2–3 and 4–6 years. These findings indicate that body fat is restored without a corresponding expansion of lean muscle mass; furthermore, these effects are exacerbated with time without directed intervention.

In the general population, not only weight gain but distribution of weight is a predictor of death. In multiple large prospective studies, with adjustment for BMI, abdominal fat distribution is positively associated with the risk of death(37-39). This association is stronger among participants with a lower BMI than among those with a higher BMI (particularly women), highlighting the importance of this metric. Currently, the direct contribution of anti-cancer therapies to abdominal girth is unknown, but given the known effects of weight gain, is likely negative.

B2.2 Metabolic alterations: glucose intolerance and dyslipidemia

Metabolic disturbances are frequently observed in cancer survivors, exacerbated by systemic therapy, radiotherapy or adjunctive medications.

Dexamethasone is a potent glucocorticoid commonly used in cancer therapy as an anti-emetic adjunct, to prevent hypersensitivity reactions associated with systemic therapy(40) and in high doses as a chemotherapeutic agent in hematologic malignancies(41). As a result, impaired insulin sensitivity and elevated serum glucose levels have been observed even with short-term, conservative doses in breast cancer patients(42). Chemotherapy dose intensity and multi-drug regimens may also influence negative metabolic changes. In survivors of HDCT/BMT, high rates of hyper-triglyceridemia are observed a median 3 years post-transplant(43). Chow also observed increased rates of renal disease, dyslipidemia and diabetes in 1491 patients surviving 2 or more years after HDCT compared to population-based controls(44).

Hormone blockade is a key therapy in breast and prostate cancer, however, conveys established adverse effects to lipid profiles. Androgen deprivation therapy is associated with increased fasting total cholesterol and triglycerides in treated prostate cancer patients compared to untreated(45, 46). Aromatase inhibitors, while demonstrating relative risk benefits for breast cancer outcomes, have demonstrated a class effect of elevated serum cholesterol(47). In 47 women ~34 months following chemotherapy completion and receiving hormonal therapy, we observed a more unfavorable lipid profile in patients receiving an aromatase inhibitor.

B2.3 Hypertension

The contribution of hypertension to development of heart failure is well-known in the general population(48, 49). This is also observed in cancer patients,

where pre-existing hypertension exacerbates cardiac dysfunction associated with anti-cancer therapies, including anthracyclines and trastuzumab(50-53). In an analysis of SEER–Medicare database data comparing known cardiac risk factors and anthracyclines in breast and lymphoma patients, only hypertension was found to potentiate heart failure risk(54, 55). In trastuzumab-treated patients, the NSABP B-31 study showed that age (>50), current use of antihypertensive medications and low baseline LVEF values (50 – 54%) predicted later cardiotoxicity (53). In a cohort of autologous HDCT patients (n = 1327) who received conservative anthracycline dose (< 250mg/m^2), the presence of hypertension resulted in a 35-fold increased risk of heart failure **(56).**

Hypertension is a frequent toxicity of anti-cancer medications targeting angiogenesis. In patients receiving sunitinib, a tyrosine kinase inhibitor of vascular endothelial growth factor (VEGF) inhibitor, hypertension rates are significant (30 – 50%)(57, 58) and occur as rapidly as within the first cycle of treatment(59). In bevacizumab, a humanized monoclonal antibody that inhibits VEGF activity, meta-analysis revealed that the summary incidence of all-grade hypertension was 23.6%(60). Hypertension is a challenging toxicity in these patients, for many have few other treatment options. Additionally, treatment must be balanced by sufficient dose to offer clinical efficacy while avoiding potential acute and long-term effects including heart failure. Interestingly, retrospective reviews of patients receiving sunitinib suggest that hypertension may be a marker of response(61, 62), highlighting this challenge.

B2.4 Exercise intolerance and cardiorespiratory dysfunction

It is concerning that maximal exercise capacity (VO_{2peak}) of cancer patients has been consistently demonstrated at approximately 30% below that of matched sedentary, healthy individuals(63-65) with a significant proportion of adult female cancer patients unable to meet the minimum threshold for independent living (VO2peak of 15mL/kg/min)(66). This finding is imperative, give that VO_{2peak} is the strongest independent predictor of mortality in healthy individuals and cardiac patients(67-71). Consequently, patients must exert maximal effort and become easily fatigued with routine activities of daily living, resulting in loss of functional independence and quality of life. This impaired aerobic capacity is observed in spite of normal left ventricular ejection fraction ($\geq$ 50%) suggesting that treatment-related injury extends to affect other oxygen-transport organs and systems(72, 73) and highlighting the effects of subclinical cardiotoxicity. Indeed, reduced systolic function is now recognized as a late effect of cardiotoxic drug exposure, not detectable until significant damage has already occurred(74).

For many cancer patients, particularly breast and hematologic, therapeutic approaches often involve multiple treatment modalities, each conveying various negative cardiovascular effects. The incidence of radiation-induced heart disease in breast cancer patients is estimated anywhere between 10 and 30% by 5 – 10 years post-treatment, with up to 88% of patients having asymptomatic abnormalities of the heart muscle, valves, pericardium, conduction abnormalities and vasculature(75). Even with modern radiotherapy techniques and complex treatment planning to avoid organs at risk(76), in left-sided breast cancers,

sufficient dose is delivered to the heart to effect myocardial perfusion defects in more than 50% of patients(77). Cytotoxic chemotherapeutic agents are all associated with some degree of acute or long-term cardiotoxic effect. In particular, anthracycline-based regimens have received attention, given their widespread use across multiple malignancies and known cardiotoxic effects. Risk factors for development of anthracycline-related cardiomyopathy include cardiac irradiation, hypertension, coronary artery disease and age > 65 years(78).

Cardiac toxicity of targeted therapies such as trastuzumab has emerged as a significant complication; clinical heart failure was observed in up to 4% of treated patients and asymptomatic declines in ventricular function have been reported in up to 18% in randomized trials(79-81). The risk of cardiac toxicity was enhanced for those receiving concurrent anthracycline chemotherapy. These multiple 'hits' 'to the cardiovascular system convey varying degrees of directly negative effects in the short and long-term(6). In 41 breast cancer patients who had received trastuzumab-based chemotherapy, we showed that at ~20 months following completion of therapy survivors consistently had higher rates of cardiovascular risk factors. Here, higher rates of overweight/obesity, low cardiorespiratory fitness (VO2peak)and unfavorable lipid profiles were observed compared to controls(82). In 47 women who had received chemotherapy and endocrine therapy for hormone-receptor positive breast cancer, we again found consistently less favorable cardiovascular risk factors compared to controls. Higher resting heart rate and systolic blood pressure were observed; peak exercise output, stroke volume and cardiac power output were significantly lower in

patients than controls(83). While these studies are limited by cross-sectional designs limiting our understanding of baseline characteristics, the results consistently identify significant clinical and sub-clinical negative cardiovascular effects of anti-cancer therapy. As such, these survivors are likely increasingly vulnerable to progressive dysfunction over time, and research into approaches to ameliorate these negative effects is urgently required.

B3.1 Effects of exercise interventions on CMS in cardiology

There is a wealth of evidence supporting exercise therapy in the rehabilitation of cardiac disease. In patients with heart failure, exercise training has been proven safe and effective, improving functional capacity and reducing symptoms of dyspnea(84, 85). Randomized trials have demonstrated improvements of VO2 peak in the range of 18 – 25%(86). The most recent systematic review produced by the Cochrane group summarizes results of 47 studies randomizing 10,794 patients to exercise-based cardiac rehabilitation or usual care. Overall, exercise-based cardiac rehabilitation provides significant health and quality of life benefit, and has proven effectiveness in reducing total and cardiovascular mortality (in medium to longer term studies) and hospital admissions (in shorter term studies).(87) As such, exercise is a Class 1 recommendation of the American College of Cardiology and the American Heart Association(88).

Training intensity and modality have been shown to influence benefits. Wisloff randomized 27 patients with stable post-infarction heart failure on optimal pharmacotherapy to either moderate continuous, higher intensity aerobic

interval training or usual care. VO2peak increased with interval training more than continuous training (46% versus 14%, P<0.001) and was associated with improved cardiac geometry and function, with ejection fraction increasing by 35%(89). No changes were observed in the control group. We undertook a meta-analysis of randomized controlled trials examining the type of exercise (aerobic vs. strength vs. combined training) on cardiac geometry in heart failure. In reviewing 14 trials (n = 812 patients) we observed improved cardiac geometry with aerobic training, which was not observed with combined aerobic and strength training. The magnitude of benefit seen with exercise training was particularly noteworthy, given that all patients were receiving optimal pharmacotherapy during their training interventions.

Importantly, exercise training has been shown to influence aspects of CMS in patients with cardiovascular disease. Exercise training promotes weight and fat loss, improved lipid profile and glucose indices and decreases blood pressure(90). Furthermore, investigations into increasing exercise intensity and exercise duration have shown to convey additional benefit for patients. Ades randomized 74 overweight patients with coronary heart disease to a high expenditure exercise program (3000 – 3500 kcal/week) compared to standard exercise programming (700 – 800 kcal/week); both groups received the same counseling and medication review. The authors found that patients in the high expenditure arm experienced double the weight loss (8.2±4 vs 3.7±5 kg, P<0.001) fat mass loss (5.9±4 vs 2.8±3 kg, P< 0.001) and a greater waist reduction (-7±5 vs. -5±5 cm, P=0.02) than standard exercise at 5 months(91). Improvements in

lipid and insulin profile and blood pressure were observed; notably, weight loss was maintained by the participants at one year. Taken together, exercise rehabilitation is considered an important and integral aspect of cardiac care, evidenced by the recommendation of specific multidisciplinary program components by the American Heart Association(92).

B3.2 Effects of exercise interventions on CMS in oncology populations

Exercise interventions have the potential to benefit cancer patients from multiple avenues. Our group and others have shown that maximal exercise testing with expired gas exchange analysis) can be safely conducted in cancer populations(82, 83, 93-95), including those with advanced disease(64, 65, 96). We also conducted a meta-analysis of RCTs to determine the effect of supervised exercise training on exercise capacity (VO2peak). Six studies were included in the analysis, with n = 449 patients randomized to varying exercise prescriptions, timing, and duration of intervention. The main finding was that relatively short-term, supervised moderate-intensity training is associated with significant improvements in VO2peak during and after curative-intent therapy(97). Specifically, the weighted mean difference in VO2peak was 2.91 ml/kg/min from baseline to post-intervention, favoring exercise training.

McNeely et al reported similar findings in a meta-analysis of randomized, controlled trials of exercise interventions in breast cancer patients(98). In spite of significant heterogeneity in the timing of interventions, type of exercise interventions and reported outcomes in the individual studies, pooled results showed a moderate to large effect size of exercise on fatigue symptoms and

exercise capacity, with VO2peak improving by 3.39mL/kg/min, or nearly 1 metabolic equivalent. While the prognostic relevance of this improvement in adult cancer patients is not yet known, mortality risk as been shown to decrease by 12% for each 1-metabolic equivalent (MET) improvement in aerobic capacity in healthy women(99). In a meta-analysis of exercise interventions in hematologic survivors of HDCT, Wiskemann reported multiple benefits, including maintenance of physical performance, maintenance of weight and lean body mass and decreased fatigue (n = 609 total patients, 15 studies)(100). Taking these and other findings into consideration, the American College of Sports Medicine has developed guidelines for cancer survivors; in their review, exercise therapy was determined to be safe, with benefits observed in terms of aerobic fitness, muscular strength and body composition(101).

Exercise interventions have also been effective in ameliorating some components of CMS. In breast cancer patients, a 15-week combination training intervention improved aerobic capacity, weight and waist/hip ratio(102). In the largest intervention study to date, 242 breast cancer patients were randomized to supervised resistance training, supervised aerobic exercise or usual care for 17 weeks, showing improvements in muscle strength and lean body mass with resistance exercise; improved aerobic fitness and body fat was seen in the aerobic exercise group(103). In prostate cancer patients receiving androgen deprivation therapy, a 20 week supervised resistance training program preserved body composition and improved muscle strength and endurance(104). Further work is required to elucidate the effects of exercise interventions on other important

aspects of CMS present at diagnosis in cancer patients, including metabolic alterations and hypertension.

B4 Future directions

In summary, there is compelling evidence to support exercise training as therapy for detrimental cardiometabolic effects of cancer treatments. However, the majority of the data is derived from breast, prostate and hematologic malignancies; data for other disease sites such as colon and gynecologic cancers remain limited. Given the wide range in treatment modalities for different malignancies, there remain significant gaps in our knowledge of exercise interventions for specific cancer patient populations, limiting our ability to make broad recommendations. It is vital that researchers adopt strategic approaches and build upon current knowledge, applying rigorous methodology(97).

Given limited resources in health care, the development of evidence-based nomograms may be useful in determining those patients most at risk. Improved understanding of baseline risk factors, effects of drug and dose intensity, duration of anti-cancer treatments and individual genetic variability will guide customized exercise interventions for each patient. As oncology enters the era of ‘personalized medicine’, accordingly research into personalized exercise interventions to ameliorate negative effects is warranted.

Questions remain regarding optimal training intensity. While moderate-intensity exercise is effective in cancer patients, it is possible that high-intensity may be beneficial and potentially more tolerable than longer duration exercise. In particular, for cancer patients with impaired exercise tolerance, brief and intense

bursts of exercise may be more manageable given easy fatigability with even routine activities of daily living. The beneficial effects of this training approach has been observed in patients with heart failure, and well-designed trials are currently underway studying the effects of exercise intensity in cancer patients(16).

Another potential area of study is the optimal multidisciplinary team complement. Given the complexities of cardiometabolic disease, individual risks and various cancer treatments, it is unlikely that one professional possesses all the necessary knowledge for effective interventions. Multidisciplinary care teams have been shown to be effective in obesity management(7); such teams have also been shown to reduce mortality and hospitalizations among heart failure patients(8-10). Currently, the effect of multidisciplinary interventions in cancer patients receiving cardiotoxic chemotherapy regimens is unknown. Our group is embarking on a primary prevention trial randomizing breast and lymphoma patients to intensive multidisciplinary and exercise intervention compared to usual care(11). Exercise physiologists, pharmacists, dieticians, nurses and physicians will regularly assess patients randomized to the intervention arm, with the aim of improving health and avoiding cardiac dysfunction. We expect our results to inform comprehensive, holistic care for patients at risk for cardiometabolic sequelae.

Most importantly, the discipline of oncology may well learn from the cardiology experience. The example of exercise rehabilitation as a class 1 recommendation and an integral aspect of cardiac care can inform similar

approaches in cancer survivors. Research into important components of multidisciplinary cancer rehabilitation, evaluating components of cardiology and other chronic diseases approaches as well as developing aspects unique for cancer survivor rehabilitation should be undertaken.

B5 References

1. Cancer Fact Sheet. World Health Organization. http://www.who.int/mediacentre/factsheets/fs297/en/. Accessed January 2012.

2. Anand SS, Yusuf S. Stemming the global tsunami of cardiovascular disease. The Lancet.377(9765):529-32.

3. Healy LA, Ryan AM, Carroll P, Ennis D, Crowley V, Boyle T et al. Metabolic Syndrome, Central Obesity and Insulin Resistance are Associated with Adverse Pathological Features in Postmenopausal Breast Cancer. Clinical Oncology.22(4):281-8.

4. Maiti B, Kundranda M, Spiro T, Daw H. The association of metabolic syndrome with triple-negative breast cancer. Breast Cancer Research and Treatment.121(2):479-83. doi:10.1007/s10549-009-0591-y.

5. Cowey S, Hardy RW. The metabolic syndrome: A high-risk state for cancer? Am J Pathol. 2006;169(5):1505-22. doi:S0002-9440(10)62617-X [pii] 10.2353/ajpath.2006.051090.

6. Kassi E, Pervanidou P, Kaltsas G, Chrousos G. Metabolic syndrome: definitions and controversies. BMC Med.9:48. doi:1741-7015-9-48 [pii] 10.1186/1741-7015-9-48.

7. Lakka HM, Laaksonen DE, Lakka TA, Niskanen LK, Kumpusalo E, Tuomilehto J et al. The metabolic syndrome and total and cardiovascular disease mortality in middle-aged men. JAMA. 2002;288(21):2709-16. doi:joc21220 [pii].

8. Horwich TB, Fonarow GC. Glucose, obesity, metabolic syndrome, and diabetes relevance to incidence of heart failure. J Am Coll Cardiol.55(4):283-93. doi:S0735-1097(09)02673-4 [pii]
10.1016/j.jacc.2009.07.029.

9. Canadian Cancer Statistics. Toronto, Canada2010.

10. Cancer Trends Progress Report - 2009/2010 Update. National Cancer Institute. http://progressreport.cancer.gov/highlights.asp. Accessed January 2012.

11. Jones LW, Haykowsky MJ, Swartz JJ, Douglas PS, Mackey JR. Early breast cancer therapy and cardiovascular injury. J Am Coll Cardiol. 2007;50(15):1435-41. doi:S0735-1097(07)02240-1 [pii]10.1016/j.jacc.2007.06.037.

12. Hanrahan EO, Gonzalez-Angulo AM, Giordano SH, Rouzier R, Broglio KR, Hortobagyi GN et al. Overall survival and cause-specific mortality of patients with stage T1a,bN0M0 breast carcinoma. J Clin Oncol. 2007;25(31):4952-60. doi:25/31/4952 [pii]
10.1200/JCO.2006.08.0499.

13. Steinherz LJ, Steinherz PG, Tan CT, Heller G, Murphy ML. Cardiac toxicity 4 to 20 years after completing anthracycline therapy. JAMA. 1991;266(12):1672-7.

14. Schunemann M, Anker SD, Rauchhaus M. Cancer fatigue syndrome reflects clinically non-overt heart failure: an approach towards onco-cardiology. Nat Clin Pract Oncol. 2008;5(11):632-3. doi:ncponc1226 [pii]10.1038/ncponc1226.

15. McNeely ML, Courneya KS. Exercise programs for cancer-related fatigue: evidence and clinical guidelines. J Natl Compr Canc Netw.8(8):945-53. doi:8/8/945 [pii].

16. Hofman M, Ryan JL, Figueroa-Moseley CD, Jean-Pierre P, Morrow GR. Cancer-related fatigue: the scale of the problem. Oncologist. 2007;12 Suppl 1:4-10. doi:12/suppl_1/4 [pii]
10.1634/theoncologist.12-S1-4.

17. de Haas EC, Oosting SF, Lefrandt JD, Wolffenbuttel BHR, Sleijfer DT, Gietema JA. The metabolic syndrome in cancer survivors. The Lancet Oncology.11(2):193-203.

18. Rock CL, Demark-Wahnefried W. Nutrition and survival after the diagnosis of breast cancer: a review of the evidence. J Clin Oncol. 2002;20(15):3302-16.

19. Irwin ML, McTiernan A, Baumgartner RN, Baumgartner KB, Bernstein L, Gilliland FD et al. Changes in body fat and weight after a breast cancer diagnosis: influence of demographic, prognostic, and lifestyle factors. J Clin Oncol. 2005;23(4):774-82. doi:23/4/774 [pii]
10.1200/JCO.2005.04.036.

20. Visovsky C. Muscle Strength, Body Composition, and Physical Activity in Women Receiving Chemotherapy for Breast Cancer. Integrative Cancer Therapies. 2006;5(3):183-91. doi:10.1177/1534735406291962.

21. Vance V, Mourtzakis M, McCargar L, Hanning R. Weight gain in breast cancer survivors: prevalence, pattern and health consequences. Obesity Reviews.12(4):282-94. doi:10.1111/j.1467-789X.2010.00805.x.

22. Prado CMM, Lieffers JR, McCargar LJ, Reiman T, Sawyer MB, Martin L et al. Prevalence and clinical implications of sarcopenic obesity in patients with solid tumours of the respiratory and gastrointestinal tracts: a population-based study. The Lancet Oncology. 2008;9(7):629-35.

23. Haseen F, Murray LJ, Cardwell CR, O'Sullivan JM, Cantwell MM. The effect of androgen deprivation therapy on body composition in men with prostate cancer: systematic review and meta-analysis. J Cancer Surviv.4(2):128-39. doi:10.1007/s11764-009-0114-1.

24. Pischon T, Boeing H, Hoffmann K, Bergmann M, Schulze MB, Overvad K et al. General and abdominal adiposity and risk of death in Europe. N Engl J Med. 2008;359(20):2105-20. doi:359/20/2105 [pii]10.1056/NEJMoa0801891.

25. Jacobs EJ, Newton CC, Wang Y, Patel AV, McCullough ML, Campbell PT et al. Waist Circumference and All-Cause Mortality in a Large US Cohort. Arch Intern Med.170(15):1293-301. doi:10.1001/archinternmed.2010.201.

26. Calle EE, Rodriguez C, Jacobs EJ, Almon ML, Chao A, McCullough ML et al. The American Cancer Society Cancer Prevention Study II Nutrition Cohort: rationale, study design, and baseline characteristics. Cancer. 2002;94(2):500-11. doi:10.1002/cncr.10197.

27. Basch E, Prestrud AA, Hesketh PJ, Kris MG, Feyer PC, Somerfield MR et al. Antiemetics: American Society of Clinical Oncology clinical practice guideline update. J Clin Oncol.29(31):4189-98. doi:JCO.2010.34.4614 [pii]10.1200/JCO.2010.34.4614.

28. Fisher RI, Gaynor ER, Dahlberg S, Oken MM, Grogan TM, Mize EM et al. Comparison of a Standard Regimen (CHOP) with Three Intensive Chemotherapy Regimens for Advanced Non-Hodgkin's Lymphoma. New England Journal of Medicine. 1993;328(14):1002-6. doi:doi:10.1056/NEJM199304083281404.

29. Hickish T, Astras G, Thomas P, Penfold S, Purandare L, Hickish TF et al. Glucose Intolerance During Adjuvant Chemotherapy for Breast Cancer. Journal of the National Cancer Institute. 2009;101(7):537. doi:10.1093/jnci/djp025.

30. Majhail NS, Flowers ME, Ness KK, Jagasia M, Carpenter PA, Arora M et al. High prevalence of metabolic syndrome after allogeneic hematopoietic cell transplantation. Bone Marrow Transplant. 2008;43(1):49-54.

31. Chow EJ, Mueller BA, Baker KS, Cushing-Haugen KL, Flowers MED, Martin PJ et al. Cardiovascular Hospitalizations and Mortality Among Recipients of Hematopoietic Stem Cell Transplantation. Annals of Internal Medicine.155(1):21-32. doi:10.1059/0003-4819-155-1-201107050-00004.

32. Saylor PJ, Smith MR. Metabolic complications of androgen deprivation therapy for prostate cancer. J Urol. 2009;181(5):1998-2006; discussion 7-8. doi:S0022-5347(09)00069-X [pii]
10.1016/j.juro.2009.01.047.

33. Braga-Basaria M, Muller DC, Carducci MA, Dobs AS, Basaria S. Lipoprotein profile in men with prostate cancer undergoing androgen deprivation therapy. Int J Impot Res. 2006;18(5):494-8.

34. Ewer MS, Gluck S. A woman's heart: the impact of adjuvant endocrine therapy on cardiovascular health. Cancer. 2009;115(9):1813-26. doi:10.1002/cncr.24219.

35. Levy D, Larson MG, Vasan RS, Kannel WB, Ho KK. The progression from hypertension to congestive heart failure. JAMA. 1996;275(20):1557-62.

36. Kannel WB. Incidence and Epidemiology of Heart Failure. Heart Failure Reviews. 2000;5(2):167-73. doi:10.1023/a:1009884820941.

37. Schocken DD, Benjamin EJ, Fonarow GC, Krumholz HM, Levy D, Mensah GA et al. Prevention of heart failure: a scientific statement from the American Heart Association Councils on Epidemiology and Prevention, Clinical Cardiology, Cardiovascular Nursing, and High Blood Pressure Research; Quality of Care and Outcomes Research Interdisciplinary Working Group; and Functional Genomics and Translational Biology Interdisciplinary Working Group. Circulation. 2008;117(19):2544-65. doi:CIRCULATIONAHA.107.188965 [pii] 10.1161/CIRCULATIONAHA.107.188965.

38. Yeh ET. Cardiotoxicity induced by chemotherapy and antibody therapy. Annu Rev Med. 2006;57:485-98. doi:10.1146/annurev.med.57.121304.131240.

39. Yeh ET, Tong AT, Lenihan DJ, Yusuf SW, Swafford J, Champion C et al. Cardiovascular complications of cancer therapy: diagnosis, pathogenesis, and management. Circulation. 2004;109(25):3122-31. doi:10.1161/01.CIR.0000133187.74800.B9 109/25/3122 [pii].

40. Tan-Chiu E, Yothers G, Romond E, Geyer CE, Jr., Ewer M, Keefe D et al. Assessment of cardiac dysfunction in a randomized trial comparing doxorubicin and cyclophosphamide followed by paclitaxel, with or without trastuzumab as adjuvant therapy in node-positive, human epidermal growth factor receptor 2-overexpressing breast cancer: NSABP B-31. J Clin Oncol. 2005;23(31):7811-9. doi:23/31/7811 [pii]
10.1200/JCO.2005.02.4091.

41. Pinder MC, Duan Z, Goodwin JS, Hortobagyi GN, Giordano SH. Congestive heart failure in older women treated with adjuvant anthracycline chemotherapy for breast cancer. J Clin Oncol. 2007;25(25):3808-15. doi:JCO.2006.10.4976 [pii]10.1200/JCO.2006.10.4976.

42. Ryberg M, Nielsen D, Skovsgaard T, Hansen J, Jensen BV, Dombernowsky P. Epirubicin cardiotoxicity: an analysis of 469 patients with metastatic breast cancer. J Clin Oncol. 1998;16(11):3502-8.

43. Armenian SH, Sun C-L, Shannon T, Mills G, Francisco L, Venkataraman K et al. Incidence and predictors of congestive heart failure after autologous hematopoietic cell transplantation. Blood.118(23):6023-9. doi:10.1182/blood-2011-06-358226.

44. Chu TF, Rupnick MA, Kerkela R, Dallabrida SM, Zurakowski D, Nguyen L et al. Cardiotoxicity associated with tyrosine kinase inhibitor sunitinib. Lancet. 2007;370(9604):2011-9. doi:S0140-6736(07)61865-0 [pii]
10.1016/S0140-6736(07)61865-0.

45. Schmidinger M, Zielinski CC, Vogl UM, Bojic A, Bojic M, Schukro C et al. Cardiac Toxicity of Sunitinib and Sorafenib in Patients With Metastatic Renal Cell Carcinoma. J Clin Oncol. 2008;26(32):5204-12. doi:10.1200/jco.2007.15.6331.

46. Khakoo AY, Kassiotis CM, Tannir N, Plana JC, Halushka M, Bickford C et al. Heart failure associated with sunitinib malate: a multitargeted receptor tyrosine kinase inhibitor. Cancer. 2008;112(11):2500-8. doi:10.1002/cncr.23460.

47. Ranpura V, Pulipati B, Chu D, Zhu X, Wu S. Increased Risk of High-Grade Hypertension With Bevacizumab in Cancer Patients: A Meta-Analysis. Am J Hypertens.23(5):460-8.

48. Scartozzi M, Galizia E, Chiorrini S, Giampieri R, Berardi R, Pierantoni C et al. Arterial hypertension correlates with clinical outcome in colorectal cancer patients treated with first-line bevacizumab. Annals of Oncology. 2009;20(2):227-30. doi:10.1093/annonc/mdn637.

49. Rini BI, Cohen DP, Lu DR, Chen I, Hariharan S, Gore ME et al. Hypertension as a Biomarker of Efficacy in Patients With Metastatic Renal Cell Carcinoma Treated With Sunitinib. Journal of the National Cancer Institute.103(9):763-73. doi:10.1093/jnci/djr128.

50. Jones LW, Haykowsky M, Pituskin EN, Jendzjowsky NG, Tomczak CR, Haennel RG et al. Cardiovascular Reserve and Risk Profile of Postmenopausal Women After Chemoendocrine Therapy for Hormone Receptor Positive Operable Breast Cancer. Oncologist. 2007;12(10):1156-64. doi:10.1634/theoncologist.12-10-1156.

51. Jones LW, Eves ND, Mackey JR, Peddle CJ, Haykowsky M, Joy AA et al. Systemic inflammation, cardiorespiratory fitness, and quality of life in patients with advanced non-small cell lung cancer. J Thorac Oncol. 2008;3(2):194-5. doi:10.1097/JTO.0b013e318160f36b
01243894-200802000-00019 [pii].

52. Jones LW, Eves ND, Mackey JR, Peddle CJ, Haykowsky M, Joy AA et al. Safety and feasibility of cardiopulmonary exercise testing in patients with advanced cancer. Lung Cancer. 2007;55(2):225-32. doi:S0169-5002(06)00540-X [pii]
10.1016/j.lungcan.2006.10.006.

53. Paterson DH, Cunningham DA, Koval JJ, St Croix CM. Aerobic fitness in a population of independently living men and women aged 55-86 years. Med Sci Sports Exerc. 1999;31(12):1813-20.

54. Jones LW, Eves ND, Haykowsky M, Joy AA, Douglas PS. Cardiorespiratory exercise testing in clinical oncology research: systematic review and practice recommendations. Lancet Oncol. 2008;9(8):757-65. doi:S1470-2045(08)70195-5 [pii]
10.1016/S1470-2045(08)70195-5.

55. Hsich E, Gorodeski EZ, Starling RC, Blackstone EH, Ishwaran H, Lauer MS. Importance of Treadmill Exercise Time as an Initial Prognostic Screening Tool in Patients With Systolic Left Ventricular Dysfunction. Circulation. 2009:CIRCULATIONAHA.109.848382. doi:10.1161/circulationaha.109.848382.

56. Grewal J, McCully RB, Kane GC, Lam C, Pellikka PA. Left Ventricular Function and Exercise Capacity. JAMA. 2009;301(3):286-94. doi:10.1001/jama.2008.1022.

57. Myers J, Prakash M, Froelicher V, Do D, Partington S, Atwood JE. Exercise Capacity and Mortality among Men Referred for Exercise Testing. N Engl J Med. 2002;346(11):793-801. doi:10.1056/NEJMoa011858.

58. Arena R, Myers J, Williams MA, Gulati M, Kligfield P, Balady GJ et al. Assessment of Functional Capacity in Clinical and Research Settings: A Scientific Statement From the American Heart Association Committee on Exercise, Rehabilitation, and Prevention of the Council on Clinical Cardiology and the Council on Cardiovascular Nursing. Circulation. 2007;116(3):329-43. doi:10.1161/circulationaha.106.184461.

59. Jones LW, Eves ND, Haykowsky M, Freedland SJ, Mackey JR. Exercise intolerance in cancer and the role of exercise therapy to reverse dysfunction. Lancet Oncol. 2009;10(6):598-605. doi:S1470-2045(09)70031-2 [pii]10.1016/S1470-2045(09)70031-2.

60. Khakoo AY, Liu PP, Force T, Lopez-Berestein G, Jones LW, Schneider J et al. Cardiotoxicity due to cancer therapy. Tex Heart Inst J.38(3):253-6.

61. Zuppinger C, Timolati F, Suter TM. Pathophysiology and diagnosis of cancer drug induced cardiomyopathy. Cardiovasc Toxicol. 2007;7(2):61-6. doi:CT:7:2:61 [pii] 10.1007/s12012-007-0016-2.

62. Carver JR, Shapiro CL, Ng A, Jacobs L, Schwartz C, Virgo KS et al. American Society of Clinical Oncology Clinical Evidence Review on the Ongoing Care of Adult Cancer Survivors: Cardiac and Pulmonary Late Effects. J Clin Oncol. 2007;25(25):3991-4008. doi:10.1200/jco.2007.10.9777.

63. Taylor CW, Bronnum D, Darby SC, Gagliardi G, Hall P, Jensen MB et al. Cardiac dose estimates from Danish and Swedish breast cancer radiotherapy during 1977-2001. Radiother Oncol.100(2):176-83. doi:S0167-8140(11)00035-1 [pii]10.1016/j.radonc.2011.01.020.

64. Marks LB, Yu X, Prosnitz RG, Zhou SM, Hardenbergh PH, Blazing M et al. The incidence and functional consequences of RT-associated cardiac perfusion defects. Int J Radiat Oncol Biol Phys. 2005;63(1):214-23. doi:S0360-3016(05)00157-4 [pii]10.1016/j.ijrobp.2005.01.029.

65. Ewer MS, Ewer SM. Cardiotoxicity of anticancer treatments: what the cardiologist needs to know. Nat Rev Cardiol.7(10):564-75. doi:nrcardio.2010.121 [pii]10.1038/nrcardio.2010.121.

66. Romond EH, Perez EA, Bryant J, Suman VJ, Geyer CE, Jr., Davidson NE et al. Trastuzumab plus adjuvant chemotherapy for operable HER2-positive breast cancer. N Engl J Med. 2005;353(16):1673-84. doi:353/16/1673 [pii] 10.1056/NEJMoa052122.

67. Suter TM, Procter M, van Veldhuisen DJ, Muscholl M, Bergh J, Carlomagno C et al. Trastuzumab-associated cardiac adverse effects in the herceptin adjuvant trial. J Clin Oncol. 2007;25(25):3859-65. doi:JCO.2006.09.1611 [pii] 10.1200/JCO.2006.09.1611.

68. Slamon D, Eiermann W, Robert N, Pienkowski T, Martin M, Press M et al. Adjuvant trastuzumab in HER2-positive breast cancer. N Engl J Med.365(14):1273-83. doi:10.1056/NEJMoa0910383.

69. Jones LW, Haykowsky M, Peddle CJ, Joy AA, Pituskin EN, Tkachuk LM et al. Cardiovascular Risk Profile of Patients with HER2/neu-Positive Breast Cancer Treated with Anthracycline-Taxane-Containing Adjuvant Chemotherapy and/or Trastuzumab. Cancer Epidemiol Biomarkers Prev. 2007;16(5):1026-31. doi:10.1158/1055-9965.epi-06-0870.

70. Jones LW, Haykowsky M, Pituskin EN, Jendzjowsky NG, Tomczak CR, Haennel RG et al. Cardiovascular reserve and risk profile of postmenopausal women after chemoendocrine therapy for hormone receptor--positive operable breast cancer. Oncologist. 2007;12(10):1156-64. doi:12/10/1156 [pii] 10.1634/theoncologist.12-10-1156.

71. Genest J, McPherson R, Frohlich J, Anderson T, Campbell N, Carpentier A et al. 2009 Canadian Cardiovascular Society/Canadian guidelines for the diagnosis and treatment of dyslipidemia and prevention of cardiovascular disease in the adult - 2009 recommendations. Can J Cardiol. 2009;25(10):567-79.

72. Arthur HM, Suskin N, Bayley M, Fortin M, Howlett J, Heckman G et al. The Canadian Heart Health Strategy and Action Plan: Cardiac rehabilitation as an exemplar of chronic disease management. Can J Cardiol.26(1):37-41.

73. Downing J, Balady GJ. The role of exercise training in heart failure. J Am Coll Cardiol.58(6):561-9. doi:S0735-1097(11)01780-3 [pii] 10.1016/j.jacc.2011.04.020.

74. Heran BS, Chen JM, Ebrahim S, Moxham T, Oldridge N, Rees K et al. Exercise-based cardiac rehabilitation for coronary heart disease. Cochrane Database Syst Rev. (7):CD001800. doi:10.1002/14651858.CD001800.pub2.

75. Hunt SA, Abraham WT, Chin MH, Feldman AM, Francis GS, Ganiats TG et al. ACC/AHA 2005 Guideline Update for the Diagnosis and Management of Chronic Heart Failure in the Adult: A Report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Writing Committee to Update the 2001 Guidelines for the Evaluation and Management of Heart Failure): Developed in Collaboration With the American College of Chest Physicians and the International Society for Heart and Lung Transplantation: Endorsed by the Heart Rhythm Society. Circulation. 2005;112(12):e154-235. doi:10.1161/circulationaha.105.167586.

76. Wisloff U, Stoylen A, Loennechen JP, Bruvold M, Rognmo O, Haram PM et al. Superior Cardiovascular Effect of Aerobic Interval Training Versus Moderate Continuous Training in Heart Failure Patients: A Randomized Study. Circulation. 2007;115(24):3086-94. doi:10.1161/circulationaha.106.675041.

77. Leon AS, Franklin BA, Costa F, Balady GJ, Berra KA, Stewart KJ et al. Cardiac Rehabilitation and Secondary Prevention of Coronary Heart Disease. Circulation. 2005;111(3):369-76. doi:10.1161/01.cir.0000151788.08740.5c.

78. Ades PA, Savage PD, Toth MJ, Harvey-Berino J, Schneider DJ, Bunn JY et al. High-calorie-expenditure exercise: a new approach to cardiac rehabilitation for overweight coronary patients. Circulation. 2009;119(20):2671-8. doi:CIRCULATIONAHA.108.834184 [pii]

10.1161/CIRCULATIONAHA.108.834184.

79. Balady GJ, Williams MA, Ades PA, Bittner V, Comoss P, Foody JM et al. Core Components of Cardiac Rehabilitation/Secondary Prevention Programs: 2007 Update. Circulation. 2007;115(20):2675-82. doi:10.1161/circulationaha.106.180945.

80. Jones LW, Courneya KS, Vallance JK, Ladha AB, Mant MJ, Belch AR et al. Association between exercise and quality of life in multiple myeloma cancer survivors. Support Care Cancer. 2004;12(11):780-8. doi:10.1007/s00520-004-0668-4.

81. Antonelli J, Freedland SJ, Jones LW. Exercise therapy across the prostate cancer continuum. Prostate Cancer Prostatic Dis. 2009;12(2):110-5. doi:pcan20094 [pii]
10.1038/pcan.2009.4.

82. Haykowsky M, Mackey JR, Jones LW, Thompson R, Paterson I. Adjuvant Trastuzumab Induces Ventricular Remodeling Despite Aerobic Exercise Training. Clinical Cancer Research. 2009;in press.

83. Jones LW, Eves ND, Peterson BL, Garst J, Crawford J, West MJ et al. Safety and feasibility of aerobic training on cardiopulmonary function and quality of life in postsurgical nonsmall cell lung cancer patients : a pilot study. Cancer. 2008;113(12):3430-9. doi:10.1002/cncr.23967.

84. Jones LW, Liang Y, Pituskin EN, Battaglini CL, Scott JM, Hornsby WE et al. Effect of exercise training on peak oxygen consumption in patients with cancer: a meta-analysis. Oncologist.16(1):112-20. doi:theoncologist.2010-0197 [pii]

10.1634/theoncologist.2010-0197.

85. McNeely ML, Campbell KL, Rowe BH, Klassen TP, Mackey JR, Courneya KS. Effects of exercise on breast cancer patients and survivors: a systematic review and meta-analysis. CMAJ. 2006;175(1):34-41. doi:175/1/34 [pii] 10.1503/cmaj.051073.

86. Gulati M, Pandey DK, Arnsdorf MF, Lauderdale DS, Thisted RA, Wicklund RH et al. Exercise capacity and the risk of death in women: the St James Women Take Heart Project. Circulation. 2003;108(13):1554-9. doi:10.1161/01.CIR.0000091080.57509.E9 01.CIR.0000091080.57509.E9 [pii].

87. Wiskemann J, Huber G. Physical exercise as adjuvant therapy for patients undergoing hematopoietic stem cell transplantation. Bone Marrow Transplant. 2008;41(4):321-9. doi:1705917 [pii] 10.1038/sj.bmt.1705917.

88. Schmitz KH, Courneya KS, Matthews C, Demark-Wahnefried W, GalvÃƒO DA, Pinto BM et al. American College of Sports Medicine Roundtable on Exercise Guidelines for Cancer Survivors. Medicine & Science in Sports & Exercise.42(7):1409-26 10.249/MSS.0b013e3181e0c112.

89. Rahnama N, Nouri R, Rahmaninia F, Damirchi A, Emami H. The effects of exercise training on maximum aerobic capacity, resting heart rate, blood pressure and anthropometric variables of postmenopausal women with breast cancer. J Res Med Sci.15(2):78-83.

90. Courneya KS, Segal RJ, Mackey JR, Gelmon K, Reid RD, Friedenreich CM et al. Effects of Aerobic and Resistance Exercise in Breast Cancer Patients Receiving Adjuvant Chemotherapy: A Multicenter Randomized Controlled Trial. Journal of Clinical Oncology. 2007;25(28):4396-404. doi:10.1200/jco.2006.08.2024.

91. Galvao DA, Nosaka K, Taaffe DR, Spry N, Kristjanson LJ, McGuigan MR et al. Resistance training and reduction of treatment side effects in prostate cancer patients. Med Sci Sports Exerc. 2006;38(12):2045-52. doi:10.1249/01.mss.0000233803.48691.8b 00005768-200612000-00002 [pii].

92. Jones L, Douglas P, Eves N, Marcom PK, Kraus W, Herndon J et al. Rationale and design of the Exercise Intensity Trial (EXCITE): A randomized trial comparing the effects of moderate versus moderate to high-intensity aerobic training in women with operable breast cancer. BMC Cancer.10(1):531.

93. Wharton S, Vanderlelie S, Sharma AM, Sharma S, Kuk JL. Feasibility of an interdisciplinary program for obesity management in Canada. Can Fam Physician.58(1):e32-8. doi:58/1/e32 [pii].

94. McAlister FA, Stewart S, Ferrua S, McMurray JJ. Multidisciplinary strategies for the management of heart failure patients at high risk for admission: a systematic review of randomized trials. J Am Coll Cardiol. 2004;44(4):810-9. doi:10.1016/j.jacc.2004.05.055 S0735-1097(04)01123-4 [pii].

95. Holland R, Battersby J, Harvey I, Lenaghan E, Smith J, Hay L. Systematic review of multidisciplinary interventions in heart failure. Heart. 2005;91(7):899-906. doi:10.1136/hrt.2004.048389.

96. Inglis SC, Pearson S, Treen S, Gallasch T, Horowitz JD, Stewart S. Extending the Horizon in Chronic Heart Failure: Effects of Multidisciplinary, Home-Based Intervention Relative to Usual Care. Circulation. 2006;114(23):2466-73. doi:10.1161/circulationaha.106.638122.

97. Alberti KG, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, Donato KA et al. Harmonizing the metabolic syndrome: a joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity. Circulation. 2009;120(16):1640-5. doi:CIRCULATIONAHA.109.192644 [pii] 10.1161/CIRCULATIONAHA.109.192644.

98. Alberti KG, Zimmet PZ. Definition, diagnosis and classification of diabetes mellitus and its complications. Part 1: diagnosis and classification of diabetes mellitus provisional report of a WHO consultation. Diabet Med. 1998;15(7):539-53. doi:10.1002/(SICI)1096-9136(199807)15:7<539::AID-DIA668>3.0.CO;2-S.

99. Grundy SM, Brewer HB, Jr., Cleeman JI, Smith SC, Jr., Lenfant C. Definition of metabolic syndrome: report of the National Heart, Lung, and Blood Institute/American Heart Association conference on scientific issues related to

definition. Arterioscler Thromb Vasc Biol. 2004;24(2):e13-8. doi:10.1161/01.ATV.0000111245.75752.C6 24/2/e13 [pii].

100. Einhorn D, Reaven GM, Cobin RH, Ford E, Ganda OP, Handelsman Y et al. American College of Endocrinology position statement on the insulin resistance syndrome. Endocr Pract. 2003;9(3):237-52.

101. Executive Summary of The Third Report of The National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, And Treatment of High Blood Cholesterol In Adults (Adult Treatment Panel III). JAMA. 2001;285(19):2486-97. doi:jsc10094 [pii].

102. Balkau B, Charles MA. Comment on the provisional report from the WHO consultation. European Group for the Study of Insulin Resistance (EGIR). Diabet Med. 1999;16(5):442-3.

APPENDIX C

Additional PhD work

RATIONALE AND DESIGN OF THE 'MULTIDISCIPLINARY APPROACH TO NOVEL THERAPIES IN CARDIOLOGY ONCOLOGY RESEARCH' TRIAL (MANTICORE 101 – BREAST): A RANDOMIZED PLACEBO-CONTROLLED TRIAL TO DETERMINE IF CONVENTIONAL HEART FAILURE PHARMACOTHERAPY CAN PREVENT TRASTUZUMAB-MEDIATED LEFT VENTRICULAR REMODLING IN PATIENTS WITH HER2+ EARLY BREAST CANCER USING CARDIAC MRI

(A version of this chapter has been published as: Pituskin E, Haykowsky M, Mackey JR, Thompson RB, Ezekowitz J, Koshman S, Oudit G, Chow K, Pagano JJ, & Paterson I.

BMC Cancer 2011, 11**:**318)

C.1 Background

Breast cancer is the most common malignancy and second leading cause of cancer death(1). Approximately 20-25% of breast cancers over-express human epidermal growth factor receptor 2 (HER2+) which is associated with poor prognosis(2, 3). Trastuzumab (Herceptin®), a humanized monoclonal antibody targeting the HER2 receptor, was previously shown to improve survival by 20% in women with HER2+metastatic disease(4-7). More recently, 4 major adjuvant trials of women with HER2+ early breast cancer (EBC) demonstrated that trastuzumab reduced 3-year breast cancer recurrence and risk of death rate by 50%(8). Given these positive findings, trastuzumab was approved in 2006 by the Food and Drug Administration for the adjuvant treatment of HER2+ breast cancer.

Despite favourable survival benefits, an adverse effect of trastuzumab is (a)symptomatic left ventricular (LV) dysfunction and heart failure (HF). In the phase III trials, HF and asymptomatic LV dysfunction was reported in 4% and 18% of patients, respectively(9-11). Although trastuzumab-related cardiotoxicity has been considered 'reversible'(12), Wadhwa et al. reported that trastuzumab was stopped in 22% of patients due to asymptomatic LV systolic dysfunction; notably, of these, 40% showed no improvement or worsening of LV function over time despite optimal pharmacotherapy(13). Similarly, Chia observed that 21.6% of women receiving adjuvant trastuzumab-based chemotherapy experienced a cardiac event requiring temporary or permanent discontinuation of trastuzumab(14). These observations are important, given the influence of more

stringent cardiac exclusion criteria in the pivotal trials compared to standard clinical practice. Furthermore, any dose reductions, delay or discontinuation due to cardiotoxicity are potentially life-threatening events from the competing risks of cancer and/or cardiac mortality. Therefore, better understanding of the pathophysiology of trastuzumab-mediated cardiotoxicity and its prevention are urgently required.

Ventricular remodeling (increased cavity size and decreased pump function) precedes overt HF(15-17). Our group has shown that aerobic exercise training has beneficial anti-remodeling benefits in clinically stable systolic HF patients(18). Based on these findings, we examined the effect of 4 months of aerobic exercise training on LV remodeling in 17 women with EBC receiving trastuzumab-based chemotherapy(19). We found that LV remodeling occurs early, confirming observations of other groups(14, 20), and that an early exercise intervention did not attenuate remodeling in this setting. Accordingly, we identified the need for examination of non-exercise interventions.

Pharmacotherapy has been shown to attenuate or reverse LV remodeling in the HF and post-myocardial infarction (MI) setting. Angiotensin-converting enzyme inhibitors (ACEI) have been proven to delay or reverse LV dilation and improve ejection fraction (EF) in multiple trials(21-24). Beta-blockers (BB) have also been shown to be beneficial, but have largely been tested in combination with other therapies(25, 26). To date, a paucity of studies have examined conventional HF therapy during anthracycline therapy(27-29). Specifically, carvedilol has been shown to be an effective single-agent therapy in anthracycline-induced

cardiomyopathy(30).Cardinale *et al* demonstrated that an ACEI can prevent a decline in EF and cardiac events in cancer patients receiving high dose anthracyclines(31). In general, however, preventive medical therapy is not considered necessary with anthracycline-based regimens, as toxicity is related to the cumulative dose (> 500mg/m^{2}) (32).

Detection and measurement of LV dysfunction may be hampered by insensitivity of routinely-available imaging modalities. Most EBC clinical trials have employed either radionuclide ventriculography (eg. MUGA) or transthoracic echocardiograms (ECHO) which may underestimate LV volumes(33). Cardiac MRI is the preferred method for the quantification of ventricular volumes and EF in individuals with impaired LV systolic function(34-36). Unfortunately, relying on a decline in EF or overt clinical symptoms limits detection of cardiotoxicity to the late stage of disease process, increasing the likelihood of irreversible myocardial damage.

In addition to detailed imaging, cardiac biomarkers may contribute to early detection, assessment and longer-term monitoring of cardiotoxicity. In patients with multiple types of advanced cancer receiving high-dose chemotherapy, Cardinale and associates demonstrated that troponin I increased in one-third of patients shortly after treatment, which was associated with a concomitant reduction in EF within the subsequent year(31). In serial evaluation of adjuvant and metastatic patients receiving trastuzumab-based chemotherapy, elevations in troponin were seen early in therapy (after 2 cycles) and with adjustment for major confounders, was the strongest independent predictor of

future EF decline, occurring within 1 – 8 months(37). In these patients (n = 42), a three-fold decrease in likelihood of recovery was observed over time despite optimal pharmacotherapy. Brain naturietic peptide (BNP), an established marker of heart failure, has also been shown to predict cardiac events including symptomatic heart failure, arrhythmias and acute coronary syndrome in patients receiving anthracycline-based chemotherapy(38). While these studies provide valuable insights, larger studies in homogeneous patient groups with similar chemotherapy regimens and biomarker collection are required before general recommendations can be implemented(39). Finally, other novel markers may have the potential to detect early cardiac dysfunction. For example, collagen-derived peptides have been correlated to the fractional volume of myocardial fibrosis in hypertensive patients(40)and HF(41). As the underlying collagen-based structure of the heart remodels, peptides are released into the plasma (42-44). While these molecules have not been studied in cancer patients(45), prospective exploration of these markers and other prospectively-collected biofluids may provide insight into disease evolution or effects of pharmacotherapy.

To date, no study has prospectively examined the effects of proven HF therapy to prevent cardiotoxicity in patients with EBC receiving trastuzumab. The median follow-up of the pivotal trastuzumab trials remains between two and three years, limiting understanding of the clinical course of patients with trastuzumab mediated cardiac toxicity and thus, the appropriate length of therapy. Therefore, the logical extension of our body of research is to examine: (1) the underlying pathogenesis of trastuzumab mediated cardiac toxicity, (2) screening tools for

detecting early LV remodeling in this patient group, (3) determine the evolution of LV remodeling beyond chemotherapy, and (4) pharmacotherapies to prevent LV remodeling for these patients.

Given this background, our cardio-oncology research group designed the *Multidisciplinary Approach to Novel Therapies In Cardiology Oncology Research (MANTICORE) trial.* The primary objective of this randomized clinical trial is to determine if conventional heart failure pharmacotherapy (ACEI or BB) can prevent trastuzumab-mediated LV remodeling among patients with HER2+ EBC, determined by 12 month change in LV end-diastolic volume (LVEDV) measured using cardiac MRI. Secondary objectives include 1) determine the evolution of LV remodeling on cardiac MRI in patients with HER2+ EBC; 2) understand the mechanism of trastuzumab mediated cardiac toxicity by assessing for the presence of myocardial injury and apoptosis using serum biomarkers and cardiac MRI; and 3) correlate cardiac biomarkers of myocyte injury and extra-cellular matrix remodeling with LV remodeling on cardiac MRI.

C2 Methods

C2.1 Design

This study is parallel 3-arm, randomized, placebo controlled, double-blind study comparing bisoprolol and perindopril for the prevention of LV remodeling in patients with HER2+ breast cancer treated with adjuvant trastuzumab. Additional inclusion requirements are adequate creatinine clearance, age > 18 years, no contraindication to MRI and willingness to provide informed consent. Exclusion criteria are known contraindication or current treatment with ACEI or

BB; history of heart failure, cardiomyopathy, baseline EF < 50%; and history of uncontrolled hypertension or myocardial infarction. Ethical approval has been secured from the two relevant institutional review Boards.

C2.2 Participants

Potential participants are consecutively identified and screened for eligibility by the study coordinator during multidisciplinary Tumor Board review of all new breast cancer case consultations. Following primary oncologist approval, potential participants are provided with a review of the study and provided with written study information. After obtaining written consent, participants will be scheduled for a baseline clinic visit for final determination of eligibility.

C2.3 Group Allocation

Participants are randomized in a 1:1:1 ratio to perindopril, bisoprolol, or placebo using a secure internet randomization service (EPICORE, www.epicore.ualberta.ca).

C2.4 Pharmacotherapy

Participants will receive drug or placebo for 1 year beginning 7 days before trastuzumab therapy. There will be no run-in period. Dosages for all groups will be systematically up-titrated as tolerated at 1 week intervals, for a total of 3 weeks. Perindopril will be initiated at 2mg and titrated to a target dose of 8mg. Bisoprolol will be initiated at 2.5mg and titrated to a 10mg target dose.

C2.5 Evaluations

Baseline demographics, medical history, cardiac risk factors, and cardiac history are evaluated. Physical examination including cardiovascular exam, blood pressure and pulse is performed. Blood is collected for the purposes of measuring baseline electrolytes, creatinine (for estimation of glomerular filtration rate [GFR]), and biomarkers and an electrocardiogram. Participants undergo a comprehensive baseline cardiac MRI as part of their routine pre-chemotherapy ventricular function evaluation in lieu of conventional modalities. An additional consent is obtained for prospective serial collection of serum and urine in the Alberta Research Tumor Bank (www.abtumorbank.com) for future analysis.

Participants initiate study drug one week prior to starting trastuzumab therapy and are scheduled for titration visits. Participants return for a visit with the study coordinator at 3 and 12 months after randomization to be assessed for signs and symptoms of HF, major adverse cardiac events (MACE), adverse effects relating to HF pharmacotherapy as well as adherence to medication. In addition, participants will be contacted by telephone at months 1, 6, 9 and 18 to ensure continued stability and minimize loss to follow-up. Participants will also undergo blood work to assess electrolytes and creatinine at 3 month intervals as per routine care while on trastuzumab therapy and have biomarkers drawn at 3 and 12 months for comparison with the concurrent cardiac MRI measures. Cardiac monitoring at 6 and 9 month intervals will be performed with routinely available modalities per standard of care. If trastuzumab therapy is discontinued due to cardiac toxicity outside of the 3 or 12 month cardiac MRI scans then

participants will return for another clinically indicated cardiac MRI within 1 week of chemotherapy termination.

Participants will return for a final visit with the study coordinator at 24 months, or 1 year after trastuzumab therapy termination. The study coordinator will perform the same clinical assessment as on previous follow-up visits at 3 and 12 months. Evaluation of medications will be done to ascertain if treatment for HF has been initiated since the 12 month visit. Patients will undergo the same collection of serum biomarkers. A non-clinically indicated research cardiac MRI will be performed to assess for the presence of ongoing LV remodeling. The long-term cardiac implications of trastuzumab therapy are unknown but we hypothesize that the exposed heart will be more vulnerable to future insults[49].

C2.6 Data Security

De-identifying case report form development, data entry, quality assurance and data analysis will be performed by EPICORE. De-identified image data from cardiac MRIs will be stored on a secure network using unique identifiers. Blood samples for biomarker analysis will also only be identified by unique identifiers and stored for batch analysis.

C2.7 Statistical Considerations

Participants will be analyzed with the intention to treat principle. The primary outcome analysis will be the difference in mean change in LVEDV at 1 year in each active treatment arm and the placebo arm, evaluated using a general linear model controlling for differences in patient characteristics at the baseline and 12 month assessments. A general linear mixed model will be used to assess

group differences with serial correlation over time based on the baseline, 3, 12, and 24 month measurements with correlated post-hoc analysis where appropriate. All statistical tests will be two-sided. Regression analyses and diagnostic tests will be performed to model changes in continuous variables from imaging with those from biomarker measures. Statistical significance will be assumed at the 5% level ($p < 0.05$). Missing data will be interpolated where possible using multivariable imputation. Data will be analyzed using SAS 9.2 (SAS, Cary, NC) software.

Based on our pilot data(19), we expect the placebo group will have a minimum change in LVEDV of +11% with 1-year of trastuzumab therapy. We assume that pharmacotherapy with ACEI would prevent 90% of the trastuzumab mediated remodeling as measured by a change in LVEDV at 1 year = +1%. ACEI and perindopril have been shown to entirely prevent LV remodeling in other models of heart failure. Note that the change in LV volume in our pilot study was observed after 4 months of trastuzumab; hence we are possibly underestimating the true magnitude of change in LV volume at 1 year in the placebo group of this study. This is supported by an interim analysis of the BCIRG 006 trial, showing that mean LV function tended to decrease out to 8 months regardless of the chemotherapy protocol(11). If the within-group standard deviation is 20 ml, with a two-2-tailed significance level $\propto = 0.05$ and a power of .80, then 47 participants are required for each group. We anticipate a 10% drop-out rate and a 3% mortality rate during our 2 year follow-up(46), therefore we will recruit a total 159 participants. If the absolute difference between the mean

LVEDV in placebo and ACE inhibitor groups is greater than 10% at 1 year, this sample size will still be sufficiently powered to detect a lower degree of remodeling prevention (Table 1).

C3 Discussion

Given our small sample size, we do not expect to find a significant decrease in cardiac clinical outcomes, however, given the overwhelming evidence of clinical efficacy in the HF literature, we expect to demonstrate a beneficial effect of pharmacotherapy in preventing trastuzumab-mediated LV remodeling. Our results would then justify a multicenter trial studying the role of ACEI and/or BB for preventing clinical HF and improving long-term outcomes of breast cancer survivors. Furthermore, if ACEI or BB are proven efficacious in the prevention of cardiac toxicity, then patients with known cardiac dysfunction (thus ineligible to receive trastuzumab) could also be studied. Effective strategies for preventing trastuzumab mediated cardiac toxicity would allow more HER2+ breast cancer patients to successfully complete their adjuvant chemotherapy, while avoiding delays or discontinuation of vital therapy.

Our secondary objectives may also yield information on appropriate biomarkers to screen for cardiac toxicity and provide additional information on the underlying pathogenesis and the evolution of cardiac injury beyond exposure to trastuzumab. Ideal biomarkers in this field have yet to be identified, however, cardiac MRI allows for precise characterization of LV dysfunction against which biomarkers can be evaluated. In future studies, we will analyze prospectively collected biofluids in the study of metabolics and genomics of trastuzumab-

related cardiotoxicity. Urine metabolomics have the capacity to sensitively measure metabolic processes at the cellular level, and may provide early insights into myocyte structural or energetic changes (47, 48). Molecular profiling may identify key pathways involved in tissue regeneration, drug metabolism and cell death (49, 50); in studying copy number variants, we have shown that toxicities associated with docetaxel-containing chemotherapy regimens may be predicted(51). Taken together, study of biomarkers and clinical outcomes in this setting will contribute to personalized care and improved therapeutic outcomes.

C4 Conclusion

Cardiac toxicity as a result of cancer therapies is now recognized as a significant health problem of increasing prevalence. While therapies such as trastuzumab potentially offer improved survival with targeted anti-tumor activities, the additional cardiac insult in addition to 'multiple-hit'(52) injuries put patients at risk of morbidity and potentially premature mortality. A limitation of the existing data is that the median follow-up in the trastuzumab adjuvant trials is between 2 and 3 years. Therefore, information is lacking on the potential for late cardiac dysfunction, or whether short-term improvements in LV remodeling or EF with medical treatment are permanent or temporary. Future trials with longer follow-up are necessary to address these remaining issues.

To our knowledge, MANTICORE will be the first randomized trial testing proven HF pharmacotherapy in the prevention of trastuzumab-mediated cardiotoxicity. We expect the findings of this trial to provide high-level evidence in the development of guidelines for preventive therapy. Relationships of

established and novel biomarkers to sensitive MRI measures will enhance our understanding of these markers, as well as development of sensitive assays for the earliest detection of cardiovascular changes in these patients. Lastly, this work will inform and support other long-term goals of our team, in the prevention, detection and treatment of cardiovascular effects of cancer therapies.

C5 References

1. Canadian Cancer Statistics. Toronto, Canada2010.

2. Slamon DJ, Clark GM, Wong SG, Levin WJ, Ullrich A, McGuire WL. Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. Science. 1987 Jan 9;235(4785):177-82. PubMed PMID: 3798106. Epub 1987/01/09. eng.

3. Slamon DJ, Godolphin W, Jones LA, Holt JA, Wong SG, Keith DE, et al. Studies of the HER-2/neu proto-oncogene in human breast and ovarian cancer. Science. 1989 May 12;244(4905):707-12. PubMed PMID: 2470152. Epub 1989/05/12. eng.

4. Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, Bajamonde A, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. N Engl J Med. 2001 Mar 15;344(11):783-92. PubMed PMID: 11248153. Epub 2001/03/15. eng.

5. Vogel CL, Cobleigh MA, Tripathy D, Gutheil JC, Harris LN, Fehrenbacher L, et al. Efficacy and safety of trastuzumab as a single agent in first-line treatment of HER2-overexpressing metastatic breast cancer. J Clin Oncol. 2002 Feb 1;20(3):719-26. PubMed PMID: 11821453.

6. Baselga J, Carbonell X, Castaneda-Soto NJ, Clemens M, Green M, Harvey V, et al. Phase II study of efficacy, safety, and pharmacokinetics of trastuzumab monotherapy administered on a 3-weekly schedule. J Clin Oncol. 2005 Apr 1;23(10):2162-71. PubMed PMID: 15800309. Epub 2005/04/01. eng.

7. Marty M, Cognetti F, Maraninchi D, Snyder R, Mauriac L, Tubiana-Hulin M, et al. Randomized phase II trial of the efficacy and safety of trastuzumab combined with docetaxel in patients with human epidermal growth factor receptor 2-positive metastatic breast cancer administered as first-line treatment: the M77001 study group. J Clin Oncol. 2005 Jul 1;23(19):4265-74. PubMed PMID: 15911866. Epub 2005/05/25. eng.

8. Baselga J, Perez EA, Pienkowski T, Bell R. Adjuvant trastuzumab: a milestone in the treatment of HER-2-positive early breast cancer. Oncologist. 2006;11 Suppl 1:4-12. PubMed PMID: 16971734. Epub 2006/09/15. eng.

9. Romond EH, Perez EA, Bryant J, Suman VJ, Geyer CE, Jr., Davidson NE, et al. Trastuzumab plus adjuvant chemotherapy for operable HER2-positive breast cancer. N Engl J Med. 2005 Oct 20;353(16):1673-84. PubMed PMID: 16236738. Epub 2005/10/21. eng.

10. Suter TM, Procter M, van Veldhuisen DJ, Muscholl M, Bergh J, Carlomagno C, et al. Trastuzumab-associated cardiac adverse effects in the herceptin adjuvant trial. J Clin Oncol. 2007 Sep 1;25(25):3859-65. PubMed PMID: 17646669. Epub 2007/07/25. eng.

11. Slamon DJ, Robert N, al. e, editors. Phase III trial comparing AC-T with AC-TH and with TCH in the adjuvant treatment of HER2 positive early breast cancer patients: a second interim efficacy analysis. . San Antonio Breast Cancer Symposium; 2006; San Antonio, TX. .

12. Ewer MS, Vooletich MT, Durand J-B, Woods ML, Davis JR, Valero V, et al. Reversibility of Trastuzumab-Related Cardiotoxicity: New Insights Based on

Clinical Course and Response to Medical Treatment. J Clin Oncol. 2005 November 1, 2005;23(31):7820-6.

13. Wadhwa D, Fallah-Rad N, Grenier D, Krahn M, Fang T, Ahmadie R, et al. Trastuzumab mediated cardiotoxicity in the setting of adjuvant chemotherapy for breast cancer: a retrospective study. Breast Cancer Res Treat. 2009 Sep;117(2):357-64. PubMed PMID: 19082707.

14. McArthur HL, Chia S. Cardiotoxicity of trastuzumab in clinical practice. N Engl J Med. 2007 Jul 5;357(1):94-5. PubMed PMID: 17611218. Epub 2007/07/06. eng.

15. Cohn JN, Ferrari R, Sharpe N. Cardiac remodeling--concepts and clinical implications: a consensus paper from an international forum on cardiac remodeling. Behalf of an International Forum on Cardiac Remodeling. J Am Coll Cardiol. 2000 Mar 1;35(3):569-82. PubMed PMID: 10716457. Epub 2000/03/15. eng.

16. Mann DL, Bristow MR. Mechanisms and Models in Heart Failure: The Biomechanical Model and Beyond. Circulation. 2005 May 31, 2005;111(21):2837-49.

17. Vasan RS, Larson MG, Benjamin EJ, Evans JC, Levy D. Left ventricular dilatation and the risk of congestive heart failure in people without myocardial infarction. N Engl J Med. 1997 May 8;336(19):1350-5. PubMed PMID: 9134875. Epub 1997/05/08. eng.

18. Haykowsky MJ, Liang Y, Pechter D, Jones LW, McAlister FA, Clark AM. A meta-analysis of the effect of exercise training on left ventricular

remodeling in heart failure patients: the benefit depends on the type of training performed. J Am Coll Cardiol. 2007 Jun 19;49(24):2329-36. PubMed PMID: 17572248. Epub 2007/06/19. eng.

19. Haykowsky MJ, Mackey JR, Thompson RB, Jones LW, Paterson DI. Adjuvant Trastuzumab Induces Ventricular Remodeling Despite Aerobic Exercise Training. Clin Cancer Res. 2009 August 1, 2009;15(15):4963-7.

20. Telli ML, Hunt SA, Carlson RW, Guardino AE. Trastuzumab-Related Cardiotoxicity: Calling Into Question the Concept of Reversibility. J Clin Oncol. 2007 August 10, 2007;25(23):3525-33.

21. St John Sutton M, Pfeffer MA, Plappert T, Rouleau JL, Moye LA, Dagenais GR, et al. Quantitative two-dimensional echocardiographic measurements are major predictors of adverse cardiovascular events after acute myocardial infarction. The protective effects of captopril. Circulation. 1994 Jan;89(1):68-75. PubMed PMID: 8281697. Epub 1994/01/01. eng.

22. Greenberg B, Quinones MA, Koilpillai C, Limacher M, Shindler D, Benedict C, et al. Effects of long-term enalapril therapy on cardiac structure and function in patients with left ventricular dysfunction. Results of the SOLVD echocardiography substudy. Circulation. 1995 May 15;91(10):2573-81. PubMed PMID: 7743619. Epub 1995/05/15. eng.

23. Ferrari R. Effects of angiotensin-converting enzyme inhibition with perindopril on left ventricular remodeling and clinical outcome: results of the randomized Perindopril and Remodeling in Elderly with Acute Myocardial

Infarction (PREAMI) Study. Arch Intern Med. 2006 Mar 27;166(6):659-66. PubMed PMID: 16567606. Epub 2006/03/29. eng.

24. Abdulla J, Barlera S, Latini R, Kjoller-Hansen L, Sogaard P, Christensen E, et al. A systematic review: effect of angiotensin converting enzyme inhibition on left ventricular volumes and ejection fraction in patients with a myocardial infarction and in patients with left ventricular dysfunction. Eur J Heart Fail. 2007 Feb;9(2):129-35. PubMed PMID: 16829187. Epub 2006/07/11. eng.

25. Randomised, placebo-controlled trial of carvedilol in patients with congestive heart failure due to ischaemic heart disease. Australia/New Zealand Heart Failure Research Collaborative Group. Lancet. 1997 Feb 8;349(9049):375-80. PubMed PMID: 9033462. Epub 1997/02/08. eng.

26. Bellenger NG, Rajappan K, Rahman SL, Lahiri A, Raval U, Webster J, et al. Effects of carvedilol on left ventricular remodelling in chronic stable heart failure: a cardiovascular magnetic resonance study. Heart. 2004 Jul;90(7):760-4. PubMed PMID: 15201244. Epub 2004/06/18. eng.

27. Kalay N, Basar E, Ozdogru I, Er O, Cetinkaya Y, Dogan A, et al. Protective Effects of Carvedilol Against Anthracycline-Induced Cardiomyopathy. Journal of the American College of Cardiology. 2006;48(11):2258-62.

28. Cardinale D, Lamantia G, Cipolla CM. Troponin I and cardiovascular risk stratification in patients with testicular cancer. J Clin Oncol. 2006 Jul 20;24(21):3508; author reply -9. PubMed PMID: 16849772. Epub 2006/07/20. eng.

29. Lipshultz SE, Lipsitz SR, Sallan SE, Simbre VC, 2nd, Shaikh SL, Mone SM, et al. Long-term enalapril therapy for left ventricular dysfunction in doxorubicin-treated survivors of childhood cancer. J Clin Oncol. 2002 Dec 1;20(23):4517-22. PubMed PMID: 12454107. Epub 2002/11/28. eng.

30. Kalay N, Basar E, Ozdogru I, Er O, Cetinkaya Y, Dogan A, et al. Protective effects of carvedilol against anthracycline-induced cardiomyopathy. J Am Coll Cardiol. 2006 Dec 5;48(11):2258-62. PubMed PMID: 17161256. Epub 2006/12/13. eng.

31. Cardinale D, Sandri MT, Martinoni A, Tricca A, Civelli M, Lamantia G, et al. Left ventricular dysfunction predicted by early troponin I release after high-dose chemotherapy. J Am Coll Cardiol. 2000 Aug;36(2):517-22. PubMed PMID: 10933366. Epub 2000/08/10. eng.

32. Lefrak EA, Pitha J, Rosenheim S, Gottlieb JA. A clinicopathologic analysis of adriamycin cardiotoxicity. Cancer. 1973 Aug;32(2):302-14. PubMed PMID: 4353012. Epub 1973/08/01. eng.

33. Jenkins C, Moir S, Chan J, Rakhit D, Haluska B, Marwick TH. Left ventricular volume measurement with echocardiography: a comparison of left ventricular opacification, three-dimensional echocardiography, or both with magnetic resonance imaging. Eur Heart J. 2009 Jan;30(1):98-106. PubMed PMID: 18997179. Epub 2008/11/11. eng.

34. Bellenger N, Pennell D. Magnetic resonance imaging in cardiology. J R Coll Physicians Lond. 1999 Jan-Feb;33(1):12-8. PubMed PMID: 10192063. Epub 1999/04/07. eng.

35. Bellenger NG, Davies LC, Francis JM, Coats AJ, Pennell DJ. Reduction in sample size for studies of remodeling in heart failure by the use of cardiovascular magnetic resonance. Journal of cardiovascular magnetic resonance : official journal of the Society for Cardiovascular Magnetic Resonance. 2000;2(4):271-8. PubMed PMID: 11545126. Epub 2001/09/08. eng.

36. Bellenger NG, Burgess MI, Ray SG, Lahiri A, Coats AJ, Cleland JG, et al. Comparison of left ventricular ejection fraction and volumes in heart failure by echocardiography, radionuclide ventriculography and cardiovascular magnetic resonance; are they interchangeable? Eur Heart J. 2000 Aug;21(16):1387-96. PubMed PMID: 10952828. Epub 2000/08/23. eng.

37. Cardinale D, Colombo A, Torrisi R, Sandri MT, Civelli M, Salvatici M, et al. Trastuzumab-induced cardiotoxicity: clinical and prognostic implications of troponin I evaluation. J Clin Oncol. Sep 1;28(25):3910-6. PubMed PMID: 20679614. Epub 2010/08/04. eng.

38. Lenihan DJ MM, Baysinger KB, et al., editor Superior detection of cardiotoxicity during chemotherapy using biomarkers. . Heart Failure Society of America; 2007 September 17, 2007; Washington, DC.

39. Cardinale D, Sandri MT. Role of biomarkers in chemotherapy-induced cardiotoxicity. Progress in cardiovascular diseases. Sep-Oct;53(2):121-9. PubMed PMID: 20728699. Epub 2010/08/24. eng.

40. Querejeta R, Varo N, Lopez B, Larman M, Artinano E, Etayo JC, et al. Serum carboxy-terminal propeptide of procollagen type I is a marker of

myocardial fibrosis in hypertensive heart disease. Circulation. 2000 Apr 11;101(14):1729-35. PubMed PMID: 10758057. Epub 2000/04/12. eng.

41. Braunwald E. Biomarkers in Heart Failure. N Engl J Med. 2008 May 15, 2008;358(20):2148-59.

42. Cicoira M, Rossi A, Bonapace S, Zanolla L, Golia G, Franceschini L, et al. Independent and additional prognostic value of aminoterminal propeptide of type III procollagen circulating levels in patients with chronic heart failure. J Card Fail. 2004 Oct;10(5):403-11. PubMed PMID: 15470651. Epub 2004/10/08. eng.

43. Zannad F, Alla F, Dousset B, Perez A, Pitt B. Limitation of excessive extracellular matrix turnover may contribute to survival benefit of spironolactone therapy in patients with congestive heart failure: insights from the randomized aldactone evaluation study (RALES). Rales Investigators. Circulation. 2000 Nov 28;102(22):2700-6. PubMed PMID: 11094035. Epub 2000/11/30. eng.

44. Iraqi W, Rossignol P, Angioi M, Fay R, Nuee J, Ketelslegers JM, et al. Extracellular cardiac matrix biomarkers in patients with acute myocardial infarction complicated by left ventricular dysfunction and heart failure: insights from the Eplerenone Post-Acute Myocardial Infarction Heart Failure Efficacy and Survival Study (EPHESUS) study. Circulation. 2009 May 12;119(18):2471-9. PubMed PMID: 19398668. Epub 2009/04/29. eng.

45. Lopez B, Gonzalez A, Diez J. Circulating biomarkers of collagen metabolism in cardiac diseases. Circulation. Apr 13;121(14):1645-54. PubMed PMID: 20385961. Epub 2010/04/14. eng.

46. Smith I, Procter M, Gelber RD, Guillaume S, Feyereislova A, Dowsett M, et al. 2-year follow-up of trastuzumab after adjuvant chemotherapy in HER2-positive breast cancer: a randomised controlled trial. Lancet. 2007 Jan 6;369(9555):29-36. PubMed PMID: 17208639. Epub 2007/01/09. eng.

47. Mazurek S, Eigenbrodt E. The tumor metabolome. Anticancer Res. 2003 Mar-Apr;23(2A):1149-54. PubMed PMID: 12820363. Epub 2003/06/25. eng.

48. Slupsky CM, Steed H, Wells TH, Dabbs K, Schepansky A, Capstick V, et al. Urine metabolite analysis offers potential early diagnosis of ovarian and breast cancers. Clin Cancer Res. Dec 1;16(23):5835-41. PubMed PMID: 20956617. Epub 2010/10/20. eng.

49. Samuel J-L, Schaub MC, Zaugg M, Mamas M, Dunn WB, Swynghedauw B. Genomics in cardiac metabolism. Cardiovasc Res. 2008 July 15, 2008;79(2):218-27.

50. Marziliano N, Grasso M, Pilotto A, Porcu E, Tagliani M, Disabella E, et al. Transcriptomic and proteomic analysis in the cardiovascular setting: unravelling the disease? J Cardiovasc Med (Hagerstown). 2009 May;10(5):433-42. PubMed PMID: 19449458. Epub 2009/05/19. eng.

51. Damaraju S SB, Ghosh S, Pituskin E, Tuszynski J, Cass CE, Mackey JR., editor Germline copy number polymorphisms associated with toxicity from adjuvant docetaxel. San Antonio Breast Cancer Symposium; 2010 December 8-12, 2010; San Antonio TX.

52. Jones LW, Haykowsky MJ, Swartz JJ, Douglas PS, Mackey JR. Early breast cancer therapy and cardiovascular injury. J Am Coll Cardiol. 2007 Oct 9;50(15):1435-41. PubMed PMID: 17919562. Epub 2007/10/09. eng.