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Cardiac dysfunction in cancer survivors unmasked during exercise

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Abstract:

Introduction: The cardiac dysfunction associated with anthracycline-based chemotherapy cancer treatment can exist sub-clinically for decades before overt presentation. Stress echocardiography, the measurement of left ventricular (LV) deformation and arterial haemodynamic evaluation have separately been used to identify sub-clinical cardiovascular (CV) dysfunction in several patient groups including those with hypertension and diabetes. The purpose of the present cross-sectional study was to determine whether the combination of these techniques could be used to improve the characterisation of sub-clinical CV dysfunction in long-term cancer survivors previously treated with anthracyclines.

Materials and methods: Thirteen long-term cancer survivors (36±10 years) with prior anthracycline exposure (11±8 years post-treatment) and 13 age-matched controls were recruited. Left ventricular structure, function and deformation were assessed using echocardiography. Augmentation index was used to quantify arterial haemodynamic load and

was measured using applanation tonometry. Measurements were taken at rest and during two stages of low-intensity incremental cycling.

Results: At rest, both groups had comparable global LV systolic, diastolic and arterial function (all $P>0.05$), however longitudinal deformation was significantly lower in cancer survivors (-18±2 v -20±2, $P<0.05$). During exercise this difference between groups persisted and further differences were uncovered with significantly lower apical circumferential deformation in the cancer survivors (-24±5 v -29±5, -29±5 v 35±8 for first and second stage of exercise respectively, both $P<0.05$).

Conclusion: In contrast to resting echocardiography the measurement of LV deformation at rest and during exercise provides a more comprehensive characterisation of sub-clinical LV dysfunction. Larger studies are required to determine the clinical relevance of these preliminary findings.

Key Words: Anthracyclines; exercise echocardiography; cardiac deformation; arterial haemodynamics

Introduction: Despite the survival benefits of anthracycline-based chemotherapy in the treatment of cancer, these drugs are known to have a dose-dependent toxic effect on the heart [1]. Indeed, cancer survivors previously exposed to anthracyclines are at greater risk of developing cardiovascular (CV) disease than from recurrent cancer [2]. Anthracycline CV toxicity is progressive in nature and may persist sub-clinically for many years prior to the presentation of overt dysfunction [1]. Recent reviews examining cancer therapeutics-related cardiac dysfunction (CTRCD) have suggested that stress (both dobutamine and exercise)

echocardiography may be useful in the characterisation of sub-clinical LV dysfunction [3,4]. Latent LV dysfunction, otherwise disguised at rest, has been successfully uncovered during exercise in other patient groups (e.g. hypertension [5] and diabetes [6]). However, stress echocardiographic studies in the oncology setting have provided contradictory and inconclusive findings [3,7,8,9,10]. This lack of clarity may be explained by the use of global measures of LV function such as the E/A ratio [7], fractional shortening [8] or cardiac index [10] which may be insensitive to sub-clinical dysfunction. More recently, the measurement of LV myocardial deformation has shown potential in the detection of sub-clinical changes to LV function in cancer patients and also in the prediction of CTRCD [3,11]. Whilst promising, these studies were carried out at rest and did not assess arterial function, which is integral to the CV response to exercise and is also susceptible to anthracycline toxicity [12]. It is possible that the combination of stress echocardiography including myocardial deformation with sensitive markers of arterial function may help thoroughly characterise sub-clinical CV dysfunction in long-term cancer survivors. Therefore, the purpose of this study was to test the hypothesis that the concurrent assessment of cardiac deformation and arterial function during exercise would improve the characterisation of sub-clinical CV dysfunction in asymptomatic cancer survivors with prior anthracycline exposure compared to assessments taken at rest.

Methods and materials:

Study population: Thirteen asymptomatic cancer survivors (age 36 ±10 years, 10 male, 3 female, with clinically recorded EF >50%) who had previously undergone anthracycline-based chemotherapy and 13 age- and gender-matched healthy participants were recruited between January 2012 and August 2014 for this cross-sectional study (Table 1). The control participants for the study were recruited from the University teaching and post-graduate

population. Cancer diagnoses and patient treatment details are presented in Table 2. Prior to enrolment in the study, a resting electrocardiogram (ECG) and echocardiogram was obtained from all cancer survivors and assessed by the study cardiologist. Exclusion criteria were: overt cardiac pathology evident on echocardiogram or ECG, atrial fibrillation, use of cardiac medications, pregnancy, uncontrolled hypertension, severe diabetic neuropathy and/or retinopathy, renal failure and any orthopaedic conditions that would prohibit exercise. The study conformed to the principles outlined in the Declaration of Helsinki. Informed consent was provided by all participants and the study was approved by the South East Wales Research Ethics Committee.

Experimental protocol: Participants attended the laboratory at Cardiff Metropolitan University on 3 occasions separated by at least 24-hours. Anthropometric measurements and a sub-maximal exercise test conducted on an upright cycle ergometer (Corival, Lode BV Medical Technology, Groningen, Netherlands) were completed in visits 1 and 2 respectively. The sub-maximal exercise test employed a ramp protocol and was stopped once participants had exceeded a respiratory exchange ratio of 1.0 [13]. Post-hoc the V-slope method [14] was applied to the sub-maximal gas exchange data (Oxycon Pro, Erich Jaeger GmbH, Hoechberg, Germany) to estimate the power output (w) associated with the individual anaerobic threshold (AT). It is acknowledged that AT determined from sub-maximal exercise test data will not be comparable to that obtained from a maximal test. However, the purpose of the present exercise test was to standardise the sub-maximal exercise intensity during the third laboratory visit. The exercise protocol in visit 3 involved 2 stages of incremental exercise on a supine cycle ergometer (Lode, Angio 2003, Groningen, Netherlands). The exercise intensities for each participant were determined by firstly correcting the upright power output for the supine position by deducting 20% [15], and then calculating 25% (exercise stage 1; Ex1) and 50%

(exercise stage 2; Ex2) of the corrected AT power. As the exercise protocol involved supine cycling sagitally rotated 45°, participants were familiarised with the ergometer during visit 1 and 2. In visit 3, following 10-minutes of rest in the rotated supine position, LV and vascular function were simultaneously investigated using echocardiography and applanation tonometry respectively. Measurements were taken at rest and during Ex1 and Ex2. Each stage of the protocol lasted approximately 10-minutes with data collected during the last 6-minutes. Brachial blood pressure was measured manually at rest and during exercise (Spirit sphygmomanometer aneroid, CK-111, Taipei, Taiwan) and heart rate (HR) was recorded continuously via the ECG attached to the echocardiograph.

Echocardiography: Echocardiographic images were collected and stored using a commercially available ultrasound machine (Vivid *q*, GE Medical Systems, Israel) equipped with a 1.5- to 4-MHz phased array sector transducer (M4S-RS). Images were acquired according to published guidelines [16,17] and were analysed using manufacturer-specific software (EchoPAC, GE Medical, Horten, Norway, version 112). Echocardiographic data were averaged over 3 cardiac cycles and images were analysed with the investigator blinded to the participant's status.

Left ventricular structure and global function: Left ventricular internal diameters and wall thicknesses were measured using 2-dimensional guided M-mode echocardiography. Left ventricular mass was determined according to the Devereux formula and indexed to body surface area [16]. Early (E) and late (A) peak diastolic filling velocities as well as the E/A ratio were determined from the trans-mitral Doppler trace. Peak myocardial tissue velocities during systole (s'), early diastole (e') and late diastole (a') were measured from the pulsed-

wave Doppler trace of the septal mitral annulus. Left ventricular volumes including end-systolic volume (ESV), end-diastolic volume (EDV) and stroke volume (SV) were calculated using the modified biplane Simpson's method [16]. Cardiac output (CO) was calculated as the product of HR and SV. Ejection fraction (EF) was derived from the following equation: [(SV/EDV)*100].

Left ventricular deformation: Left ventricular deformation was quantified by measuring LV strain using speckle tracking echocardiography as described previously [18]. Briefly, 4-chamber long-axis (longitudinal strain) and basal and apical short-axis (circumferential strain) LV video loops were recorded and the endocardial border manually traced using specialised software (EchoPAC, GE Medical, Horten, Norway, version 112). Following initial processing the raw strain data were exported to custom software (2D strain analysis tool, version 1.0β14, Stuttgart, Germany) for further analysis resulting in the generation of peak longitudinal systolic strain and peak basal and apical circumferential systolic strain data.

Arterial function: Pulse wave velocity (PWV) and augmentation index (AIx) were employed as markers of arterial function in this study and were measured using applanation tonometry. Carotid-femoral pulse wave velocity (PWV), the current non-invasive gold-standard technique for the assessment of aortic stiffness, was measured at rest while augmentation index (AIx), a marker of arterial haemodynamic load, was evaluated at rest and during exercise [19]. Duplicate carotid-femoral PWV measurements were obtained using the "foot-to-foot" methodology described in detail previously [19]. The measurement of AIx involved the collection of radial pressure waveforms using a high-fidelity micromanometer (SPC-301; Millar Instruments, Texas, Houston), which were then transformed into central aortic waveforms using a generalised transfer function (GTF) (SphygmoCor7.01; AtCor

Medical, Sydney, Australia). From this waveform AIx was automatically derived by the SphygmoCor software [20]. The GTF has been validated both at rest [21] and during exercise [22]. Augmentation index data are reported as absolute values and, as this variable varies inversely with heart rate, relative to a heart rate of 75 bpm (AIx@75) [22].

Statistical analysis: All data are presented as mean ± SD unless otherwise stated. Differences in resting haemodynamics and global CV structure and function between the cancer survivors and controls were explored using independent-samples t-tests. Differences in LV strain and AIx between groups at rest and during exercise were analysed using independent samples t-tests with a Holm-Bonferroni correction applied for multiple comparisons. Statistical significance was set *a priori* at <0.05. Intra-observer reliability for selected echocardiographic variables at rest and during exercise was determined using intraclass correlation coefficients (ICC) with 95% confidence intervals in a separate test-retest study (n=10). Both at rest and during exercise, ICC for longitudinal and basal and apical circumferential strain varied between 0.91 and 0.99 (all $P<0.0001$).

Results: Participant characteristics are reported in Table 1. The cancer survivor group was similar to the control group in age, sex, height, body mass and body surface area. None of the participants were taking any medications and all were free from co-morbidities. All of the cancer survivors successfully completed the two stages of exercise. The cancer survivors and control participants showed a similar oxygen uptake and power output during the sub-maximal exercise test. Resting CV structure and function variables are presented in Table 3 whilst exercise haemodynamic and LV and arterial function data are reported in Table 4.

Cardiovascular structure and function at rest: Resting LV wall thicknesses, cavity dimensions, volumes, HR and blood pressure were similar between cancer survivors and controls however cancer survivors had a significantly smaller LV mass than controls. Despite this difference, the cancer survivors were still well within normal reference ranges [16]. Global resting LV systolic (EF, SV, CO) and diastolic (E/A ratio) function were not different between groups. Measures of arterial function (PWV and AIx) were also similar in both groups at rest. Circumferential strain and a' were not significantly different between groups at rest, in contrast, the cancer survivors had a lower resting longitudinal strain (Figure 1) and slower s' and e' compared to controls.

Cardiovascular function during exercise: During exercise, blood pressures, HR, EF and CO were comparable between the cancer survivors and controls. Longitudinal strain and s' remained significantly lower in the cancer survivors compared to controls during exercise. Despite similar resting basal and apical circumferential strain between the two groups, on exercise these variables were significantly lower in the cancer survivors during Ex1 (basal and apical circumferential strain) and Ex2 (apical circumferential strain). Although significantly lower at rest, e' was similar in both groups throughout the exercise protocol. In contrast, a' which was comparable at rest was significantly lower in the cancer survivors during Ex1. Cancer survivors had consistently higher AIx compared to controls during exercise but the difference did not reach statistical significance.

Discussion: This study examined whether the concurrent assessment of cardiac deformation and arterial function during exercise would improve the characterisation of sub-clinical CV dysfunction in asymptomatic cancer survivors with prior anthracycline exposure compared to

assessments taken at rest. We found that despite having preserved global LV function (EF) at rest, cancer survivors have reduced resting LV long-axis function (lower longitudinal strain and slower s' and e'). In addition, as hypothesised, further differences were uncovered during exercise with cancer survivors having reduced short-axis function (decreased circumferential strain) and slower late diastolic myocardial velocities (a'). However, no differences in arterial function were identified between the groups either at rest or during exercise. The findings from this study suggest that the assessment of cardiac deformation during exercise may provide a more comprehensive characterisation of sub-clinical LV dysfunction in cancer survivors previously exposed to anthracyclines than resting measures alone.

Detection of sub-clinical cardiovascular dysfunction at rest

The use of myocardial deformation indices in the detection [23,24] and prediction [11] of sub-clinical LV dysfunction in cancer patients undergoing anthracycline-based chemotherapy is well established. In contrast, there are only a limited number of research studies investigating the role of these indices in the early identification of sub-clinical changes to LV function in long-term cancer survivors. The present cohort of asymptomatic cancer survivors had preserved global LV function (EF) 10+ years post-treatment but reduced longitudinal deformation and slower systolic (s') and early diastolic (e') myocardial velocities at rest. These findings are consistent with previous studies involving long-term cancer survivors [25,26]. Sub-endocardial myofibers play a key role in LV long-axis function [27] and are particularly susceptible to a loss of functional myocytes, a common finding in biopsies taken from patients with prior anthracycline exposure [28]. Accordingly, endocardial fibre impairment may explain the reduced LV long-axis function observed at rest in the present cohort of cancer survivors.

The toxic effects of anthracyclines are not confined to the heart but also damage the arterial system causing irregular vascular tone, impaired nitric oxide production and the induction of endothelial apoptosis [29]. Previously, it has been shown that aortic wall stiffness is increased 4 months after chemotherapy in breast cancer, leukaemia and lymphoma patients [30]. In contrast to the previously observed short-term effects of anthracyclines, neither aortic stiffness nor arterial haemodynamic load in the present investigation were chronically increased in cancer survivors several years after the cessation of treatment. Earlier studies evaluating aortic stiffness in long-term cancer survivors have also found either a partial reversal 14 months post-treatment [31] or no differences between controls and cancer survivors 10+ years after treatment [32]. Whilst this may point to a recovery of the vasculature from prior anthracycline exposure, it is also possible that sub-clinical dysfunction is not evident at rest in young otherwise healthy cancer survivors.

Unmasking latent sub-clinical cardiovascular dysfunction during exercise

Blood supply to working muscles during exercise is enhanced via local processes such as decreased vasomotor tone and increased nitric oxide production [33], changes which also lead to reduced arterial haemodynamic load (AIx) [34]. As anthracycline exposure impairs these processes, it was hypothesised that exercise would provoke the appearance of latent differences in AIx between cancer survivors and controls. Yet there were no statistically significant differences in AIx between the groups during exercise suggesting no impairment of the vasculature of long-term cancer survivors at sub-maximal exercise intensities. Whether higher intensities are required to uncover differences in AIx in such an asymptomatic group of cancer survivors requires further investigation.

Conversely, the exercise stimulus was effective in unmasking additional differences in LV systolic (reduced circumferential strain) and diastolic (slower a') function that were not apparent at rest. Moreover, longitudinal deformation remained lower in the cancer survivors throughout the exercise protocol. Tan and colleagues reported similar findings i.e. lower longitudinal function at rest and reduced short axis function only on exercise in older (~71 years) hypertensive patients with NYHA Stage II and III heart failure [5]. Reduced circumferential deformation tends to occur in the more advanced stages of heart failure (NYHA Stage III and IV) while longitudinal deformation is reduced in the earlier stages (Stage I) of the condition [35]. In line with this pathophysiological progression, it appears that the current low-intensity exercise stimulus precipitated a degree of impaired LV short axis function in the cancer survivors more commonly associated with advanced cardiac damage. The timely identification of such damage may allow for improved risk stratification of cancer survivors and earlier intervention in those at most risk. However, longitudinal studies are firstly required to ascertain the association if any between reduced LV short axis function on exercise and the development of overt CV disease in this patient group.

While the assessment of cardiac deformation during exercise appears to improve the characterisation of sub-clinical LV dysfunction in asymptomatic cancer survivors the clinical utility of such an approach is uncertain. Exercise echocardiography is an extremely challenging skill and this reduces the likelihood of it being rapidly adopted into clinical practice. Furthermore our data suggests that while providing more detail, CV evaluation during exercise did not distinguish sub-clinical CV dysfunction beyond that already determined by the resting assessment of LV longitudinal deformation. Larger studies are required to confirm our preliminary findings and to ascertain the added benefit of exercise echocardiography in the detection of sub-clinical CV dysfunction.

Limitations: There were several limitations to the present study including the small sample size and the cross-sectional design, which limits the conclusions that can be drawn from the current findings i.e. the effect of different treatment regimens on the outcome variables. Whilst the identification of sub-clinical LV dysfunction in cancer survivors contributes to the understanding of the pathological process underpinning CTRCD it has not yet been confirmed if these sub-clinical findings are associated with the development of overt clinical CV disease. Owing to ethical restrictions peak $\dot{V}O_2$ was not measured in the present investigation. Consequently despite having similar sub-maximal $\dot{V}O_2$, cancer survivors may have had lower peak $\dot{V}O_2$ values, which in turn may have affected the interpretation of the data.

Conclusion: The assessment of cardiac deformation during exercise appears to improve the characterisation of sub-clinical LV dysfunction in asymptomatic cancer survivors previously exposed to anthracyclines beyond that achieved with simple resting measures. However for the purpose of detecting sub-clinical LV dysfunction, the measurement of myocardial deformation at rest may be sufficient.

References:

1. Yeh ET, Bickford CL. Cardiovascular complications of cancer therapy: incidence, pathogenesis, diagnosis and management. *J Am Coll Cardiol* 2009;**53**:2231-47.

2. Lipshultz SE, Adams MJ, Colan SD, Constine LS, Herman EH, Hsu DT *et al*. Long-term cardiovascular toxicity in children, adolescents, and young adults who receive cancer therapy: pathophysiology, course, monitoring, management, prevention and research

directions: a scientific statement from the American Heart Association. *Circulation* 2013;**128**:1927-95.

3. Plana JC, Galderisi M, Barac A, Ewer MS, Ky B, Scherrer-Crosbie M *et al*. Expert consensus for multimodality imaging evaluation of adult patients during and after cancer therapy: a report from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Soc Echocardiogr* 2014;**27**:911-39.

4. Rosa GM, Gigli L, Tagliasacchi MI, Di Iorio C, Carbone F, Nencioni A *et al*. Update on cardiotoxicity of anti-cancer treatments. *Eur J Clin Invest* 2016;**46**:264-84.

5. Tan YT, Wenzelburger F, Lee E, Heatlie G, Frenneaux M, Sanderson JE. Abnormal left ventricular function occurs on exercise in well-treated hypertensive subjects with normal resting echocardiography. *Heart* 2010;**96**:948-55.

6. Ha JW, Lee HC, Kang ES, Ahn CM, Kim JM, Ahn JA *et al*. Abnormal left ventricular longitudinal functional reserve in patients with diabetes mellitus: implication for detecting subclinical myocardial dysfunction using exercise tissue Doppler echocardiography. *Heart* 2007;**93**:1571-6.

7. Bountioukos M, Doorduijn JK, Roelandt JR, Vourvouri EC, Bax JJ, Schinkel AF *et al*. Repetitive dobutamine stress echocardiography for the prediction of anthracycline cardiotoxicity. *Eur J Echocardiogr* 2003;**4**:300-5.

8. Lanzarini L, Bossi G, Laudisa ML, Klersy C, Aricò M. Lack of clinically significant cardiac dysfunction during intermediate dobutamine doses in long-term childhood cancer survivors exposed to anthracyclines. *Am Heart J* 2000;**140**:315-23.

9. Jarfelt M, Kujacic V, Holmgren D, Bjarnason R, Lannering B. Exercise echocardiography reveals subclinical cardiac dysfunction in young adult survivors of childhood acute lymphoblastic leukemia. *Pediatr Blood Cancer* 2007;**49**:835-40.

10. Khouri MG, Hornsby WE, Risum N, Velazquez EJ, Thomas S, Lane A *et al*. Utility of 3-dimensional echocardiography, global longitudinal strain, and exercise stress echocardiography to detect cardiac dysfunction in breast cancer patients treated with doxorubicin-containing adjuvant therapy. *Breast Cancer Res Treat* 2014;**143**:531-9.

11. Thavendiranathan P, Poulin F, Lim KD, Plana JC, Woo A, Marwick TH. Use of myocardial strain imaging by echocardiography for the early detection of cardiotoxicity in patients during and after cancer chemotherapy: a systematic review. *J Am Coll Cardiol* 2014;**63**:2751-68.

12. Drafts BC, Twomley KM, D'Agostino R Jr, Lawrence J, Avis N, Ellis LR *et al*. Low to moderate dose anthracycline-based chemotherapy is associated with early non-invasive imaging evidence of subclinical cardiovascular disease. *JACC Cardiovas Imaging* 2013;**6**:877-85.

13. Nikooie R, Gharakhanlo R, Rajabi H, Bahraminegad M, Ghafari A. Non-invasive determination of anaerobic threshold by monitoring the %SpO2 changes and respiratory gas exchange. *J Strength Cond Res* 2009;**23**:2107-13.

14. Beaver WL, Wasserman K, Whipp BJ. A new method for detecting anaerobic threshold by gas exchange. *J Appl Physiol* 1986;**60**:2020-7.

15. Doucende G, Schuster I, Rupp T, Startun A, Dauzat M, Obert P *et al*. Kinetics of left ventricular strains and torsion during incremental exercise in healthy subjects: the key

role of torsional mechanics for systolic-diastolic coupling. *Circ Cardiovasc Imaging* 2010;**3**:586-94.

16. Lang RM, Badano LP, Mor-Avi V, Afilalo J, Armstrong A, Ernande L *et al*. Recommendations for chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Soc Echocardiogr* 2015;**28**:1-39.

17. Hill JC, Palma RA. Doppler tissue imaging for the assessment of left ventricular diastolic function: a systematic approach for the sonographer. *J Am Soc Echocardiogr* 2005;**18**:80-8.

18. Stöhr EJ, González-Alonso J, Shave R. Left ventricular mechanical limitations to stroke volume in healthy humans during incremental exercise. *Am J Physiol Heart Circ Physiol* 2011;**301**:478-87.

19. Laurent S, Cockcroft J, Van Bortel L, Boutouyrie P, Giannattasio C, Hayoz D *et al*. Expert consensus document on arterial stiffness: methodological issues and clinical applications. *Eur Heart J* 2006;**27**:2588-605.

20. Wilkinson IB, Fuchs SA, Jansen IM, Spratt JC, Murray GD, Cockcroft JR *et al*. Reproducibility of pulse wave velocity and augmentation index measured by pulse wave analysis. *J Hypertens* 1998;**16**:2079-84.

21. Pauca AL, O'Rourke MF, Kon ND. Prospective evaluation of a method for estimating ascending aortic pressure form the radial artery pressure waveform. *Hypertension* 2001;**38**:932-7.

22. Sharman JE, Lim R, Qasem AM, Coombes JS, Burgess MI, Franco J *et al*. Validation of a generalized transfer function to non-invasively derive central blood pressure during exercise. *Hypertension* 2006;**47**:1203-8.

23. Poterucha JT, Kutty S, Lindquist RK, Li L, Eidem BW. Changes in left ventricular longitudinal strain with anthracycline chemotherapy in adolescents precede subsequent decreased left ventricular ejection fraction. *J Am Soc Echocardiogr* 2012;**25**:733-40.

24. Sawaya H, Sebag IA, Plana JC, Januzzi JL, Ky B, Tan TC *et al*. Assessment of echocardiography and biomarkers for the extended prediction of cardiotoxicity in patients treated with anthracyclines, taxanes, and trastuzumab. *Circ Cardiovasc Imaging* 2012;**5**:596-603.

25. Cheung YF, Hong WJ, Chan GC, Wong SJ, Ha SY. Left ventricular myocardial deformation and mechanical dyssynchrony in children with normal ventricular shortening fraction after anthracycline therapy. *Heart* 2010;**96**:1137-41.

26. Ho E, Brown A, Barrett P, Morgan RB, King G, Kennedy MJ *et al*. Subclinical anthracycline- and trastuzumab-induced cardiotoxicity in the long-term follow-up of asymptomatic breast cancer survivors: a speckle tracking echocardiographic study. *Heart* 2010;**96**:701-7.

27. Geyer H, Caracciolo G, Abe H, Wilansky S, Carerj S, Gentile F *et al*. Assessment of myocardial mechanics using speckle tracking echocardiography: fundamentals and clinical applications. *J Am Soc Echocardiogr* 2010;**23**:351-69.

28. Ewer MS, Ali MK, Mackay B, Wallace S, Valdivieso M, Legha SS *et al*. A comparison of cardiac biopsy grades and ejection fraction estimations in patients receiving Adriamycin. *J Clin Oncol* 1984;**2**:112-7.

29. Soultati A, Mountzios G, Avgerinou C, Papaxoinis G, Pectasides D, Dimopoulos MA *et al*. Endothelial vascular toxicity from chemotherapeutic agents: preclinical evidence and clinical implications. *Cancer Treat Rev* 2012;**38**:473-83.

30. Chaosuwannakit N, D'Agostino R Jr, Hamilton CA, Lane KS, Ntim WO, Lawrence J *et al*. Aortic stiffness increases upon receipt of anthracycline chemotherapy. *J Clin Oncol* 2010;**28**:166-72.

31. Grover S, Lou PW, Bradbrook C, Cheong K, Kotasek D, Leong DP *et al*. Early and late changes in markers of aortic stiffness with breast cancer therapy. *Intern Med J* 2015;**45**:140-7.

32. Vandecruys E, Mondelaers V, De Wolf D, Benoit Y, Suys B. Late cardiotoxicity after low dose of anthracycline therapy for acute lymphoblastic leukemia in childhood. *J Cancer Surviv* 2012;**6**:95-101.

33. Hellsten Y, Nyberg M, Jensen LG, Mortensen SP. Vasodilator interactions in skeletal muscle blood flow regulation. *J Physiol* 2013;**590**:6297-305.

34. Sharman JE, McEniery CM, Campbell RI, Coombes JS, Wilkinson IB, Cockcroft JR. The effect of exercise on large artery haemodynamics in healthy young men. *Eur J Clin Invest* 2005;**35**:738-44.

35. Kosmala W, Plaksej R, Strotmann JM, Weigel C, Herrmann S, Niemann M, et al. Progression of left ventricular functional abnormalities in hypertensive patients with heart failure: an ultrasonic two-dimensional speckle tracking study. *J Am Soc Echocardiogr* 2008;**21**:1309-17.

Figure legend:

Figure 1: **Differences in peak left ventricular strain between control and anthracycline groups at rest and during exercise (mean ±SD).** The anthracycline group had significantly lower (less negative) longitudinal strain at rest and during Ex1 and Ex2 compared to controls (top). Despite comparable apical (middle) and basal (bottom) circumferential strain at rest the anthracycline group had significantly lower values (less negative) in these parameters during exercise. Ex1: exercise stage 1; Ex2: exercise stage 2; **P*<0.05; †*P*<0.01.

Table 1: **Demographics of study population (mean ±SD).**

Variable	**ANT (n=13)**	**Control (n=13)**	***P* value**
Age (years)	36 ±10	35 ±12	0.742
Gender (M/F)	10/3	10/3	----
Height (cm)	174 ±9	175 ±11	0.906
Body mass (kg)	80 ±13	79 ±15	0.868
Body surface area (m^{-2})	1.9 ±0.2	1.9 ±0.2	0.940
Co-morbidities	None	None	----
Medication	None	None	----
$\dot{V}O_2$ at AT ($ml \cdot min^{-1} \cdot kg^{-1}$)*	18 ±4	19 ±4	0.369
Workload at AT (W)*	95 ±30	110 ±38	0.294

ANT: anthracycline; M: male; F: female; $\dot{V}O_2$: oxygen uptake; AT: anaerobic threshold; *: AT determined using V Slope method applied to sub-maximal gas exchange data.

Table 2: **Cancer diagnoses and treatment details (mean ±SD, % or range; n=13).**

Variable		
Cancer type	Hodgkin's lymphoma	3 (23%)
	Non-Hodgkin's lymphoma	6 (46%)
	Ewing's sarcoma	1 (8%)
	Acute lymphoblastic leukaemia	3 (23%)
Years since therapy	11 ±8 (range: 2-33)	
Age at cancer (years)	25 ±13 (range: 4-47)	
ANT type	Doxorubicin	13 (100%)
ANT cumulative dose ($mg \cdot m^{-2}$)	317 ±106 (range: 150-450)	
Radiotherapy	Yes/No	5 (38%)/8 (62%)
Radiotherapy location	Abdomen	1 (8%)
	Mediastinum	2 (15%)
	Brain and spinal cord	1 (8%)
	Neck	1 (8%)

ANT: anthracycline.

Table 3: **Global cardiovascular structure and function at rest (mean ±SD).**

Variable	**ANT (n=13)**	**Control (n=13)**	***P* value**
LV structure			
LVPWs (cm)	1.4 ±0.3	1.5 ±0.3	0.164
LVIDs (cm)	3.3 ±0.4	3.4 ±0.4	0.562
IVSs (cm)	1.4 ±0.2	1.5 ±0.3	0.453
LVPWd (cm)	0.9 ±0.1	1.0 ±0.2	0.081
LVIDd (cm)	4.7 ±0.4	4.9 ±0.5	0.295
IVSd (cm)	1.0 ±0.2	1.1 ±0.2	0.106
LV mass (g)	153 ±45	194 ±55	**0.049**
LV mass index ($g \cdot m^{-2}$)	78 ±18	99 ±20	**0.010**
LV volumes			
End-systolic volume (ml)	47 ±12	45 ±12	0.702
End-diastolic volume (ml)	101 ±19	102 ±23	0.886
Global LV systolic function			
Ejection fraction (%)	54 ±5	56 ±4	0.269
Global LV diastolic function			
Trans-mitral E vel. ($m \cdot s^{-1}$)	0.7 ±0.1	0.7 ±0.2	0.682
Trans-mitral A vel. ($m \cdot s^{-1}$)	0.4 ±0.1	0.3 ±0.1	0.361

E/A ratio	2.06 ±0.63	2.26 ±1.27	0.624
Aortic stiffness			
C-F pulse wave velocity ($m \cdot s^{-1}$)	6.1 ±1.0	6.6 ±1.6	0.385

ANT: anthracycline; LV: left ventricle; LVPW: left ventricular posterior wall; LVID: left ventricular internal diameter; IVS: inter-ventricular septum; s: systole; d: diastole; E: early diastolic; A: late diastolic; vel: velocity; C-F: carotid-femoral.

Table 4: **Haemodynamics and global left ventricular and arterial function at rest and during exercise (mean ±SD).**

	Exercise Intensity		
	Rest	**Ex1**	**Ex2**
Haemodynamics			
Systolic blood pressure (mmHg)			
ANT	113 ±13	124 ±14	132 ±15
Control	115 ±19	126 ±17	135 ±21
Diastolic blood pressure (mmHg)			
ANT	68 ±9	78 ±8	79 ±5
Control	68 ±11	74 ±10	77 ±11
Heart rate (bpm)			
ANT	58 ±5	84 ±7	95 ±9
Control	53 ±8	76 ±9	85 ±12

Global LV function			
Ejection fraction (%)			
ANT	54 ±6	59 ±5	59 ±5
Control	56 ±4	60 ±4	64 ±4
Cardiac output ($L \cdot min^{-1}$)			
ANT	3.6 ±1.1	5.8 ±1.2	6.7 ±1.7
Control	4.0 ±1.0	6.2 ±1.6	7.4 ±2.0
s' ($m \cdot s^{-1}$)			
ANT	0.07 ±0.01†	0.08 ±0.01†	0.10 ±0.02†
Control	0.09 ±0.01	0.10 ±0.01	0.12 ±0.01
e' ($m \cdot s^{-1}$)			
ANT	0.09 ±0.02†	0.11 ±0.02	0.12 ±0.03
Control	0.12 ±0.03	0.13 ±0.03	0.14 ±0.03
a' ($m \cdot s^{-1}$)			
ANT	0.07 ±0.02	0.09 ±0.02†	0.10 ±0.03
Control	0.09 ±0.02	0.12 ±0.02	0.13 ±0.03
Arterial Function			
Augmentation index (%)			

ANT	20 ±15	13 ±14	7 ±14
Control	11 ±12	8 ±10	1 ±8
AIx@75 (%)			
ANT	12 ±14	18 ±14	17 ±13
Control	1 ±12	9 ±9	6 ±10

ANT: anthracycline; Ex1: exercise stage 1; Ex2: exercise stage 2; LV: left ventricular; s': systolic myocardial velocity; e': early diastolic myocardial velocity; a': late diastolic myocardial velocity; AIx@75: augmentation index normalised to heart rate 75 bpm; **P*<0.05; †*P*<0.01.

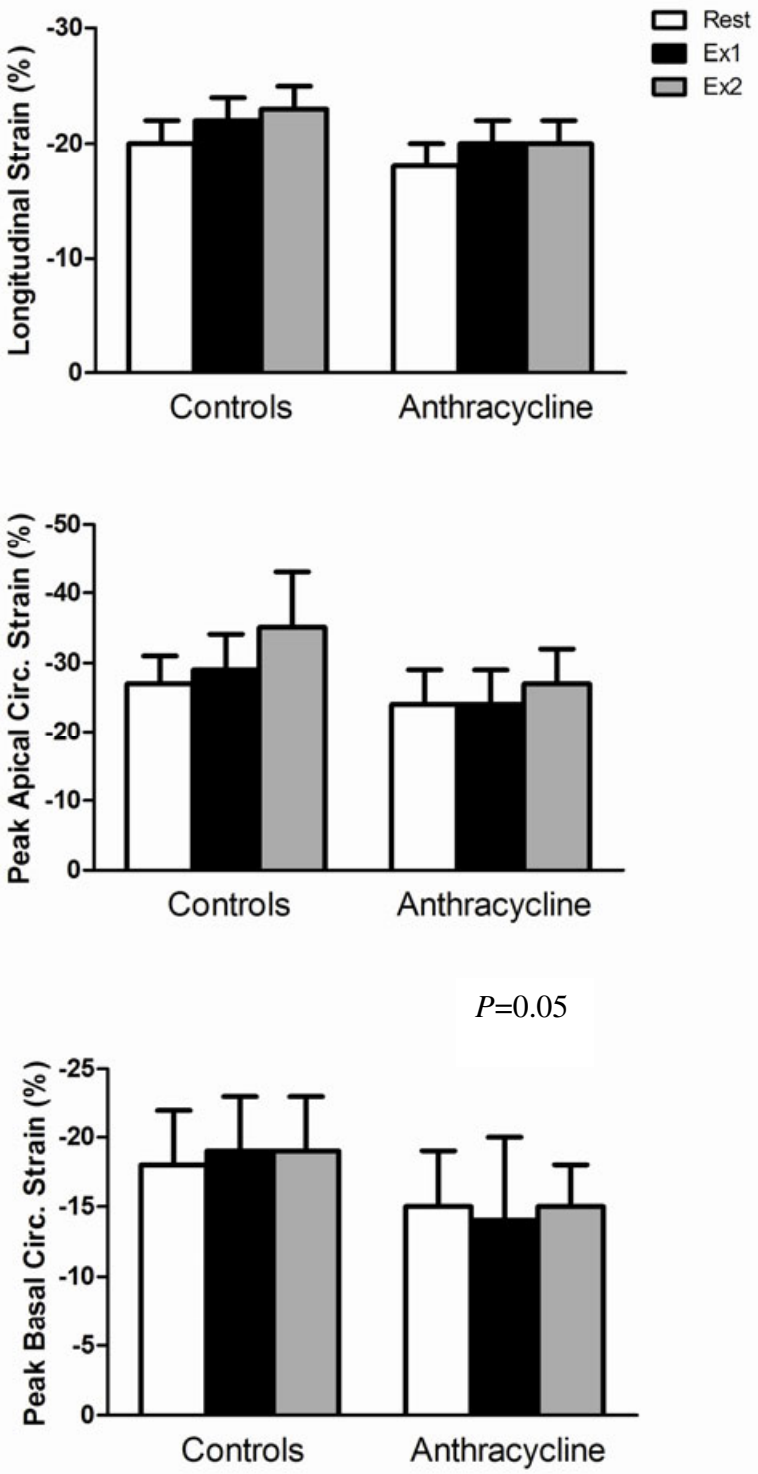

Figure 1: **Differences in peak left ventricular strain between control and anthracycline groups at rest and during exercise (mean ±SD).**