Possibilities of Early Detection of Cardiotoxicity in Patients with Malignancy

PhD thesis

Dr. András Csaba NAGY

Tutor:
Prof. Dr. Tamás Forster MD, PhD, DsC

2nd Department of Medicine and Cardiology Center, Medical Faculty,
Albert Szent-Györgyi Clinical Center,
University of Szeged

Szeged
2018

List of full papers that served as the basis of the PhD thesis

I. Nagy ACs, Tolnay E, Nagykálnai T. et al.: Kemoterápia okozta szívizom-károsodás: a diagnosztika lehetőségei. Orvosi Hetilap 2004 Oct 24;145(43):2171-6.

II. Nagy ACs, Tolnay E, Forster T et al.: Cardiotoxicity of anthracycline in young breast cancer female patients: the possibility of detection of early cardiotoxicity. TDI. Neoplasma. 2006;53(6):511-7.

III. Nagy ACs, Cserép Zs, Tolnay E et al.: Early diagnosis of chemotherapy induced cardiomyopathy: a prospective tissue Doppler imaging study. Pathol.Oncol.Res. 2008; 14:69-77.

IV. Nagy ACs, Gulácsi-Bárdos P, Cserép Zs, et al.: Late cardiac effect of anthracycline therapy in physically active breast cancer survivors - a prospective study. Neoplasma 2017;64(1):92-100.

Other publications

I. Nagy András Csaba: A cardiovascularis prevenció - a kockázat csökkentésének lehetőségei, 2010, LAM (Lege Artis Medicinæ) - 2010;20(08)

II. Székely A, Cserép Z, Sápi E, Breuer T, Nagy CA, Vargha P, Hartyánszky I, Szatmári A, Treszl A.: Risks and predictors of blood transfusion in pediatric patients undergoing open heart surgery.
Annals of Thoracic Surgery, Ann Thorac Surg. 2009 Jan;87(1):187-97.

III. Nagy A, Cserép Z.: Link between diabetes and diastolic dysfunction and the diagnostic role of echocardiography. Orv Hetil. 2009 Nov 8;150(45):2060-7. Review. Hungarian.

IV. Nagy, András Csaba MD., Cserép, Zsuzsanna MD., Tolnay, Edina MD., Forster, Tamás MD.: Diastolic disfunction due to chemotherapy: early diagnosis with tissue Doppler imaging Poster – ESC Euroecho – 2008 Liszabon, Portugália European Journal of Echocardiography 8, Suppl. 1, Dec, 2007, 1525-2167.

V. Nagy, András Csaba MD., Cserép, Zsuzsanna MD., Tolnay, Edina MD., Forster, Tamás MD.: Early prediction of chemotherapy induced late cardiotoxicity: the role of Tissue Doppler Imaging in detection of diastolic dysfunction Poster – ESMO Congress, Stockholm, Sweden, 2008, 12-16 Sept. Annals of Oncology 19, 2008 Suppl.8., 261

VI. Nagy A., Tolnay E., Cserep Z., Forster T.: Chemotherápia okozta kései szívizomkárosodás előrejelzése: a szöveti Doppler szerepe a diasztolés diszfunkció kimutatásában. Poster Magyar Belgyógyász Társaság 2008 évi Nagygyűlése, Magyar Belorvosi Archivum, 2008. Suppl.

Table of contents

Abbreviation

A – late mitral inflow wave velocity

AC – adriamycine and cyclophosphamide

Ae – segmental late diastolic wave velocity

CK – creatine kinase

CK MB – creatine kinase Muscle/ Brain Isoenzyme

CV – cardiovascular

D – Pulmonaric venous D wave velocity

DD - diastolic dysfunction

DT - deceleration time

E – early mitral inflow wave velocity

E/Ea - early mitral inflow wave velocity/segmental early diastolic wave velocity ratio

Ea – segmental early diastolic wave velocity

Ea/Aa – Ea/Aa ratio

E/A – E wave/ A wave ratio

EC – epirubicin and cyclophosphamide

ECG – electrocardiography

EF – ejection fraction

FS - fractional shortening

GLS - global longitudinal strain

LVEDd – left ventricular end diastolic diameter in systole

LVEDs – left ventricular end diastolic diameter in systole

LVEF – left ventricular ejection fraction

MAV - mitral annular velocity

PA - physically active

PNA - physically inactive

S – Pulmonaric venous S wave velocity

S/D – S/D ratio

TDI - tissue Doppler imaging

3D – three dimension

2D – two dimension

Introduction

Chemotherapy-induced cardiotoxicity was first described in the literature in 1967 in children with leukemia treated with doxorubicin (1). Subsequently, an increasing number of studies have addressed this new entity (2, 3), and cardiotoxicity has received increasing attention as a potential chemotherapy-related adverse event. There are a growing number of publications on cardiotoxicity, and the protocol of the most recent international trials must include careful monitoring of cardiovascular parameters. Prevention became a widespread approach in this field as well, similarly to trends in cardiology in recent decades. Aggressive combination antitumour therapy may lead to remission or healing in many types of malignancies, but it comes with a price. A large body of literature data has been published on the myocardial side effects of widely used anthracyclines (4, 5, 6), such as doxorubicin and epirubicin, but these adverse effects corresponded mostly to acute cardiomyopathy and were associated with decreased ejection fraction and pump function, and manifested in the symptoms of heart failure.

Administering smaller doses at less frequent intervals helped to avoid these acute damages. However, the so-called late cardiotoxicity became increasingly common, most of the times occurred in a subclinical form, often progressive, potentially severe and sometimes causing fatal heart failure in children (7). There are two forms of late or chronic cardiotoxicity: the early-onset and the late-onset chronic progressive cardiotoxicity (8). The former develops within one year after the anthracycline therapy, while the latter up to decades after the therapy. Furthermore, the awareness of the risk factors which may be involved in anthracycline therapy-induced cardiomyopathy is increasing. For example, in addition to known cardiovascular risk factors (such as known CV condition, age, gender, genetic background, smoking, unhealthy eating habits, hypertension, hypercholesterinemia, diabetes and obesity), the type of the antitumour agent (anthracycline), higher cumulative dose, rapid infusion, combination therapy and concomitant chest radiotherapy also play a role (9).

The fight against cardiotoxicity has come to the fore because survival of the patients significantly improved despite the underlying malignancy.

Later complaints are caused by secondary organ impairment and cardiomyopathy (resulting from previous medication), as well as by progressive heart failure (resulting from cardiomyopathy), and these ultimately lead to fatal outcome.

This is the reason why both oncologists and cardiologists focus more and more on this topic. An independent subfield was born, that is cardio-oncology.

Anthracycline-induced cardiomyopathy may develop even after many years of the treatment. Some signs have already been described which may be indicative of subclinical cardiotoxicity, even in the absence of specific symptoms or radiological signs (10), such as a change in fractional shortening (FS) observed using echocardiography, or a decrease of left ventricular function below 50%, potentially early-onset dyskinesia, akinesia or abnormal wall motion. Early detection of subclinical cardiotoxicity can help choosing the appropriate early treatment and improving the chance of long-term survival (11). The need for long-term monitoring and multidisciplinary care of breast cancer survivors was pointed out by Burstein and Winer in their publication in 2000 (12).

Several methods can be used to detect chemotherapy-induced cardiomyopathy. Of the non-invasive methods, the ECG, heart frequency variability, determination of ventricular repolarization time and echocardiography provide a basis for assessing the cardiac impairment (13). Of the laboratory tests, the literature highlights monitoring of the increased levels of myocardial biomarkers (CK, CK-MB, Troponin I and T) in case of damage, and the determination of natriuretic peptides, which can be used even in the early detection of subclinical damages (14,15). Of the imaging techniques, antimyosin (In-111-antimyosin-Fab antibody) and Thallium 201 perfusion scintigraphy is used as a direct procedure to detect myocardial damage, and the radionuclide ventriculography has long been the gold standard in measuring the ejection fraction (16, 17). Sequential MRI can provide better resolution than various isotope methods. The predominant method is the non-invasive echocardiography which provides valuable data and can be repeated several times and used to determine both left ventricular systolic and diastolic functions. Using the above-mentioned well-defined measurements, it is currently widely used as gold standard to measure and monitor the ejection capacity of the heart muscle (18). However, the measurement of the diastolic function provides a measure of the possible damage to the relaxation capacity only with approximate accuracy (19). Accurate data on this is important because, on the one hand, they provide preliminary information on the damage of the myocardium, and on the other hand, the systolic

function can remain normal for a long period of time in the case of subclinical cardiomyopathy, despite the late onset of heart failure symptoms.

Though the invasive catheter technique has long been used as a standard method to measure diastolic function (20), as echocardiography developed, this technique has been increasingly replaced by the evaluation of the mitral inflow Doppler pattern (21). However, in addition to the diastolic properties of the left ventricle, traditional E/A inflow pattern can be influenced by other factors, namely preload, afterload, frequency, atrioventricular delay, ventricular interaction, viscoelastic properties, and pericardial limiting factors. Thus, the mitral inflow pattern is easy to record, but the above-mentioned factors should also be taken into account in the evaluation. In many cases, knowing the pulmonary venous flow or the deceleration time is of no avail.

Tissue Doppler imaging (TDI), described for the first time in 1989 (22), was the new method which allowed not only to measure the regional myocardial function (23) and obtain a value comparable to global left ventricular function by measuring the systolic mitral annular velocity (24), but also made it possible to accurately measure the diastolic properties of a specific portion of the ventricular wall using pulse Doppler technique (25), regardless of the presence of the above-mentioned influencing factors.

In addition to the management of the above-mentioned risk factors, there is an increasing body of evidence that physical activity plays a role in the prevention in patients with malignancies. In the case of breast cancer, regular exercise can reduce the risk of the disease by up to 50%. Physical activity before doxorubicin therapy can reduce the severity of anthracycline-induced myocardial dysfunction by maintaining fractional fibre shortening and preserving pressure and contractility. Exercise increases cardiovascular efficacy, improves cardiac output and stroke volume, reduces resting pulse rate, and improves breathing and oxygen transport from the lungs to the cells (26). Animal studies have confirmed the protective effect of one-time and regular exercise against doxorubicin-induced cardiac damage, which may be explained by increased antioxidant capacity and oxygen use as a result of this exercise (27, 28).

Objectives

Obj. 1. This prospective study attempted to establish whether the anthracycline-containing treatment leads to the development of so-called subclinical myocardial damage in the early stages in female patients with breast cancer and without cardiovascular risk factors.
Obj. 2. Furthermore, this study explored the extent to which tissue Doppler imaging can be used in the early diagnosis of subclinical cardiomyopathy in patients with mammary tumours treated with anthracycline.
Obj. 3. A further objective of this study was to confirm the suitability for use of tissue Doppler echocardiography (TDI) in the early detection of subclinical myocardial damage using long-term measurements.
Obj. 4. The extended study aimed at confirming the preventive effect of regular exercise, already confirmed by literature data, regarding the anthracycline-induced myocardial damage and possible heart failure.

Materials and methods

Obj. 1. Measuring early subclinical damage using traditional methods

A number of 40 women, aged between 31 and 65 years of age (mean age: 50±9 years), treated between April 2003 and January 2006 at the Department of Radiation Oncology of the Uzsoki Hospital, who also received chemotherapy after surgery for malignant mammary carcinoma, were enrolled in this study. The patients had no cardiovascular risk factors (Table 1). From this population, we enrolled patients with breast cancer on the right side in order to avoid the radiation of the left side of the chest. Female patients with mammary carcinoma of low or intermediate invasivity underwent postoperative chemotherapy, which included 4 series of EC or AC infusions. The cumulative mean dose of doxorubicin and epirubicin was 240 mg/ m^2 and 360 mg/m^2, respectively.

Supportive treatment for the underlying condition did not constitute an exclusion criterion (e.g. palliative radiation treatment in other regions, pain relief, antiemetics, and bisphosphonates).

Table 1. Exclusion criteria

- hypertension (severe or mild)
- anemia
- diabetes mellitus
- coronary artery disease
- left ventricular hypertrophy
- severe aortic stenosis
- mitral valve disease
- cardiomyopathy
- radiation of the left chest or mediastinum

The control group included 20 women with similar age distribution, between 33 and 62 years of age (mean age: 49±10 years), without cardiovascular risk. Participants in the control group were examined according to the same protocol.

Cardiology examination

In addition to the patients' cardiac history, their physical status was recorded, paying particular attention to their cardiovascular status. Resting 12-leads ECG and blood pressure measurements were performed.

Standard echocardiography

This echocardiography was performed using a Vivid 3 ultrasound machine. The measurements were carried out in M mode, at the parasternal longitudinal section of the left ventricle, below the apex of the mitral valve. Standard parameters were measured during systole and diastole: dimensions of the left ventricle, E-septum distance to the mitral valve, the width of the aorta and the diameter of the left atrium. In the apical 4-chamber view, the

left ventricular ejection fraction was calculated using the modified Simpson formula; atrial dimensions were measured; the mitral inflow curve was assumed so that the depth of the pulse Doppler was positioned between the mitral valves in the middle of the flow. We also examined the peak velocity of the early (E wave) and late (A wave) inflow, the E/A ratio and the deceleration time (DT). The pulmonary venous curve was obtained at the opening of the right upper pulmonary vein, approximately at a depth of 1 cm. We recorded the pulmonary venous peak systolic (S) and peak diastolic (D) flow velocities, the S/D ratio, and the velocity and duration of the peak reverse flow. Ideally, the diastolic function of the heart can be determined using these values.

We also examined wall motions in each view, the anatomy of the valve, alterations and potential pericardial effusion.

Schedule of examinations

Examination 1: Before the initiation of the chemotherapy: data collection and recording cardiac status using the methods described above.

Examination 2: 12 months after the initiation of the treatment.

Statistical analysis

Data were evaluated using Kolmogorov-Smirnov normality test, and different statistical tests and analyses were performed accordingly. Data were presented as a mean ± SD of three measurements. Basic data were compared using independent *t*-test. Heart rate, blood pressure, standard ultrasound values and mitral inflow velocity values were analyzed at all time points using *t*-test. These values were analyzed using a statistics software (SPSS, Version 10.0, SPSS Inc., Chicago, III), with a significance limit of $p<0.05$.

Obj. 2. Measuring early subclinical damage using tissue Doppler imaging

The analysis included the study group and the control group detailed in section Obj. 1, and the schedule was developed accordingly.

Cardiac examination and standard echocardiography

These were performed as detailed in section Obj. 1.

Tissue Doppler imaging (TDI)

This examination was performed using a Vivid 3 ultrasound machine in TDI mode. The mitral annular velocity (MAV) was measured using pulse TDI from transthoracic view. The velocity was measured in 4 different points of the mitral annulus from apical view. The lateral velocity patterns were obtained from the proximal annulus of the septal (septum) or lateral wall by placing the TDI cursor on the proximal mitral annulus of the septum in the apical 4-chamber view. Similarly, the anterior and inferior velocity patterns were obtained from the apical 2-chamber view. The mean of velocity values recorded in 3 cycles from this position was used in the statistical calculations. Three high velocity peak values were obtained from the 4 mitral annular sites. One value is the positive systolic velocity (S) when the mitral ring moves toward the heart apex, as well as two negative diastolic velocities, when the ring moves away from the apex toward the basis. The first and the second velocity provide the early diastolic (Ea) wave and late diastolic (Aa) wave, respectively. Similar measurements were performed in the parasternal longitudinal section by placing the Doppler cursor in the middle of the septum and posterior wall. The ratio of the E wave of the traditional inflow and the early (Ea) diastolic wave of the mitral annulus measured with TDI describes the atrial filling pressure well, which was evaluated as a derived parameter. The Ea/Aa value measured during the tissue Doppler measurement (TDI) is considered abnormal below 1, which is an artificial limit, but it provides a good description of the essential damage to the diastolic function.

Statistical analysis

Data were analyzed using statistical calculations described in section Obj. 1. Similarly to the above, the annular velocity values were analyzed at all time points using the t-test.

Obj. 3. Measuring long-term subclinical damage

Our extended study included the study group and the control group detailed in section Obj. 1.

Study protocol

Schedule of detailed cardiac examinations:
Before the initiation of the chemotherapy (T0), at 1 month after the first chemotherapy cycle (T1), at 1 month after the second chemotherapy (T2), at 1 year (T3), then at two years (T4) after the initiation of the chemotherapy. In addition to the cardiac history of the patients, we also recorded their physical status with particular attention to their cardiovascular status and possible symptoms of circulatory insufficiency. Resting 12-leads ECG and blood pressure measurements were also performed. Echocardiography was carried out at each time point.

Standard echocardiography and tissue Doppler imaging (TDI)

Performed as described in sections Obj. 1 and Obj. 2.

Obj. 4. Preventive effects of physical activity

The extended study included 55 young women with breast cancer and without cardiovascular risk, treated at the Department of Radiation Oncology of the Uzsoki Hospital; most of them were from the study group described above (Obj. 1). We used the same anthracycline dose as above, while the cyclophosphamide dose was 500 mg/m2 in the study group, according to the standard protocol. Supportive treatment for the underlying condition did not constitute an exclusion criterion (e.g. analgesics, antiemetics, palliative radiation treatment in other regions, and bisphosphonates).

Patients were included into two groups depending on whether they were physically active before their disease or not. Intensive physical activity carried out at least 4 or 5 days a week, for at least half an hour, including any individual or team sports, was considered exercise. Based on the above-mentioned definition, 36 patients and 19 patients were included in the physically active group (PA) and the physically inactive group (PNA), respectively.

Schedule of examination

The cardiac examinations were carried out in four phases in this prospective study: before the chemotherapy (T1); at mid-term of the anthracycline therapy (T2); at one year after the first chemotherapy cycle (T3); at 2 years after the first chemotherapy cycle (T4). We assessed the occurrence of symptoms (such as breathlessness, reduced capacity, fatigue, foot swelling, nocturia) indicative of cardiac failure by phone 5 years after the first examination in these patients (T1+5 year).

Standard echocardiography and tissue Doppler imaging (TDI)

Performed as described in sections Obj. 1 and Obj. 2.

Statistical analysis

The two groups were compared using the Mann-Whitney test and the Fischer exact test. The results were presented as mean ± SD.

Results

Obj. 1. Measuring early subclinical damage using traditional methods

A number of 40 female patients with breast cancer were enrolled in this study, with a control group of 20 persons of similar gender and with negative cardiac history and risk factors. We evaluated the traditional systolic and diastolic function parameters. The age distribution of the study group and the control group was the same.

Table 2 shows the basic data and standard echocardiographic values of the study group and the control group at baseline and at 1 year.

	Begining			After 1 year		
	Patient (n=40)	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*
Gender (year)	49,2 ± 10,1	50,1 ± 8,5	ns			
Heart rate (1/min)	78,7 ± 12,6	72,6 ± 9,5	ns	83,0 ± 11,3	72,1 ± 15,5	0,003
Blood pressure (mmHg)	131/81 ± 16/8	128/77 ± 11/6	ns	128/78 ± 13/7	124/77 ± 10/4	ns
Aortic root (cm)	2,64 ± 0,26	2,52 ± 0,36	ns	2,58 ± 0,25	2,44 ± 0,27	ns
Left atrium (cm)	3,83 ± 0,45	3,97 ± 0,37	ns	4,89 ± 0,32	4,40 ± 0,87	0,030
LVEDd (cm)	5,11 ± 0,56	5,09 ± 0,53	ns	5,41 ± 0,43	5,06 ± 0,52	0,007
E-septum separation (cm)	0,50 ± 0,18	0,46 ± 0,12	ns	0,51 ± 0,13	0,45 ± 0,09	0,070
EF (Simpson) %	66,9 ± 5,8	64,2 ± 4,7	ns	65,7 ± 3,6	67,7 ± 2,8	0,030
Mitral E velocity (m/s)	0,78 ± 0,13	0,78 ± 0,09	ns	0,65 ± 0,11	0,76 ± 0,09	0,001*
Mitral A velocity (m/s)	0,62 ± 0,13	0,59 ± 0,12	ns	0,73 ± 0,10	0,57 ± 0,08	0,001*
E/A ratio	1,30 ± 0,29	1,35 ± 0,26	ns	0,91 ± 0,18	1,33 ± 0,23	0,001*
Deceleration time (ms)	253 ± 49	275 ± 53	ns	301 ± 42	280 ± 56	ns
Pulmonary S (m/s)	0,56 ± 0,10	0,59 ± 0,08	ns	0,43 ± 0,12	0,57 ± 0,08	0,001*
Pulmonary D (m/s)	0,36 ± 0,09	0,38 ± 0,08	ns	0,55 ± 0,14	0,36 ± 0,11	0,001*
S/D ratio	1,58 ± 0,29	1,57 ± 0,27	ns	0,87 ± 0,51	1,55 ± 0,11	0,001*

* means significant difference in values within the patient group at the end as compared to the initial status

Table 2. Basic data and standard echocardiographic parameters of patients

There were no differences in the basic data and standard ultrasound measurements between the two groups at baseline. The pulse rate at 1 year was significantly higher in the study group. As regards blood pressure, there was no substantial difference at 1 year, which is important because the change in blood pressure could not cause diastolic dysfunction (DD) in either group.

Based on the cardiac physical examination, the ECG and the evaluation of the symptoms, it can be established that there were no significant differences between the two groups in their circulatory status.

After 1 year, there was a significant difference in the standard ultrasound parameters (aortic root, left atrium, E-septum separation, LVEDd); the heart cavities were enlarged in the treatment group as a result of the cardiotoxicity. However, this was not associated with either clinical symptoms or decreased systolic function. The EF did not decrease below 60%, that is, there was a clinically insignificant, but statistically measurable difference between the two groups.

As regards the traditional measurements of diastolic dysfunction (mitral E velocity, mitral A velocity, E/A ratio, deceleration time, S/D ratio), there was a significant difference within the study group in the values obtained at the end of the study compared to the baseline, and between the study group and the control group at the last measurement. However, the E/A ratio and the deceleration time showed differences indicative of diastolic dysfunction (DD) in only 30 patients (75% of the cases), in other words, this is the number of patients who supposedly developed diastolic dysfunction within 1 year (Figure 1).

Obj. 2. Measuring early subclinical damage using tissue Doppler imaging

Table 3 contains TDI measurements by each segment (6 measurements) at the baseline and at the end of the study (at 1 year) both in the patient and the control groups (Table 3).

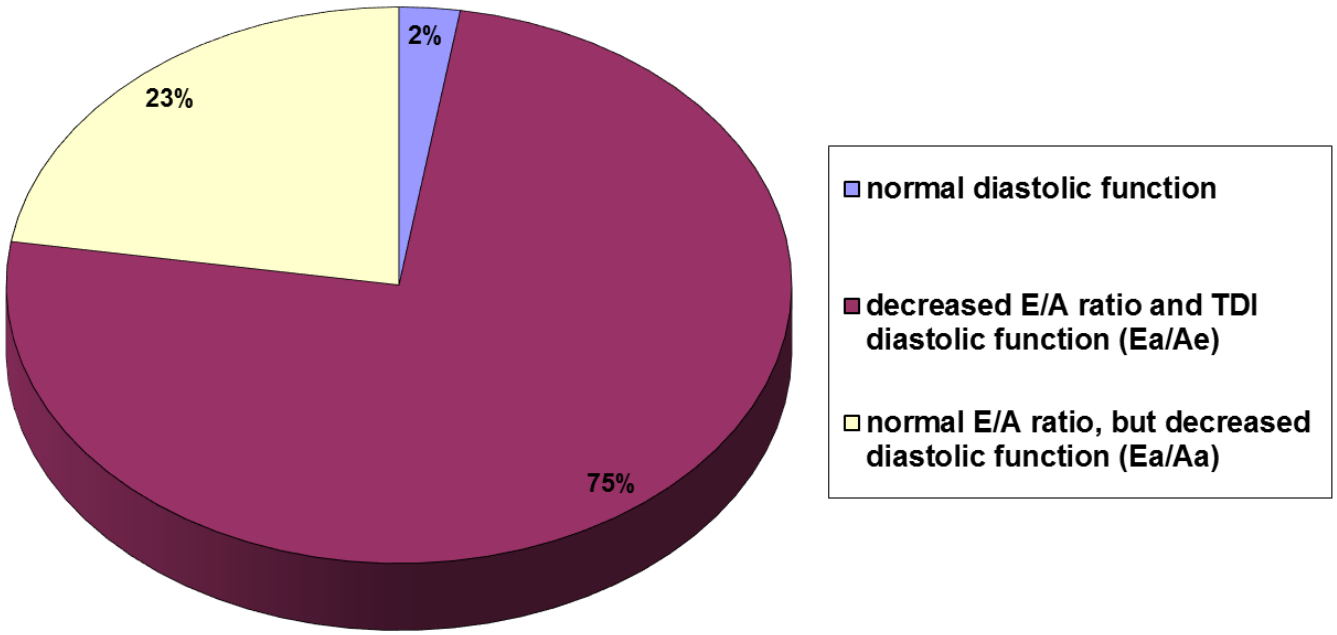

Figure 1. Diastolic function in patients after 1 year

	Begining			After 1 year		
	Patient (n=40)	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*
Septal						
S velocity (cm/s)	9,96 ± 2,21	10,32 ± 1,92	ns	8,18 ± 1,67	10,28 ± 1,96	0,001
Ea diast.vel.(cm/s)	9,97 ± 1,95	10,80 ± 1,78	ns	6,13 ± 1,33	11,18 ± 2,05	0,001
Aa diast.vel.(cm/s)	6,11 ± 1,35	6,25 ± 1,28	ns	9,41 ± 2,23	6,50 ± 1,30	0,001
Ea/Aa ratio	1,65 ± 0,24	1,74 ± 0,21	ns	0,70 ± 0,31	1,72 ± 0,16	0,001
Posterior						
S velocity (cm/s)	7,22 ± 1,59	7,64 ± 1,12	ns	6,54 ± 1,07	7,24 ± 0,95	0,004
Ea diast.vel.(cm/s)	11,68 ± 2,27	11,60 ± 1,70	ns	7,09 ± 2,01	11,36 ± 1,80	0,001
Aa diast.vel.(cm/s)	6,61 ± 1,61	6,29 ± 1,09	ns	9,07 ± 1,95	6,43 ± 1,09	0,001
Ea/Aa ratio	1,81 ± 0,37	1,86 ± 0,26	ns	0,84 ± 0,42	1,77 ± 0,17	0,001
Septal'						
S velocity (cm/s)	9,65 ± 1,81	9,91 ± 1,84	ns	8,41 ± 1,37	9,75 ± 1,65	0,002
Ea diast.vel.(cm/s)	12,23 ± 1,89	13,43 ± 1,95	ns	8,10 ± 1,54	13,61 ± 1,56	0,001
Aa diast.vel.(cm/s)	9,11 ± 1,93	8,81 ± 1,56	ns	11,27 ± 2,04	8,47 ± 1,47	0,001
Ea/Aa ratio	1,38 ± 0,27	1,54 ± 0,21	ns	0,74 ± 0,20	1,62 ± 0,19	0,000
Lateral						
S velocity (cm/s)	10,09 ± 2,14	9,88 ± 1,53	ns	8,87 ± 1,54	9,67 ± 1,66	0,070
Ea diast.vel.(cm/s)	14,21 ± 2,21	14,34 ± 2,79	ns	10,06 ± 2,67	14,38 ± 2,55	0,001
Aa diast.vel.(cm/s)	9,32 ± 1,53	8,92 ± 1,55	ns	11,91 ± 2,38	8,87 ± 1,45	0,001
Ea/Aa ratio	1,54 ± 0,27	1,63 ± 0,32	ns	0,90 ± 0,41	1,63 ± 0,22	0,001
Inferior						
S velocity (cm/s)	10,16 ± 1,72	10,13 ± 1,35	ns	9,12 ± 1,28	9,94 ± 1,28	0,020
Ea diast.vel.(cm/s)	15,12 ± 2,40	14,00 ± 1,82	ns	9,88 ± 2,96	13,93 ± 1,35	0,001
Aa diast.vel.(cm/s)	10,03 ± 1,35	9,41 ± 1,64	ns	12,21 ± 1,91	9,11 ± 1,27	0,001
Ea/Aa ratio	1,52 ± 0,28	1,50 ± 0,17	ns	0,84 ± 0,36	1,54 ± 0,18	0,001
Anterior						
S velocity (cm/s)	8,70 ± 1,55	9,14 ± 1,77	ns	7,98 ± 1,46	8,69 ± 1,63	0,090
Ea diast.vel.(cm/s)	10,77 ± 1,94	11,33 ± 1,40	ns	7,58 ± 2,01	11,75 ± 1,29	0,001
Aa diast.vel.(cm/s)	7,04 ± 1,30	6,90 ± 1,12	ns	9,90 ± 2,27	6,65 ± 1,16	0,001
Ea/Aa ratio	1,55 ± 0,28	1,66 ± 0,23	ns	0,82 ± 0,38	1,79 ± 0,24	0,001

Table 3. TDI measurements at the baseline vs at the end of the study both in the patient group and the control group

This results show that there were no differences between TDI values before chemotherapy compared to the control group. However, values obtained at 1 year show a clear difference between the patient group and the control group in the fibre shortening velocity values in a specific segment, during both systole and diastole. The resulting Ea/Aa ratio showed a clear significant difference in all cases, which characterized the diastolic dysfunction of a specific segment. The baseline E/Ea ratio indicative of left ventricular filling pressure values were also similar in the groups, and the measurement at 1 year detected a measurable difference indicating an increase in the filling pressure in the study group (Table 4).

Obj. 3. Measuring long-term subclinical damage

There were no differences in mean age, risk factors, overall physical parameters, blood pressure and pulse rate between the two groups before the initiation of the chemotherapy (T0). Similarly, there were no significant differences in standard echocardiographic parameters and values obtained using tissue Doppler measurements in either segment. The two groups were similar based on the above-mentioned parameters.

After therapies, the basic circulation parameters at different time points showed only slight changes (Table 5).

		Begining			After 1 year	
	Patient (n=40)	**Control (n=20)**	**p**	**Patient (n=40)**	**Control (n=20)**	**p**
anterior	7,46±1,61	7,00±1,04	n.s.	9,14±2,60	6,53±1,01	0,001
inferior	5,24±1,04	5,70±1,07	n.s.	7,05±1,91	5,49±0,85	0,001
lateralis	5,61±1,20	5,68±1,34	n.s.	6,63±2,13	5,81±1,31	n.s.
posterior	6,97±1,69	6,91±1,34	n.s.	9,83±2,86	6,90±1,68	0,001
septalis	8,16±2,12	7,43±1,41	n.s.	11,06±2,70	6,96±1,28	0,001
septalis'	6,56±1,42	5,95±1,04	n.s.	8,25±1,55	5,62±0,76	0,001

Table 4. E/Ea ratio in all segments referring to the left ventricular filling pressure in the groups before the chemotherapy and one year later

	T0			T1			T2			T3			T4		
	Patient (n=40)	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*
Gender (year)	49.2 ± 10.1	50.1 ± 8.5	ns												
Heart rate (1/min)	78.7 ± 12.6	72.6 ± 9.5	ns	79.7 ± 11.6	75.3 ± 8.7	ns	81.4 ± 11.8	79.1 ± 8.2	ns	82.8 ± 11.3*	76.6 ± 7.6	0.03	78.6 ± 11.8	73.2 ± 15.2	ns
Blood pressure (mmHg)	131/81 ± 16/8	128/77 ± 11/6	ns	133/82 ± 13/8	125/76 ± 11/5	0.02	126/81 ± 12/7*	124/78 ± 11/6	ns	128/78 ± 14/7	124/77 ± 10/4	ns	127/80 ± 10/6	123/80 ± 8/4	ns
Aortic root (cm)	2.64 ± 0.26	2.52 ± 0.36	ns	2.67 ± 0.27	2.63 ± 0.38	ns	2.56 ± 0.30	2.50 ± 0.40	ns	2.59 ± 0.28	2.44 ± 0.27	0.04	2.68 ± 0.26	2.61 ± 0.27	ns
Left atrium (cm)	3.83 ± 0.45	3.97 ± 0.37	ns	3.95 ± 0.51	4.16 ± 0.53	ns	4.05 ± 0.43*	4.06 ± 0.36	ns	4.21 ± 0.52*	4.40 ± 0.87	ns	4.18 ± 0.45*	4.05 ± 0.50	ns
LVEDd (cm)	5.11 ± 0.56	5.09 ± 0.53	ns	5.08 ± 0.47	5.17 ± 0.57	ns	5.08 ± 0.44	5.20 ± 0.54	ns	5.22 ± 0.42*	5.05 ± 0.51	0.05	5.27 ± 0.57*	5.17 ± 0.49	ns
E-septum separatio (cm)	0.50 ± 0.18	0.46 ± 0.12	ns	0.47 ± 0.13	0.44 ± 0.12	ns	0.46 ± 0.12	0.45 ± 0.11	ns	0.46 ± 0.09	0.45 ± 0.09	ns	0.50 ± 0.13	0.43 ± 0.11	ns
EF (Simpson) %	66.9 ± 5.8	64.2 ± 4.7	ns	65.8 ± 5.7	65.4 ± 4.2	ns	67.7 ± 5.4	64.1 ± 3.9	ns	65.9 ± 4.2	67.7 ± 2.8	ns	65.9 ± 3.7	65.3 ± 2.9	ns
Mitral E velocity (cm/s)	78 ± 13	78 ± 09	ns	72 ± 10*	78 ± 10	0.04	69 ± 12*	76 ± 12	0.03	66 ± 10*	76 ± 10	0.002	67 ± 10*	75 ± 14	0.02
Mitral A velocity (cm/s)	62 ± 13	59 ± 12	ns	68 ± 11*	60 ± 08	0.006	73 ± 12*	58 ± 08	0,000	73 ± 10*	57 ± 08	0,000	74 ± 12*	66 ± 22	0.05
E/A ratio	1.30 ± 0.29	1.35 ± 0.26	ns	1.08 ± 0.25*	1.31 ± 0.16	0.001	0.98 ± 0.28*	1.35 ± 0.25	0,000	0.93 ± 0.21*	1.33 ± 0.23	0,000	0.93 ± 0.24*	1.20 ± 0.33	0.001
Deceleration time (ms)	253 ± 49	275 ± 53	ns	267 ± 55	261 ± 44	ns	272 ± 53	298 ± 49	ns	300 ± 39*	280 ± 56	ns	280 ± 47*	302 ± 60	ns
Pulmonaric venous S (cm/	56 ± 10	59 ± 08	ns	48 ± 13*	55 ± 07	0.04	47 ± 12*	53 ± 08	ns	46 ± 11*	57 ± 08	0,000	50 ± 11*	57 ± .09	0.02
Pulmonaric venous D (cm/	36 ± 09	38 ± 08	ns	42 ± 12*	39 ± 07	ns	51 ± 16*	36 ± 08	0,000	49 ± 14*	36 ± 11	0.002	44 ± 12*	38 ± 06	ns
S/D ratio	1.58 ± 0.29	1.57 ± 0.27	ns	1.31 ± 0.77*	1.46 ± 0.25	ns	1.01 ± 0.44*	1.52 ± 0.23	0,000	1.05 ± 0.43*	2.16 ± 0.26*	0.01	1.23 ± 0.43*	1.50 ± 0.29	0.01

* means significant difference in values within the patient group at the end as compared to the initial status. P<0.05

p means the difference between the two goups (patient and control)

Table 5. Basic data and standard echocardiographic parameters of patients

In the study group, the pulse rate increased after the first (T1) and the second (T2) chemotherapy cycle; this increase reached a significant level only during the one year follow-up period (T3), but was not clinically relevant. During the two-year follow-up period (T4), the pulse rate returned to normal.

The blood pressure in the cancer study group showed a mildly significant, but still irrelevant decrease after the second cycle (T2), after which it normalized. In the control group, neither the pulse rate nor the blood pressure showed a substantial change during the two year follow-up period.

We closely monitored the symptoms of circulatory insufficiency. It is clear that neither group had chest complaints or symptoms of circulatory insufficiency. Furthermore, there were no changes in the ECG in the study group.

During the echocardiography, a slight increase was observed in the volume of the left atrium after the second cycle (T2) and of the left ventricle at 1 year (T3) in the study group, which was significant compared to the first measurements. However, there was no substantial change in the ejection fraction in line with the clinical status.

The traditional ultrasound parameters characterized by the diastolic function showed relaxation dysfunction almost right after the first therapy (T1). The peak of the first mitral inflow wave (mitral E wave velocity) began to decrease gradually, and at the same time, the peak of the second wave (mitral A wave velocity) showed an increasing tendency. Accordingly, their ratio (E/A) showed a decrease with a significant change at time point T1. This decrease was detectable during the entire study. The other parameter describing the diastolic function, the two waves of the pulmonary venous curve (pulmonary venous S and D wave) and their mean (S/D) also provided a good measure of DD.

Similarly to the values measured using the traditional method, the Ea and Aa diastolic velocities and their ratio (Ea/Aa), represented by the tissue Doppler echocardiography used to describe the diastolic function, showed a change at time point T1, which remained significant until the end of the study (Table 6).

	T0			T1			T2			T3			T4		
	Patient (n=40)	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*	*Patient (n=40*	*Control (n=20)*	*p*	*Patient (n=40)*	*Control (n=20)*	*p*
Septal															
S velocity (cm/s)	9.96 ± 2.21	10.32 ± 1.92	ns	9.55 ± 2.29	10.63 ± 2.26	ns	9.05 ± 1.91	9.12 ± 1.70	ns	8.36 ± 1.78*	10.54 ± 2.07	0,00	9.45 ± 2.19*	10.85 ± 1.70	0.01
Ea diast.vel.(cm/s)	9.89 ± 2.08	10.80 ± 1.78	ns	7.91 ± 2.71*	10.68 ± 2.13	0,00	6.46 ± 2.11*	9.02 ± 1.69	0,00	6.34 ± 1.59*	11.18 ± 2.05	0,00	6.37 ± 1.96*	10.11 ± 2.04	0,000
Aa diast.vel.(cm/s)	6.19 ± 1.41	6.25 ± 1.28	ns	8.02 ± 2.39*	6.51 ± 1.66	0.01	8.87 ± 2.21*	5.28 ± 1.15	0,00	8.83 ± 2.34*	6.50 ± 1.30	0,00	10.28 ± 2.32*	7.51 ± 2.33	0,000
Ea/Aa ratio	1.63 ± 0.29	1.74 ± 0.21	ns	1.11 ± 0.55*	1.66 ± 0.19	0,00	0.80 ± 0.42*	1.72 ± 0.16	0,00	0.79 ± 0.40*	1.72 ± 0.12	0,00	0.62 ± 0.17*	1.45 ± 0.41	0,000
Posterior															
S velocity (cm/s)	7.22 ± 1.59	7.64 ± 1.12	ns	7.22 ± 1.71	7.73 ± 1.22	ns	6.92 ± 1.33	7.24 ± 1.26	ns	6.76 ± 1.11*	7.39 ± 0.95	0.03	7.42 ± 1.76*	8.24 ± 1.04	0.05
Ea diast.vel.(cm/s)	11.68 ± 2.27	11.60 ± 1.70	ns	9.61 ± 3.44*	10.80 ± 1.91	ns	7.91 ± 2.90*	10.08 ± 1.45	0.003	7.64 ± 2.34*	11.36 ± 1.80	0,00	7.00 ± 2.54*	11.97 ± 2.10	0,000
Aa diast.vel.(cm/s)	6.61 ± 1.61	6.29 ± 1.09	ns	7.75 ± 2.32*	6.24 ± 1.34	0.01	8.15 ± 2.30*	5.52 ± 1.15	0,00	8.33 ± 2.35*	6.43 ± 1.09	0.001	9.58 ± 2.47*	6.68 ± 1.24	0,000
Ea/Aa ratio	1.81 ± 0.37	1.86 ± 0.26	ns	1.36 ± 0.63*	1.77 ± 0.33	0.009	1.12 ± 0.69*	1.87 ± 0.34	0,00	1.06 ± 0.64*	1.77 ± 0.17	0,00	0.78 ± 0.40*	1.82 ± 0.33	0,000
Septal'															
S velocity (cm/s)	9.65 ± 1.81	9.91 ± 1.84	ns	9.20 ± 1.73	10.03 ± 1.37	ns	8.88 ± 1.37*	9.25 ± 1.25	ns	8.72 ± 1.34*	9.75 ± 1.65	0.01	10.13 ± 2.28*	10.12 ± 1.94	ns
Ea diast.vel.(cm/s)	12.23 ± 1.89	13.43 ± 1.95	ns	9.88 ± 2.56*	13.42 ± 1.48	0,00	9.34 ± 2.00*	12.33 ± 1.80	0,00	8.58 ± 1.87*	13.61 ± 1.56	0,00	9.08 ± 2.39*	13.51 ± 2.78	0,000
Aa diast.vel.(cm/s)	9.11 ± 1.93	8.81 ± 1.56	ns	10.47 ± 2.48*	9.13 ± 1.36	0.03	11.27 ± 2.64*	8.02 ± 1.38	0,00	11.01 ± 2.23*	8.47 ± 1.47	0,00	12.05 ± 2.20*	9.23 ± 1.46	0,000
Ea/Aa ratio	1.38 ± 0.27	1.54 ± 0.21	ns	1.02 ± 0.41*	1.49 ± 0.22	0,00	0.87 ± 0.30*	1.56 ± 0.27	0,00	0.82 ± 0.30*	1.62 ± 0.19	0,00	0.78 ± 0.27*	1.51 ± 0.42	0,000
Lateral															
S velocity (cm/s)	10.09 ± 2.14	9.88 ± 1.53	ns	9.71 ± 2.01*	10.29 ± 1.86	ns	9.62 ± 2.38	10.04 ± 1.98	ns	9.09 ± 1.60*	9.67 ± 1.66	ns	9.56 ± 2.26	9.69 ± 1.19	ns
Ea diast.vel.(cm/s)	14.21 ± 2.21	14.34 ± 2.79	ns	12.85 ± 3.30*	14.03 ± 2.73	ns	11.31 ± 3.21*	13.66 ± 2.79	0.007	10.77 ± 2.79*	14.38 ± 2.55	0,00	9.96 ± 2.96*	13.24 ± 3.34	0,000
Aa diast.vel.(cm/s)	9.32 ± 1.53	8.92 ± 1.55	ns	10.48 ± 2.84*	9.34 ± 2.39	ns	11.23 ± 2.94*	8.26 ± 1.83	0,00	11.15 ± 2.70*	8.87 ± 1.45	0.001	12.00 ± 2.56*	8.81 ± 1.85	0,000
Ea/Aa ratio	1.54 ± 0.27	1.63 ± 0.32	ns	1.33 ± 0.54*	1.57 ± 0.41	ns	1.10 ± 0.50*	1.71 ± 0.45	0,00	1.06 ± 0.51*	1.63 ± 0.22	0,00	0.87 ± 0.36*	1.57 ± 0.49	0,000
Inferior															
S velocity (cm/s)	10.16 ± 1.72	10.13 ± 1.35	ns	9.95 ± 1.62	10.28 ± 1.18	ns	9.78 ± 1.33	10.14 ± 1.41	ns	9.40 ± 1.31*	9.94 ± 1.28	ns	9.62 ± 1.20	9.93 ± 1.38	ns
Ea diast.vel.(cm/s)	15.12 ± 2.40	14.00 ± 1.82	ns	12.45 ± 3.09*	14.23 ± 2.32	0.02	11.06 ± 3.56*	13.97 ± 2.33	0.002	10.23 ± 3.30*	13.93 ± 1.35	0,00	10.26 ± 2.77*	14.04 ± 3.03	0,000
Aa diast.vel.(cm/s)	10.03 ± 1.35	9.41 ± 1.64	ns	11.51 ± 2.45*	10.81 ± 2.47	ns	12.42 ± 2.29*	9.23 ± 1.80	0,00	12.16 ± 2.02*	9.11 ± 1.27	0,00	12.78 ± 2.05*	9.45 ± 2.00	0,000
Ea/Aa ratio	1.52 ± 0.28	1.50 ± 0.17	ns	1.15 ± 0.46*	1.38 ± 0.38	ns	0.94 ± 0.43*	1.54 ± 0.27	0,00	0.88 ± 0.40*	1.54 ± 0.18	0,00	0.83 ± 0.33*	1.55 ± 0.48	0,000
Anterior															
S velocity (cm/s)	8.70 ± 1.55	9.14 ± 1.77	ns	8.64 ± 1.89	9.78 ± 2.17	0.04	8.28 ± 1.82	8.43 ± 0.97	ns	8.18 ± 1.18	8.69 ± 1.63	ns	8.73 ± 2.10	8.99 ± 1.49	ns
Ea diast.vel.(cm/s)	10.77 ± 1.94	11.36 ± 1.38	ns	9.70 ± 2.37*	11.75 ± 1.84	0.001	8.63 ± 2.37*	10.16 ± 1.39	0.01	7.94 ± 2.03*	11.75 ± 1.29	0,00	7.42 ± 2.38*	11.52 ± 2.81	0,000
Aa diast.vel.(cm/s)	7.04 ± 1.30	6.90 ± 1.12	ns	8.25 ± 2.04*	7.63 ± 1.76	ns	9.19 ± 2.48*	6.29 ± 1.01	0,00	9.33 ± 2.41*	6.65 ± 1.16	0,00	10.01 ± 2.14*	7.17 ± 1.31	0,000
Ea/Aa ratio	1.55 ± 0.28	1.66 ± 0.23	ns	1.27 ± 0.49*	1.61 ± 0.42	0.01	1.04 ± 0.51*	1.66 ± 0.43	0,00	0.93 ± 0.42*	1.79 ± 0.24	0,00	0.76 ± 0.28*	1.67 ± 0.57	0,000

Table 6. TDI measurements of the 6 segments at different times both in patient and control groups# (septal, posterior, septal', lateral, inferior and anterior indicate the sites of the mitral annulus, where the TDI cursor was placed)

p means the difference between the two groups (study and control)

*P<0.05, means significant difference in values within the study group at the end as compared to baseline.

It should be noted that the velocity of systolic fibre shortening of different segments (S waves) also changed: they showed a decreasing trend from time point T1 through T3, but the values started to improve at time point T4.

It should be noted that in addition to the above mean values, individual analysis of patients showed that 22 patients (55%) had normal diastolic function, and 16 patients (40%) had DD at time point T1 that was detected using the traditional method, while for 18 patients (45%) tissue Doppler imaging had to be used to demonstrate DD (Figure 2). However, by the end of the study (T4) these ratios changed and we succeeded to detect late myocardial damage using some methods in all patients. At the end of the study, we confirmed DD in 27 patients (67.5%) using a joint analysis of the traditional mitral inflow pattern and the pulmonary venous pattern, while in the case of 13 patients (32.5%) only a TDI helped.

The analysis of the E/Ea ratio representing the left ventricular filling pressure in the segments showed a significant increase of the filling pressure in all segments after the second chemotherapy cycle (T2) (Figure 3).

Obj. 4. Preventive effects of physical activity

The two groups of patients (physically active: n=36 - vs physically inactive: n=19) enrolled in the prospective study had a similar age distribution (49.2 vs 50.1 +/-SD) (Table 7). There were no differences between the two groups in the patients' cardiovascular risk status due to the inclusion criteria.
The basic data of these two groups are presented in Table 7.

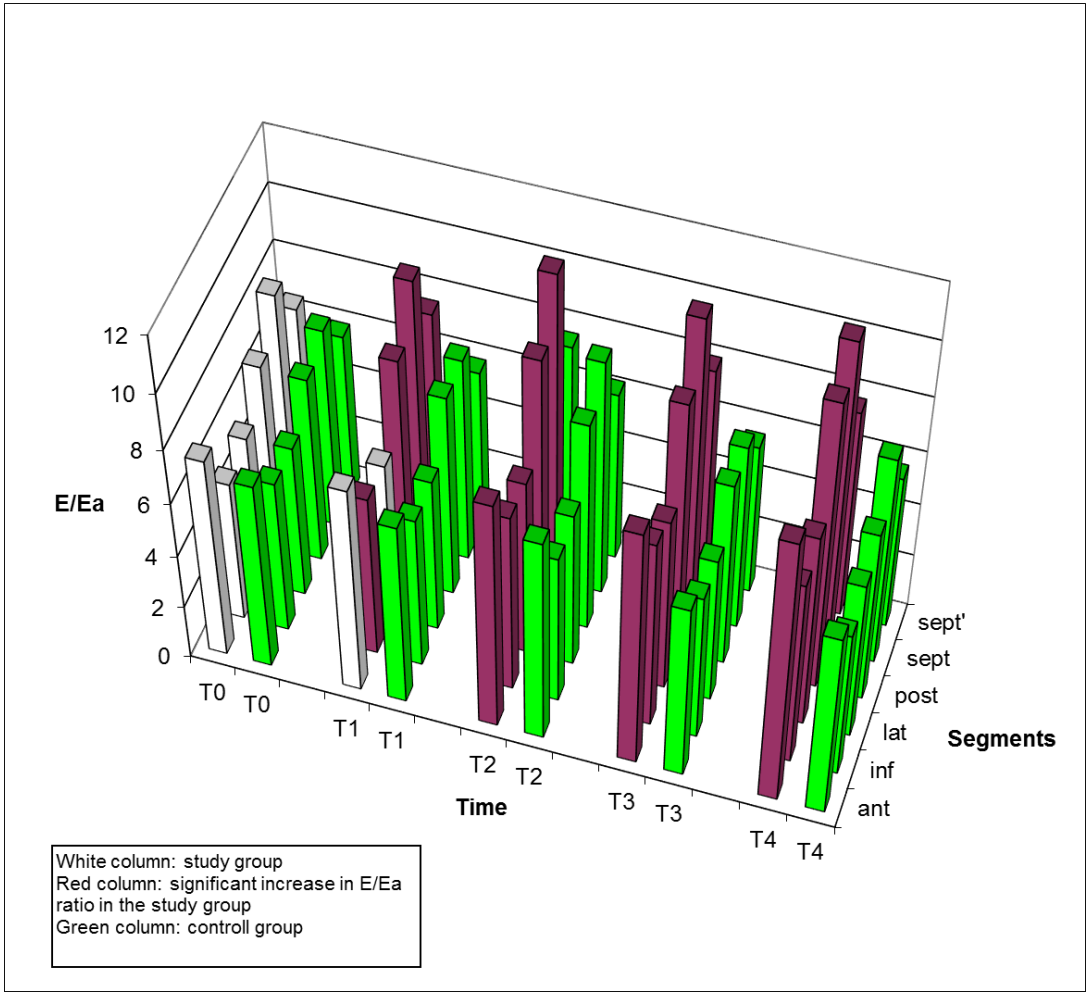

Figure 3. Analysis of E/Ea representing the left ventricular filling pressure

	T1			T2			T3			T4		
	S	NS	p	S	NS	p	S	NS	p	S	NS	p
Mean age (years)	**49.2±10.1**	**50.1±8.5**										
Systolic blood pressure (mmHg)	**129±12$^{\#}$**	**129±18**	**ns**	**130±10$^{\#}$**	**135±14**	**ns**	**124±8**	**128±12**	**ns**	**127±10$^{\#}$**	**128±13**	**ns**
Diastolic blood pressure (mmHg)	**82±8**	**78±8**	**ns**	**83±7**	**81±7**	**ns**	**80±7$^{\$}$**	**80±7**	**ns**	**78±7$^{\$,x}$**	**79±7**	**ns**
Heart rate (1/min)	**78.0±13.1$^{\#}$**	**78.1±13.0**	**ns**	**78.3±11.5$^{\#}$**	**79.3±10.0**	**ns**	**82.5±10.9**	**81.2±14.8**	**ns**	**82.0±11.1**	**81.1±14.3**	**ns**

Table 7.
Demographic data of the patients
S: patients exercising regularly, NS: patients not exercising regularly
p: difference between the two groups at each time point
#: $p<0.05$ vs. T3 among S patients
$: $p<0.05$ vs. T2 among S patients
x: $p<0.05$ vs. T1 among S patients

There were no significant differences between the two groups at baseline (time point T1) or at a later time. The differences observed at various time points within each group, although statistically significant, cannot be considered clinically relevant because both the blood pressure and the pulse rate remained within normal limits.

As regards echocardiographic parameters, there were no significant differences between the two groups and within each group in the aorta width, the end-diastolic diameter of the left atrium and the left ventricle, and the E-septum data (these parameters were omitted from the table). The systolic function of the left ventricle showed a significant change within the physically inactive group (EF T3 vs T2), but this was not clinically relevant either (Table 8).

There were more significant and measurable changes in the diastolic function during this study. One of the parameters of diastolic function, the value (S/D) calculated from the two waves of the pulmonary inflow showed a significant decrease in both groups over time, which indicated an impairment of the diastolic function. The differences between the two groups were not significant, and this value decreased below 1 at time point T3 in both groups. However, the difference became significant at time point T4 compared to time point T1 in both groups. An evaluation of the mitral inflow patterns showed a clear change in the diastolic function. The decrease of the E/A value was visible at time point T2 in both groups, which already decreased below 1 in the physically inactive group, though the difference between the two groups was not yet significant at that time. However, a significant difference was revealed at T3 both between the two groups and compared to time point T1. The E/A ratio in the physically active group decreased below 1 only at time point T4 (Figure 4).

	T1			T2			T3			T4		
	S	NS	p	S	NS	p	S	NS	p	S	NS	p
EF (Simpson, %)	**66.1±5.1**	**68.3±7.6**	**ns**	**64.8±7.5**	**65.9±5.0**	**ns**	**66.2±3.4**	**69.4±5.3^{+}**	**ns**	**64.7±3.5**	**67.8±4.4**	***0.060***
Mitral E velocity (m/s)	**0.76±0.16**	**0.82±0.08**	**ns**	**0.71±0.11**	**0.69±0.12**	**ns**	**0.70±0.12**	**0.67±0.13**	**ns**	**0.67±0.11**	**0.64±0.13**	**ns**
Mitral A velocity (m/s)	**0.59±0.14**	**0.66±0.14**	**ns**	**0.66±0.10**	**0.74±0.11**	0.044	**0.69±0.10**	**0.79±0.11**	0.014	**0.76±0.09**	**0.81±0.10**	**ns**
E/A ratio	**1.30±0.15**	**1.31±0.47**	**ns**	**1.10±0.25**	**0.95±0.22**	**ns**	**1.05±0.28$^{\#}$**	**0.86±0.25***	0.038	**0.89±0.19$^{\#,\$}$**	**0.78±0.16***	**ns**
Deceleration time (ms)	**246±56**	**264±43**	**ns**	**264±55**	**282±54**	**ns**	**287±47**	**264±64**	**ns**	**299±42**	**295±52**	**ns**
Pulmonaric venous S (m/s)	**0.59±0.10**	**0.54±0.10**	**ns**	**0.47±0.14**	**0.49±0.15**	**ns**	**0.46±0.12**	**0.46±0.13**	**ns**	**0.44±0.12**	**0.42±0.09**	**ns**
Pulmonaric venous D (m/s)	**0.38±0.09**	**0.34±0.08**	**ns**	**0.43±0.10**	**0.43±0.16**	**ns**	**0.51±0.14**	**0.56±0.21**	**ns**	**0.53±0.11**	**0.58±0.16**	**ns**
S/D ratio	**1.59±0.33**	**1.60±0.29**	**ns**	**1.21±0.61**	**1.43±0.25**	**ns**	**0.98±0.40$^{\#}$**	**0.94±0.51**	**ns**	**0.87±0.36$^{\#}$**	**0.78±0.32***	**ns**

Table 8.
Echocardiographic parameters of the patients
S: patients exercising regularly, NS: patients not exercising regularly
p: difference between the two groups at each time point
*: $p<0.05$ vs. T1, among NS patients, #: $p<0.05$ vs. T1 among S patients
+: $p<0.05$ vs. T2 among NS patients, $: $p<0.05$ vs. T2 among S patients

The measurements in different segments of the left ventricle allow a more accurate evaluation of the diastolic function, but they have different sensitivity in the detection of the damage (Table 9).

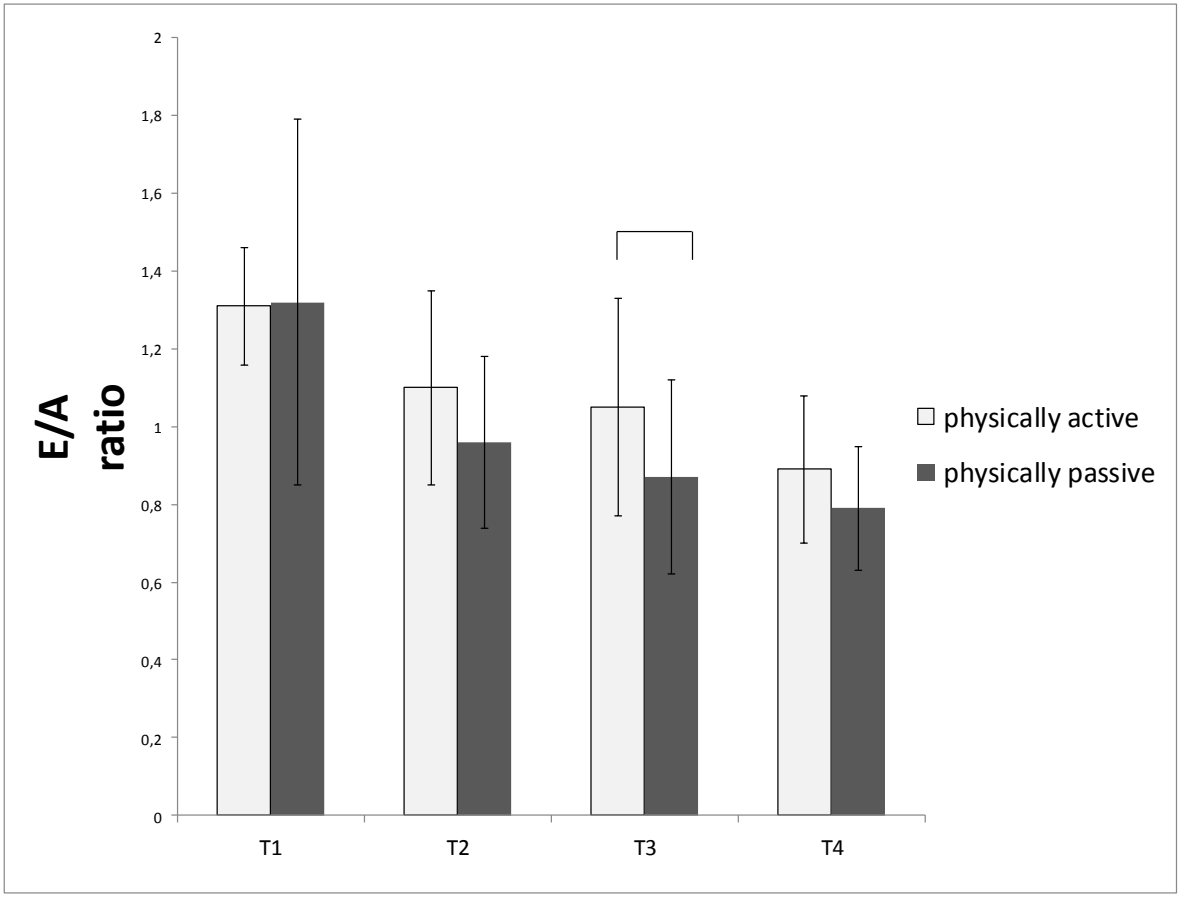

Figure 4.
The E/A ratio in the physically active and physically inactive group at different time points
The black lines between the columns indicate significant differences ($p<0.01$)

	T1			T2			T3			T4		
	S	NS	p	S	NS	p	S	NS	p	S	NS	p
Ea/Aa septal	**1.63±0.23**	**1.67±0.4**	**ns**	**1.10±0.55**	**0.81±0.44**	**ns**	**0.90±0.45**	**0,62±0.23***	0.007	**0.71±0.31#**	**0.61±0.28***	***0.065***
Ea/Aa posterior	**1.82±0.28**	**1.81±0.53**	**ns**	**1.29±0.56**	**1.19±0.53**	**ns**	**1.17±0.78#**	**0.99±0.55***	**ns**	**0.80±0.43#,$**	**0.81±0.45***	**ns**
Ea/Aa septal'	**1.41±0.24**	**1.36±0.23**	**ns**	**0.97±0.42#**	**0.84±0.35&**	**ns**	**0.93±0.35#**	**0.78±0.17&**	**ns**	**0.71±0.14#,$**	**0.63±0.07***	**ns**
Ea/Aa inferior	**1.52±0.32**	**1.49±0.26**	**ns**	**1.19±0.54**	**1.01±0.41**	**ns**	**1.00±0.46#**	**0.79±0.35***	**ns**	**0.87±0.35#,$**	**0.67±0.19***	**ns**
Ea/Aa lateral	**1.57±0.29**	**1.53±0.27**	**ns**	**1.39±0.61**	**1.12±0.49**	**ns**	**1.14±0.54#**	**0.96±0.44***	**ns**	**0.93±0.41#**	**0.81±0.31***	**ns**
Ea/Aa anterior	**1.53±0.32**	**1.51±0.15**	**ns**	**1.18±0.55**	**1.08±0.43**	**ns**	**1.11±0.52#**	**0.96±0.54***	**ns**	**0.78±0.33#,$**	**0.70±0.32***	**ns**
E/septE	**7.97±2.41**	**8.70±2.71**	**ns**	**9.43±2.68**	**11.42±3.23**	***0.09***	**10.34±2.61#**	**12.77±3.52***	0.012	**11.25±3.26#**	**11.00±3.03**	**ns**

Table 9.
TDI parameters of the patients in different segments
S: patients exercising regularly, NS: patients not exercising regularly
p: difference between the two groups at each time point
*: $p<0.05$ vs. T1, among NS patients
#: $p<0.05$ vs. T1 among S patients
$: $p<0.05$ vs. T2 among S patients
&: $p=0.059$ vs. T1 among NS patients

In this study, the most sensitive measurement in the detection of the damage to the diastolic function was the measurement performed in the sept and sept' segments. In the physically inactive group the Ea/Aa ratio decreased below 1 at time point T2 in the sept segment, while in sept' segment a value below 1 was detected at the same time point in both groups. At time point T3, this value was below 1 in all segments in the physically inactive group, while the same value decreased below 1 only in the sept' and the sept segments in the physically active group. At time point T4, a value below 1 was detected in both groups and all segments.

As regards the change of the parameters over time, the Ea/Aa ratio showed a decreasing trend in all segments in both the sporting and the physically inactive group. This decrease became significant at time point T3 compared to baseline (T1) in both groups, but this significance was confirmed at time point T4, as well. The Ea/Aa value showed a significant decrease at time point T4 compared to time point T2 in the inf, sept', post and ant segments in the physically active groups, which indicates that the diastolic function remained intact for a longer period of time in the physically active group.

A comparison of the values of the two groups showed that the Ea/Aa values were lower in the physically inactive group than in the physically active group in all segments beginning with time point T2. This difference was significant only in the septal segment at time point T3, while a tendency difference was detected in the same value at time point T4 (p=0.065) (Figure 5).

The E/Ea sept value, which is a good indicator for the left atrial filling pressure exceeded 10 sooner in the physically inactive group, at time point T2. This limit was reached in both groups at time point T3 and became significant compared to time point T1 (Figure 6).

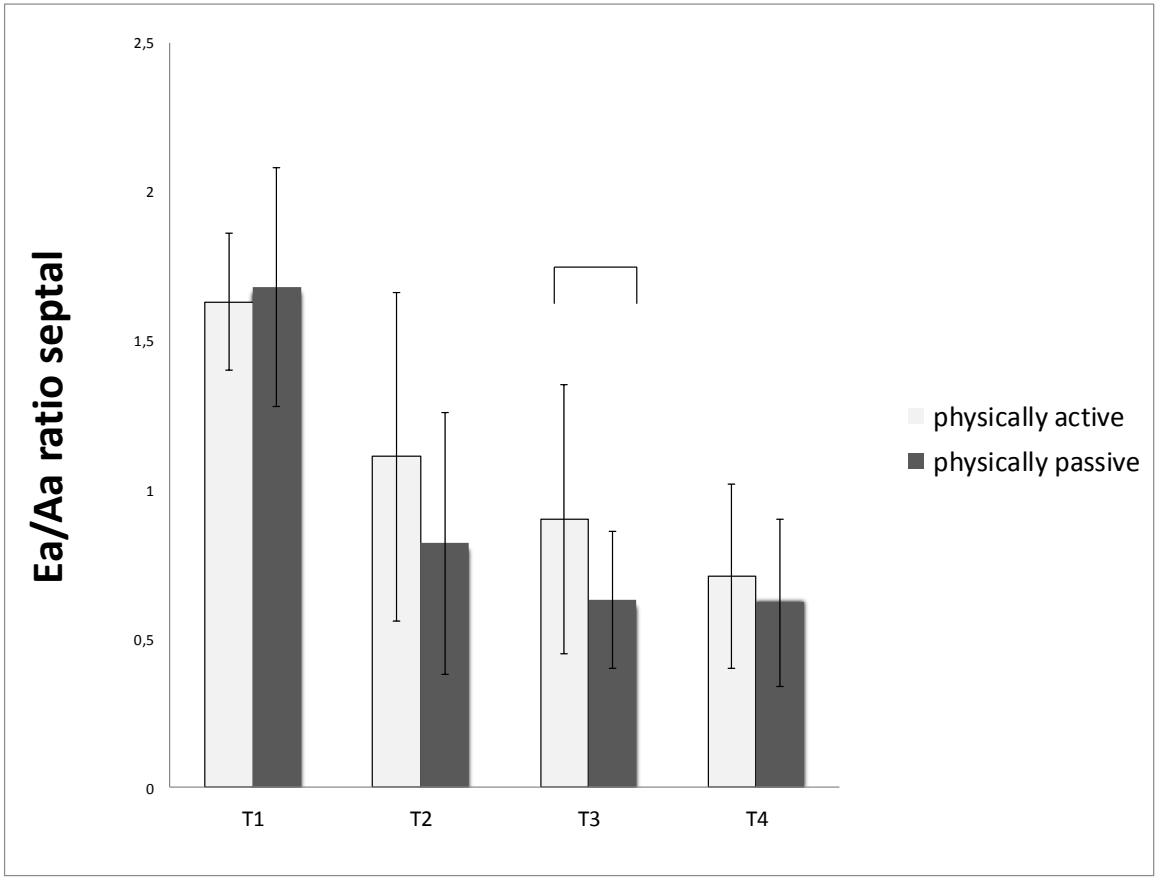

Figure 5.
The Ea/Aa ratio in the physically active and physically inactive group at different time points
The black lines between the columns indicate significant differences (p<0.01)
The dashed lines between the columns indicate non-significant differences (p<0.065)

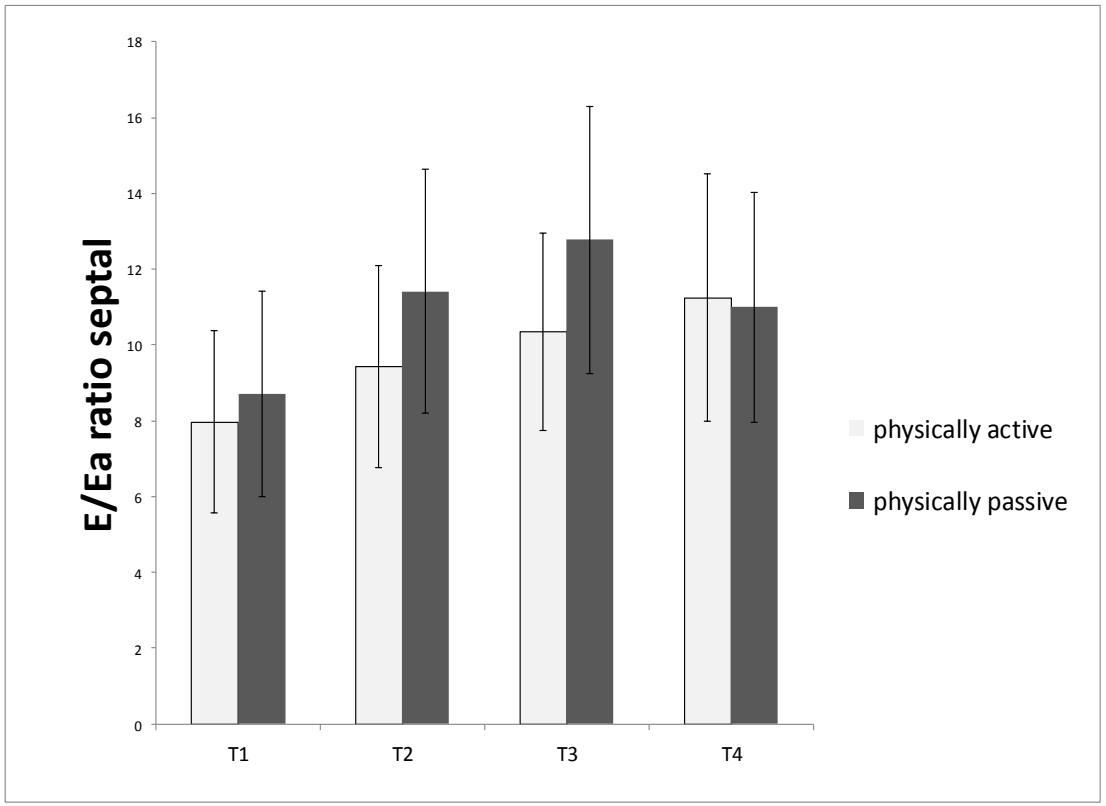

Figure 6.
The E/Ea ratio in the physically active and physically inactive group in different times, which is a good indicator of left atrial filling pressure
The black lines between the columns indicate significant differences (p<0.01)
The dashed lines between the columns indicate non-significant differences (p<0.065)

This elevation was more marked in the physically inactive group and was significant compared to time point T2. This ratio remained above 10 in both group at time point T4.

Five years after the short term tests, the patients have been asked by phone. According to the responses given, the symptoms of cardiac failure developed in 18.1% of physically active patients while in the case of physically inactive patients, this proportion was significantly higher, 69.2% (Figure 7). 81.2% of physically active patients and only 30.8% of physically inactive patients were free of symptoms.

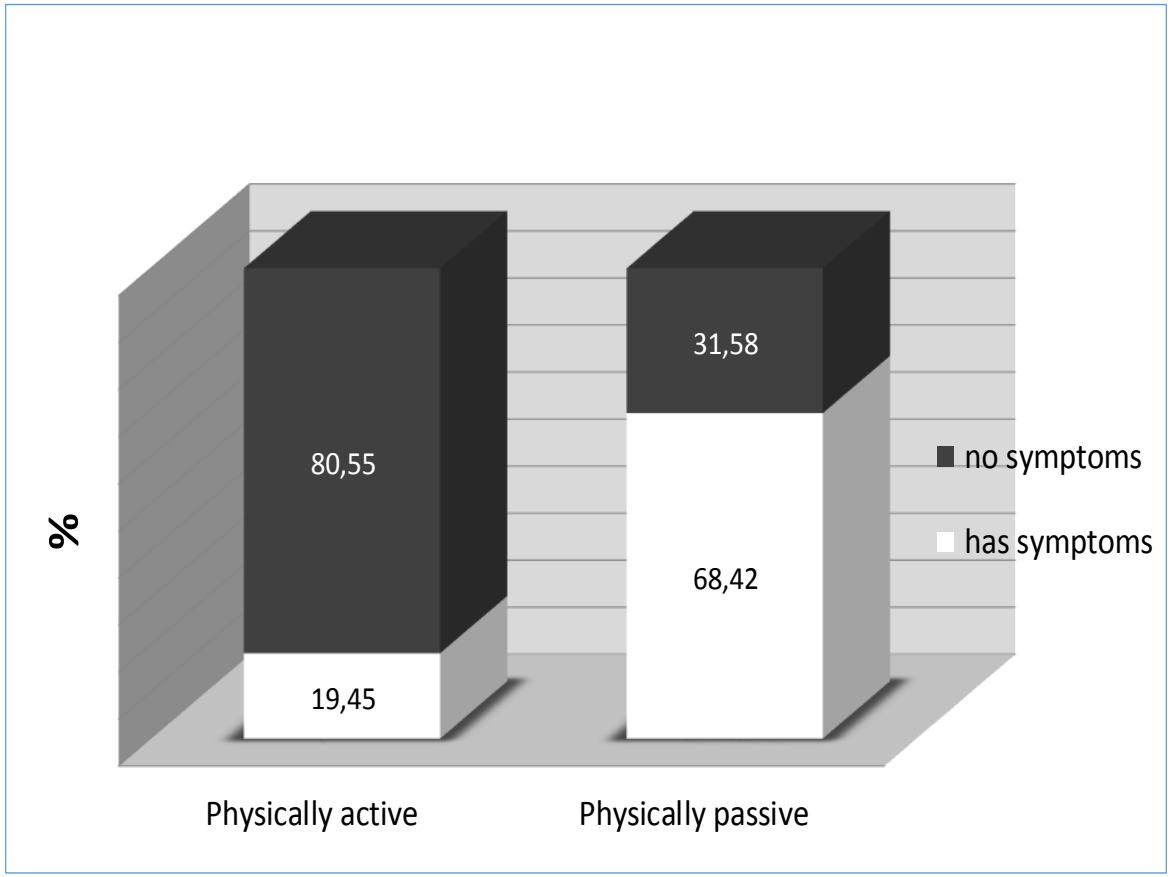

Figure 7.
Incidence of symptoms characteristic to heart failure in the physically active and physically inactive group after 5 years the completion of the short-term assessments, based on phone interviews.
White column means the proportion of patients with any symptoms
Black column means the proportion of patients free of symptoms

Discussion

Anthracyclines have long been used as part of the adjuvant chemotherapy in the treatment of patients with breast cancer. The cardiotoxicity caused by these agents have been described for several decades (1, 2). Clinically relevant cardiotoxicities include congestive heart failure, malignant arrhythmias, impulse generation and conduction disorders, and sudden cardiac death, which can develop years or even decades after the treatment (29, 30, 31, 32, 7). These entities have been addressed by a number of synthesis studies and prior guidelines (33, 34). One of the most common and most investigated form of cardiotoxicities is myocardial damage (left ventricular dysfunction and heart failure), which received particular attention because it is associated with a far greater mortality than any cardiomyopathy of other etiology (35). However, it should also be noted that the proportion and relevance of risk factors that contribute to the development of cardiotoxicity is much higher in real life in tumour patients than in the well-defined conditions of prospective randomized studies (see the Introduction). Patients enrolled in the planned studies do not represent the average population of tumour patients because patients with severe comorbidities or cardiovascular conditions, as well as too young or too old patients were excluded right from the start. Indeed, the retrospective population data show a much higher incidence both for cardiomyopathy and heart failure (15.5-41.9%) (36). Anthracyclines had been long seen as the dominant cause of cardiotoxicity; however, these adverse events are increasingly observed with the newest antitumour treatments, as well. Thus, targeted molecular tumour therapies, including monoclonal antibodies and tyrosine kinase inhibitors, have also “signed up” as cardiotoxic agents (37).

At the beginning, in the 1960s cardiotoxicities were confirmed by establishing the diagnosis of heart failure. Echocardiography became available for routine examination only in 1980s and was used to describe the severity of toxic damage on the basis of the change in the left ventricular EF for several decades. This non-invasive testing method became the gold standard in the detection of cardiotoxicities as well. Nevertheless, due to the heart’s substantial reserve capacity, the systolic function (LVEF) showed a significant decrease that can be easily detected using ultrasound scan only years or decades after the toxic damage caused by the treatment. These data have become

apparent for the first time from observations performed on survivors of childhood cancer (38).

Attention has been increasingly focused on the early detection of late left ventricular dysfunction. We have mentioned the follow-up of FS changes in the Introduction. A slight decrease in EF observed during repeated LVEF measurements may already indicate the damage that potentially led to heart failure at the end (39).

Early detection of subclinical cardiotoxicity and prevention of the resulting heart failure has always been a great challenge in oncology care. It has become increasingly obvious that the improvement of non-invasive diagnostic methods led to an increase in the incidence of asymptomatic myocardial damage caused by cumulative low doses of anthracycline. However, late damages show an increasing incidence, and can develop even decades after the treatment causing often unmanageable heart failure (40).

The rate of subclinical cardiotoxicity detected with traditional methods is estimated by various papers to be between 20% and 75% (51, 30, 52, 7, 53, 54). In addition to limiting the dose of the anthracyclines used and amending the therapeutic protocol, the attempt to minimize the extent of the myocardial damage also required routine cardiac monitoring and detailed knowledge of the cardiovascular risk status. The incidence of heart failure related to the anthracycline treatment is 15-20% and 2% in patients with risk factors and patients without risk factors, respectively (55). These data emphasise not only the importance of evaluating cardiovascular status and systematic follow-up, but also require a method which can be used to detect cardiotoxicity as early as possible.

The best method to prevent cardiotoxicity is early treatment of the subclinical myocardial damage confirmed in due time. The first stage of the cardiac damage is the diastolic damage. The most commonly used method to evaluate the diastolic function, available in routine clinical settings, is echocardiography. Of the traditional heart ultrasound parameters those suitable to evaluate the relaxation disorder using the M mode are as follows: the extent and rate of the volume fluctuation in the ventricular systolic and diastolic function, the change in the wall thickness in diastole, and the duration of the isovolumetric relaxation. Doppler echocardiography can be used to measure mitral filling velocity in early and late diastole. As noted in the Introduction, the detection of diastolic dysfunction using the traditional Doppler mitral inflow

imaging is impacted by a variety of factors. The change in velocity depends on the change in the preload, afterload and heart rate as well, and therefore the diagnosis of diastolic dysfunction and diastolic heart failure may not be based solely on the E/A definition.

During recent years, new methods have been investigated in order to determine diastolic dysfunction more accurately. One technique that can be very easily performed and is becoming increasingly accessible is tissue Doppler imaging (TDI), which measures and represents the moving velocity of a specific segment (most commonly the septal and lateral segment) proximal to the mitral annulus as a function of time. The resulting curve is the tissue Doppler curve, where the first positive wave (S wave) shows the contraction rate of myocardium fibres during systole, and then, in the next phase, the first negative, normally greater wave (Ea wave) results from the rate of relaxation of the myocardium fibres during diastole. The third negative, smaller wave (Aa wave) is a curve of the additional relaxation caused by the atrial contraction. The underlying principle is the measurement of the contraction and relaxation velocity of subendocardial longitudinal fibres. The ratio of the two negative waves (diastolic waves, Ea/Aa) is a representation of the diastolic function just as the traditional mitral inflow curve (56). Furthermore, it is important to mention the mitral E/Ea ratio used to evaluate the left ventricular filling pressure, which contributes to the exploration of the diastolic dysfunction by measuring high filling pressure, irrespective of the systolic function. As far as we know, a value less than 8 excludes, while a value above 15 confirms the presence of elevated filling pressure with certainty, which is caused by an impairment of the relaxation of the heart, that is, diastolic insufficiency. There is a grey area between the two above-mentioned values, where additional tests are needed to explore the cause of the difference (57).

Several studies investigated the development of diastolic dysfunction after chemotherapy (51, 58). It is known that the diastolic dysfunction precedes the development of the systolic dysfunction. In addition, isolated diastolic dysfunction may also develop as a result of the cardiotoxicity, which can lead to heart failure after a long latency period. The above findings reflect the importance of the measurement and follow-up of this parameter in relation to the treatment of malignancies.

The tissue Doppler echocardiography has been an innovation since it made possible that the regional alterations be detected in an early stage, before the development of any change in the global function. The relaxation properties of the myocardium are complex and are determined by the internal processes and the structure of the heart (such as active and passive flexibility) and other external factors (such as right ventricular pressure, the filling pressure, that is, preload, afterload, heart rate, pericardial and pleural pressure). Contrary to the standard Dopple echocardiography, TDI can measure myocardial tissue velocity during both systole and diastole, which directly describe myocardial contractility and relaxation properties (59).

Obj. 1. This study aimed at detecting the diastolic dysfunction to help early detection of subclinical cardiotoxicity using both traditional echocardiography and tissue Doppler imaging by comparing the accuracy of these methods.

Most of the times, the symptoms, the possible heart failure and arrhythmias cannot help in the early recognition of the subclinical cardiotoxicity. In this study, none of the patients developed circulatory failure during the one-year follow-up.

Based on the heart rate measurement, it can be established that the initial equivalency between the study group and the control group disappeared and a significantly higher pulse rate was measured in the study group after one year. During the last measurements, the higher heart rate is likely to be caused by cardiotoxicity. While the post-treatment tachycardia is clearly demonstrated in the acute phase (60), possible arrhythmias are more likely to be caused by the myocardial damage, cardiomyopathy, the remodelling developed (53) and the associated sympathicotonia.

There were no differences in the blood pressure neither in the control group nor in the study group, and thus, blood pressure as independent risk factor cannot lead to diastolic dysfunction. We found no literature data on whether chemotherapy-induced chronic myocardial damage is associated with blood pressure.

As expected, we detected no toxic effect that directly influences the systolic function during the treatment in line with the literature data. Nevertheless, compared to literature data, a new finding is that cardiotoxicity (diastolic dysfunction) can be confirmed using objective parameters 6 months after the treatment in a patient group with no other comorbidity.

In female patients treated with chemotherapy, without cardiovascular risk factors, changes indicative of cardiotoxicity not yet associated with clinical symptoms could be detected very early, after only one year after the initiation of the treatment, using TDI. For some parameters, objective changes can be detected even before the deterioration in the clinical status of the patients.

The extended study attempted to find out whether there will be any changes in the above-mentioned diastolic dysfunction that can be detected only one year after chemotherapy, two years after the initiation of this therapy, and whether the systolic function deteriorates two years after the therapy. Tassan-Mangina *et al.* found that after anthracycline-containing therapy in patients with different tumours, early diastolic dysfunction can be detected using TDI, followed by systolic dysfunction in some cases after many years (41).

The difference in the heart rate found at 1 year disappeared by the end of the second year, and which probably was a reversible process, that is, transient tachycardia.

There was no clinically relevant difference in blood pressure between the two groups, and so blood pressure, as a separate risk factor, could not lead to diastolic dysfunction.

However, the measurements showed a difference in the size of the left ventricle and the left atrium after the first year, and this significant difference remained unchanged until the end of the study. This is only a small change in the size, which was not associated with either a clinical deterioration of the circulatory status or a significant change in the pump function. Similarly to our study, several authors described a non-significant enlargement (61, 62), a process that was not followed by a substantial deterioration of the ejection fraction in our case.

A comparison of the traditional method and the new method used to measure diastolic dysfunction showed that both methods are suitable to detect late-onset subclinical cardiotoxicity (diastolic dysfunction) after anthracycline-containing chemotherapy in young female patients without risk factors. Both methods confirmed significant changes in the traditional parameters (E, A, E/A, S/D) and the TDI parameters (Ea, Aa, Ea/Aa) used to detect diastolic dysfunction.

Several publications (41, 63) confirmed the early decrease in the mitral annulus Ea velocity in patients treated with anthracycline, which remained low both during and

after the treatment, as also shown by the results of this study. The decrease in segmental Ea was heterogenous, assuming different levels of stress, apoptosis and fibrosis (64).

The advantages of the modern TDI method were clearly revealed by the individual analysis of the patients. Figure 2 clearly shows that TDI is more a sensitive method for it could detect diastolic dysfunction in 18 patients (45%) in contrast to the 16 patients (40%) evaluated using the traditional method. The difference in the sensitivity of the two methods became even more obvious at time point T4 (27 versus 40 patients). If we also take into account another advantage of the TDI method that it does not depend on a number of factors and the measurement presents a far less technical challenge, no one should have problems with accepting it as part of routine diagnostics.

As mentioned above, the detection of the diastolic dysfunction plays an important role in confirming cardiotoxicity. The diastolic dysfunction is an individual clinical entity, similarly to diastolic heart failure.

For more than a decade, there has been a big debate about the detection and importance of diastolic dysfunction. A joint position of the European Association of Echocardiography (EAE) of the European Association of Cardiology (EAC) and the American Society of Echocardiography (ASE) addresses in detail the possibilities to analyse and detect the diastolic function. In addition to traditional methods (E/A, DT, S/D, left atrial volume), it emphasises the role of the tissue Doppler imaging (Ea, E/Ea), and considers that these methods should be used together (65). It also notes the role of myocardial strain in measuring regional contractility and diastolic function. A position published by the above-mentioned associations in 2014 addresses in detail the possibilities of using multimodal testing of patients with tumour (66). This position provides a precise definition of cardiotoxicities and the methods to be used to detect them. Furthermore, it emphasises the role of echocardiography which is non-invasive method that can be repeated in unlimited times. In the monitoring of the systolic function, it gives preference to 3D EF measurement, but where it is not accessible, it allows for the use of 2D EF measurement. However, in this case it is important which method is used to calculate EF for it primarily recommends the Simpson method. Due to high intraobserver variability, the same examiner should follow-up the left ventricular function of a specific patient at different time points, whenever possible. In this study, all above-mentioned conditions have been met.

According to this position, in the detection of subclinical left ventricular dysfunction, considered to be an early marker of cardiotoxicity, in addition to a decrease in LVEF, a decisive factor is the change in the global longitudinal strain. If this change exceeds 15%, the damage caused by the antitumour therapy can be established.

According to the current recommendations, it can be stated that in the early detection of cardiotoxicity is based primarily on the 3D EF measurement and the change in the global longitudinal strain (GLS), but these methods are barely accessible in clinical settings. An acceptable alternative for detecting myocardial damage caused by the antitumour treatment is an accurate 2D EF measurement and diastolic parameters measurement. Nevertheless, there is limited evidence available on whether the diastolic parameters have prognostic power for cardiotoxicity.

An additional large-scale study is required in order to demonstrate the above.

Obj. 4.

This study demonstrated that the deterioration of the diastolic function could be detected in patients with intact heart after the initiation of chemotherapy both in the fit (physically active) and the unfit (physically inactive) groups; in the physically inactive group this deterioration occurred earlier and it could be detected after only one year, while in the physically active group it could be detected only after two years. The difference in the diastolic function between the two groups became significant after one year. This study evaluated the function of different segments and found that the septal segment is the first to be damaged in both groups. In the physically inactive patients, there were significantly more cases with symptoms indicative of heart failure based on the phone interview conducted 5 years after the completion of the short term studies.

This study included 55 young women with breast cancer. These patients were, in part, patients from earlier studies, and in part, patients enrolled after these earlier studies. During patient enrolment, exclusion criteria were traditional cardiovascular risk factors and radiation of the left chest, and so the changes in the left ventricular function during chemotherapy were side effects of the medication. The patients were examined at 4 time points. The echocardiographic parameters measured before starting the treatment, at time point T1, were considered baseline values. Additional study time points

corresponded to the development of the subacute (T2), early chronic (T3) and late chronic (T4 and the time of the phone interview) cardiotoxic damage.

As regards the diastolic function, the value (S/D) calculated from the two waves of the pulmonary inflow showed a significant decrease in both groups over time, which indicated an impairment of the diastolic function. The differences between the two groups were not significant, and this value decreased below 1 at time point T3 in both groups. However, the difference became significant at time point T4 compared to time point T1 in both groups.

During the tissue Doppler imaging (TDI), as regards the change of the parameters over time, the Ea/Aa ratio showed a decreasing trend in all segments in both the physically active and physically inactive groups. This decrease became significant at time point T3 compared to baseline (T1) in both groups, but this significance was confirmed at time point T4, as well. A study published by Lisi et al. presented similar findings, according to which TDI is more sensitive in the detection of diastolic dysfunction than the traditional E/A measurement (67).

The Ea/Aa value showed a significant decrease at time point T4 compared to time point T2 in the inf, sept', post and ant segments in the physically active groups, which indicates that the diastolic function remained intact for a longer period of time in the physically active group.

The measurements in different segments of the left ventricle allow a more accurate evaluation of the diastolic function, but they have different sensitivity in the detection of the damage (Table 9). A comparison of the values of the two groups showed that the Ea/Aa values were lower in the physically inactive group than in the physically active group in all segments beginning with time point T2. This difference was significant only in the septal segment at time point T3, while a tendency difference was detected in the same value at time point T4 (p=0.065) (Figure 5).

In this study, the most sensitive measurement in the detection of the damage to the diastolic function was the measurement performed in the sept and sept' segments. In the physically inactive group the Ea/Aa ratio decreased below 1 at time point T2 in the sept segment, while in sept' segment a value below 1 was detected at the same time point in both groups. At time point T3, this value was below 1 in all segments in the physically inactive group, while the same value decreased below 1 only in the sept' and the sept

segments in the physically active group. At time point T4, a value below 1 was detected in both groups and all segments.

The sensitivity of TDI measurements was confirmed by the fact that a decreased Ea/Aa ratio could be measured in a specific segment (sept') as early as the time of the subacute damage in the physically active group, as well. The velocity profile of the mitral annulus measured by tissue Doppler imaging method is a commonly used technique in the determination of global left ventricular function. Due to the direction of the Doppler measurement, the rate of the radial shortening and dilation of the left ventricle is the best suitable to measure global left ventricular function, irrespective whether the measurement is performed in either the lateral or septal segment. Nevertheless, similar studies showed no differences in the shortening rate of different segments after treatment with medium doses of anthracycline (41).

The E/A ratio in the physically active group decreased below 1 only at time point T4 (Figure 4). In the physically inactive group, diastolic dysfunction could be measured as early as the expected time of the subacute damage (T2) (see Tables 8 and 9). In contrast, in the physically active group diastolic dysfunction (65) defined according to the ESC Guideline became apparent only in measurements performed after one year (T3-T4). Similar dynamics was observed in connection to the change in the left atrial filling pressure. The E/Ea sept value, which is a good indicator for the left atrial filling pressure exceeded 10 sooner in the physically inactive group, at time point T2. This limit was reached in both groups at time point T3 and became significant compared to time point T1 (Figure 6). This elevation was more marked in the physically inactive group and was significant compared to time point T2. This ratio remained above 10 in both group at time point T4. Establishing the importance of the left ventricular filling pressure between 8 and 15 requires further evaluation. This study measured the elevation of the filling pressure; however, it did not confirmed signs indicative of more serious damages which could have caused the elevation of the filling pressure. The change in the filling pressure in this patient group has not been evaluated yet.

During the longer term follow-up (5 years after the completion of the chemotherapy), there were significantly more cases with symptoms of heart failure in the physically inactive group, which may have an impact on quality of life and life expectancy.

Based on the phone interview conducted 5 years after the completion of the short term studies, symptoms of heart failure occurred in 19.45% of physically active patients and 68.42% of physically inactive patients, that is, in significantly more cases (Figure 7). Conversely, 80.55% of physically active patients were free of symptoms, while only 31.58% of physically inactive patients remained free of symptoms. The positive physiological effects and function of physical activity in ameliorating the cardiovascular risk have been long known. Numerous epidemiological studies demonstrated the relationship between regular physical activity and the risk of malignancies (68). The studies published over the last decade provided evidence that physical activity plays an important role in the prevention of breast cancer (69). In addition to primary prevention, regular physical activity may also play an important role in the secondary prevention, that is, the prevention of cardiotoxicities occurring during the chemotherapy administered in the treatment of malignancies. Several animal models demonstrated the protective effect of regular aerobic physical exercise on the functional, histological and molecular changes caused by doxorubicin and the increase of mortality (70). The positive/preventive effects of physical activity are explained by several physiological factors (70). A number of human clinical studies also demonstrate the positive effect of physical activity performed during and after the antitumour treatment on the cardiac and pulmonary function and quality of life. Based on the results of Schneider *et al.*, physical activity performed during and after chemotherapy had a positive impact on the systolic and diastolic tension, heart rate and pulmonary function in patient with breast cancer. In addition, it also had a positive effect on fatigue and exhaustion associated with the disease, as well as certain sensory and cognitive functions (71). Courneya *et al.* reached a similar conclusion in post-menopausal female patients in the case of physical training performed after chemotherapy and radiation (72).

The Framework PEACE study distinguishes six key stages of malignancies (pre-screening, screening, pre-treatment, treatment, post-treatment and resumption), in each of which the positive effect of physical activity can be demonstrated. During pre-screening and screening, exercise can help with the diagnosis of malignancies by directly influencing the sensitivity and specificity of the method used to detect them, and by indirectly improving the adherence to cancer screening. Furthermore, one of the positive effects of physical activity is that it improves the physical fitness of cancer patients before the treatment. During the treatment, it ameliorates the side effects (such

as fatigue, pain, nausea and depression). After the treatment, physical activity is an efficient facilitator of the rehabilitation (73). This study was conducted both during and after the treatment, and the positive effects of physical activity were observed in both cases.

Conclusions - Theses

Ad 1. Compared to literature data, a new finding is that cardiotoxicity (diastolic dysfunction) can be confirmed using objective parameters 6 months after the treatment in a patient group with no other comorbidity or cardiovascular risk.

Ad 2. In female patients treated with chemotherapy, without cardiovascular risk factors, changes indicative of cardiotoxicity not yet associated with clinical symptoms could be detected very early, after only one year after the initiation of the treatment, using TDI. For some parameters, objective changes can be detected even before the deterioration in the clinical status of the patients.

Ad 3. A comparison of the traditional method and the new method used to measure diastolic dysfunction showed that both methods are suitable to detect late-onset subclinical cardiotoxicity (diastolic dysfunction) after anthracycline-containing chemotherapy in young female patients without risk factors. (One or more years later).

Ad 4. Our additional studies confirmed the positive effects of physical activity in the prevention of cardiotoxic damage

Acknowledgements

First of all I wish to express my special thanks to my supervisor Professor Tamás Forster director of the Second Department of Internal Medicine and Cardiology Center, University of Szeged for his support and scientific guidance of my work.

I am grateful to my co-authors, Edina Tolnay, Zsuzsanna Cserép and Petra Gulácsi-Bárdos for their sustained work and permanent support. I would like especially to thank Tamás Nagykálnai for his enthusiastic professional support and motivation who has encouraged and inspired me since the beginning.

I am very grateful to Professor László Hangody for his valuable remarks and recommendations.

I have to thank Mária Olajos and Andrea Gondi for their committed help in data input.

I have to thank Péter Varga for his help with the statistical calculations.

I'm very thankful to the physicians, nurses, administrators and assistants of the Department of Internal Medicine and Cardiology I of the Uzsoki Hospital for their support and patience during the last years.

Last but not least, allow me to express my gratitude to my wife, Margit Tóth, my children, my parents and my friends for their support, strength and encouragement during the last several years.

References

1. *Tan C, Tasaka H, Yu KP, et al.: Daunomycin, an antitumour antibiotic, in the treatment of neoplastic disease: clinical evaluation with special reference to childhood leukemia. Cancer 1967; 20(3): 333-53.*
2. *Lefrak EA, Pitha J, Rosenheim S,et al.: A clinicopathologic analysis of adriamycin cardiotoxicity. Cancer. 1973 Aug;32(2):302-14.*
3. *Wheeler DM: Another hypothesis concerning the antitumour activity and cardiotoxicity of daunorubicin (NSC-82151) and adriamycin (NSC-123127). Cancer Chemother Rep. 1975 Mar-Apr;59(2 Pt 1):258-9.*
4. *Deborah L. Keefe: Anthracyclin-Induced Cardiomyopathy. Seminars in Oncology. 2001 August;28(4):2-7.*
5. *Bristow MR, Thompson PD, Martin RP, et al.: Early anthracycline cardiotoxicity. Am J Med. 1978 Nov;65(5):823-32.*
6. *Villani F, Beretta G, Guindani A.: Evaluation of early doxorubicin-induced cardiotoxicity by means of systolic time intervals. Cancer Chemother Pharmacol. 1979;3(4):249-51.*
7. *Steinherz LJ, Steinherz PG, Tan C.: Cardiac failure and dysrhythmias 6-19 years after anthracycline therapy: a series of 15 patients. Med Pediatr Oncol. 1995 Jun;24(6):352-61.*
8. *Vinita BP, Milap CN: Cardiotoxicity of Chemotherapeutic Agents. Drug Safety 2000 Apr; 22(4): 263-302.*
9. *Lotrionte M, Biondi-Zoccai G, Abbate A, et al.: Review and meta-analysis of incidence and clinical predictors of anthracycline cardiotoxicity. Am J Cardiol. 2013 Dec 15;112(12):1980-4. doi: 10.1016/j.amjcard.2013.08.026. Epub 2013 Sep 25.*
10. *Hequet O, Le QH, Moullet I, et al.: Subclinical Late Cardiomyopathy After Doxorubicin Therapy for Lymphoma in Adults. J Clin Oncol 22:1864-1871.*
11. *Bu'Lock FA, Mott MG, Oakhill A, et al.: Early identification of anthracycline cardiomyopathy: possibilities and implications. Arch Dis Child. 1996 Nov;75(5):416-22.*
12. *Burstein H, Winer E: Primary care for survivors of breast cancer. N Engl J Med, 2000; 343 (15), 1086-1094. Review.*

13. *Salminen E, Syvanen K, Korpela J et al.: Docetaxel with epirubicin--investigations on cardiac safety. Anticancer Drugs. 2003 Jan; 14(1): 73-7.*
14. *Cardinale D, Sandri MT, Martinoni A et al.: Left ventricular dysfunkción predicted by early Troponin I release after high-dose chemotherapy. J Am Coll Cardiol. 2000 Aug; 36(2):517-22.*
15. *Meinardi MT, van Veldhuisen DJ, Gietema JA et al.: Prospective evaluation of early cardiac damage induced by Epirubicin-containing adjuvant chemotherapy and locoregional radiotherapy in breast cancer patients. J Clin Oncology 2001May 15; 19 (10): 2746-2753.*
16. *Carrio I, Estorch M, Berna L et al.: Assessment of anthracycline-induced myocardial damage by quantitative indium 111 myosin-specific monoclonal antibody studies. Eur J Nucl Med 1991; 18:806-812.*
17. *Cottin Y, Touzery C, Dalloz F et al.: Comparison of Epirubicin and Doxorubicin cardiotoxicity induced by low doses: evolution of the diastolic and systolic parameters studied by radionuclid angiography. Clin.Cardiol. 1998, 21:665-670.*
18. *Elbl L, Vasova I, Kral Z, et al.: Echocardiographic evaluation of early and chronic cardiotoxicity in adult patients treated for Hodgkin's disease with ABVD regimen. Neoplasma. 2006;53(1):586-590.*
19. *Horácek JM, Pudil R, Tichy M, et al.: The use of biochemical markers in cardiotoxicity monitoring in patients treated for leukemia. Neoplasma. 2005;52(5):430-4.*
20. *Kass DA: Assesment of diastolic dysfunkción. Invasive modalities. Cardiol Clin. 2000 Aug;18(3):571-86.*
21. *Iarussi D, Galderisi M, Ratti G, et al.: Left ventricular systolic and diastolic funkción after anthracycline chemotherapy in childhood. Clin Cardiol. 2001; 24(10), 663-669.*
22. *Isaaz K, Thompson A, Ethevenot G, et al: Doppler echocardiographic measurement of low velocity motion of the left ventricular posterior wall. J Am Soc Cardiol. 1989; 64:66-75.*
23. *Uematsu M, Miyatake K, Tanaka N, et al.: Myocardial velocity gradient as a new indicator of regional left ventricular contraction: detection by a two-dimensional tissue Doppler imaging technique. J Am Coll Cardiol 1995; 26: 217-223.*

24. *Gulati VK, Katz WE, Follansbee WP, et al.: Mitral annular descent velocity by tissue Doppler echocardiography as an index of global left ventricular funkción. Am J Cardiol. 1996; 77:979-984.*
25. *Shimizu Y, Uematsu M, Shimizu H, et al.: Peak negative myocardial velocity gradient in early diastole as a noninvasive indicator of left ventricular diastolic funkción: comparison with transmitral flow velocity indices. J Am Coll Cardiol 1998; 32:1418-1425.*
26. *Schneider C, Dennehy C, Carter S: Exercise and Cancer Recovery. 1st ed. Champaign, Ill: Human Kinetics Publishers; 2003.*
27. *Ascensao A, Magalhaes J, Soaresi J, et al.: Endurance training attenuates doxorubicin-induced cardiac oxidative damage in mice. Int J Cardiol 2005; 100: 451–460. https:/doi.org/10.1016/j.ijcard.2004.11.004*
28. *Chicco AJ, Schneider CM, Hayward R: Voluntary exercise protects against acute doxorubicin cardiotoxicity in the isolated perfused rat heart. Am J Physiol Regul Integr Comp Physiol 2005; 289: R424-R431. https:/doi.org/10.1152/ajpregu.00636.2004*
29. *Krischer JP, Epstein S, Cuthbertson DD, et al.: Clinical cardiotoxicity following anthracycline treatment for childhood cancer: the Pediatric Oncology Group experience. J Clin Oncol 1997;15:1544–1552.*
30. *Lipshultz SE, Colan SD, Gelber RD, et al.: Late cardiac effects of doxorubicin therapy for acute lymphoblastic leukemia in childhood. N Eng J Med 1991;324:808–815.*
31. *Lipshultz SE, Colan SD, Walsh EP, et al.:Ventricular tachycardia and sudden unexplained death in late survivors of childhood malignancy treated with doxorubicin. Pediatr Res 1990;27:145A.*
32. *Lipshultz SE, Lipsitz SR, Mone SM, et al.: Female sex and drug dose as risk factors for late cardiotoxic effects of doxorubicin therapy for childhood cancer. N Eng J Med 1995; 333:1359–1360.*
33. *Brian R.J.,Bird H, Swain SM: Cardiac Toxicity in Breast Cancer Survivors: Reviewof Potential Cardiac Problems. Clin Cancer Res 2008 January 1;14(1): 14-24.*
34. *G. Curigliano, D. Cardinale, T. Suter et al.: Cardiovascular toxicity induced by chemotherapy, targeted agents and radiotherapy: ESMO Clinical Practice*

Guidelines. Annals of Oncology 23 (Supplement 7): vii155–vii166, 2012; doi:10.1093/annonc/mds293.

35. *Felker GM, Thomson RE, Hare JM et al.: Underlying Causes and Long-Term Survival in Patients with Initially Unexplained Cardiomyopathy NEJM 2000 April 13; 342:1077-1084*
36. *Lenihan DJ, Oliva S, Chow EJ, et al.: Cardiac toxicity in cancer survivors. Cancer. 2013 Jun 1;119 Suppl 11:2131-42. doi: 10.1002/cncr.28061.*
37. *Curigliano G1, Mayer EL, Burstein HJ, et al.: Cardiac toxicity from systemic cancer therapy: a comprehensive review.*
38. *Oefflinger KC, Mertens AC, Sklar CA et al.: Chronic health conditions in adult survivors of childhood cancer. NEJM 2006;355(15):1572.*
39. *Swain SM, Whaley FS, Ewer M: Congestive heart failure in patients treated with doxorubicin: a retrospective analysis of three trials. Cancer 2003; 97:2869-79.*
40. *Hequet O, Le QH, Moullet I, et al.: Subclinical late cardiomyopathy after doxorubicin therapy for lymphoma in adults. J Clin Oncol. 2004 May 15;22(10):1864-71.*
41. *Tassan-Mangina S, Codorean D, Metivier M, et al.: Tissue Doppler imaging and conventional echocardiography after anthracycline treatment in adults: early and late alterations of left ventricular function during a prospective study. Eur J Echocardiogr 2006;7:141-6.*
42. *Ho E, Brown A, Barrett P, 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.*
43. *Ganame J, Claus P, Uyttebroeck A, et al.: Myocardial dysfunction late after low-dose anthracycline treatment in asymptomatic pediatric patients. J Am Soc Echocardiogr 2007;20:1351-8.*
44. *Di Lisi D1, Bonura F, Macaione F, et al.: Chemotherapy-induced cardiotoxicity: role of the tissue Doppler in the early diagnosis of left ventricular dysfunction. Anticancer Drugs. 2011 Jun;22(5):468-72.*
45. *Sadao S, Masayo K, Shinkan T, et al.: Effect of Physical Activity on Breast Cancer Risk: Findings of the Japan Collaborative Cohort Study. Cancer Epidemiol Biomarkers Prev 2008;17:3396-3401.*

46. *Vainio H, Kaaks R, Bianchini F.: Weight control and physical activity in cancer prevention: international evaluation of the evidence. Eur J Cancer Prev. 2002 Aug;11 Suppl 2:S94-100.*
47. *Scott JM, Khakoo A, Mackey JR, et al.: Modulation of anthracycline-induced cardiotoxicity by aerobic exercise in breast cancer: current evidence and underlying mechanisms. Circulation. 2011 Aug 2;124(5):642-50*
48. *Schneider CM, Hsieh CC, Sprod LK, et al.: Effects of supervised exercise training on cardiopulmonary function and fatigue in breast cancer survivors during and after treatment. Cancer. 2007 Aug 15;110(4):918-25.*
49. *Courneya SK, Mackey JR, Bell GJ, et al.: Randomized Controlled Trial of Exercise Training in Postmenopausal Breast Cancer Survivors: Cardiopulmonary and Quality of Life Outcomes. Journal of Clinical Oncolog. 2003 May 1;21(9) 1660-1668*
50. *Courneya KS1, Friedenreich CM.: Framework PEACE: an organizational model for examining physical exercise across the cancer experience. Ann Behav Med. 2001 Fall;23(4):263-72.*
51. *Bu'Lock FA, Mott MG, Oakhill A, et al.: Left ventricular diastolic function after anthracycline chemotherapy in childhood: relation with systolic function, symptoms, and pathophysiology. Br Heart J 1995;73:340–350.*
52. *Nysom K, Colan SD, Lipshultz SR: Late cardiotoxicity following anthracycline therapy for childhood cancer. Prog Pediatr Cardiol 1998;8:121–128.*
53. *Steinherz LJ, Steinherz PG, Tan CT, et al.: Cardiac toxicity 4 to 20 years after completing anthracycline therapy. JAMA 1991;266:1672–1677.*
54. *Uematsu M, Nakatani S, Yamagishi M, et al.: Usefulness of myocardial velocity gradient derived from two-dimensional tissue Doppler imaging as an indicator of regional myocardial contraction independent of translational motion assessed in atrial septal defect. Am J Cardiol 1997; 79:237–241.*
55. *Kantrowitz NE, Bristow MR: Cardiotoxicity of antitumour agents. Prog Cardiovasc Dis 1984;27:195–200.*
56. *Lengyel M., Nagy A., Zorándi A.: [Tissue Doppler echocardiography: a new technique to assess diastolic function]. Orv. Hetil., 2002; 143(7): 333–339*
57. *Chahal NS1, Lim TK, Jain P, et al.: Normative reference values for the tissue Doppler imaging parameters of left ventricular function: a population-based study. Eur J Echocardiogr. 2010 Jan;11(1):51-6.*

58. *Mori T, Yanagi N, Maruyama T et al.: Left ventricular diastolic dysfunkción induced by cyclophosphamide in blood stem cell transplantation. Jpn Heart J. 2002 May;43(3):249-261.*
59. *Shimizu Y, Uematsu M, Shimizu H, et al.:Peak negative myocardial velocity gradient in early diastole as a noninvasive indicator of left ventricular diastolic function: comparison with transmitral flow velocity indices. J Am Coll Cardiol 1998;32:1418–1425*
60. *Steinberg JS, Cohen AJ, Wasserman AG, et al.: Acute arrhythmogenicity of doxorubicin administration. Cancer 1987; 60(6): 1213–1218.*
61. *Kapusta L, Thijssen JM, Groot-Loonen J, et al.: Tissue Doppler imaging in detection of myocardial dysfunction in survivors of childhood cancer treated with anthracyclines. Ultrasound Med Biol 2000;26:1099–1108.*
62. *Krupicka J, Markova J, Pohlreich D, et al.: German Hodgkin's Lymphoma Study Group. Echocardiographic evaluation of acute cardiotoxicity in the treatment of Hodgkin disease according to the German Hodgkin's Lymphoma Study Group. Leuk Lymphoma 2002;43:2325–2329.*
63. *Ho E, Brown A, Barrett P, 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.*
64. *Ganame J, Claus P, Uyttebroeck A, et al.: Myocardial dysfunction late after low-dose anthracycline treatment in asymptomatic pediatric patients. J Am Soc Echocardiogr 2007;20:1351-8.*
65. *Nagueh SF, Appleton CP, Gillebert TC et al.: Recommendations for the Evaluation of Left Ventricular Diastolic Function by Echocardiography. European J Echocard 2009;10:165–193. doi:10.1093/ejechocard/jep007*
66. *Plana JC, Galderisi M, Barac A 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.*
67. *Di Lisi D1, Bonura F, Macaione F, et al.: Chemotherapy-induced cardiotoxicity: role of the tissue Doppler in the early diagnosis of left ventricular dysfunction. Anticancer Drugs. 2011 Jun;22(5):468-72.*

68. *Suzuki S,Kojima M, Tokudome S, et al.: Effect of Physical Activity on Breast Cancer Risk: Findings of the Japan Collaborative Cohort Study. Cancer Epidemiol Biomarkers Prev 2008;17:3396-3401.*
69. *Vainio H, Kaaks R, Bianchini F: Weight control and physical activity in cancer prevention: international evaluation of the evidence. Eur J Cancer Prev. 2002 Aug;11 Suppl 2:S94-100.*
70. *Scott JM, Khakoo A, Mackey JR, et al.: Modulation of anthracycline-induced cardiotoxicity by aerobic exercise in breast cancer: current evidence and underlying mechanisms. Circulation. 2011 Aug 2;124(5):642-50*
71. *Schneider CM, Hsieh CC, Sprod LK, et al.: Effects of supervised exercise training on cardiopulmonary function and fatigue in breast cancer survivors during and after treatment. Cancer. 2007 Aug 15;110(4):918-25.*
72. *Courneya KS, Mackey JR, Bell GJ, et al.: Field, and Adrian S. Fairey: Randomized Controlled Trial of Exercise Training in Postmenopausal Breast Cancer Survivors: Cardiopulmonary and Quality of Life Outcomes. Journal of Clinical Oncology, 2003 May 1;21(9):1660-1668*
73. *Courneya KS1, Friedenreich CM. Framework PEACE: an organizational model for examining physical exercise across the cancer experience. Ann Behav Med. 2001 Fall;23(4):263-72.*