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THE USE OF CARDIAC BIOMARKERS IN DETECTION OF CARDIOTOXICITY ASSOCIATED WITH CONVENTIONAL AND HIGH-DOSE CHEMOTHERAPY FOR ACUTE LEUKEMIA

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Aim: **Monitoring of cardiotoxicity of conventional and high-dose chemotherapy (HD-CT) with multiple biomarkers of cardiac injury — glycogen phosphorylase BB (GPBB), heart-type fatty acid binding protein (H-FABP), cardiac troponins (cTnT, cTnI), creatine kinase MB (CK-MB mass), myoglobin.** ***Methods:*** **A total of 47 adult acute leukemia patients were studied — 24 patients treated with conventional CT containing anthracyclines (ANT) and 23 patients treated with HD-CT (myeloablative preparative regimen) followed by hematopoietic cell transplantation (HCT). Cardiac biomarkers were assessed prior to treatment (before CT/HD-CT), after first CT with ANT, after last CT with ANT in the first group, after HD-CT and after HCT in the second group. Values above the reference range were considered elevated.** ***Results:*** **Before CT/HD-CT, all biomarkers of cardiac injury were below the cut-offs in all patients. GPBB increased above the cut-off (7.30 μg/L) in 4 (16.7%) patients after first CT and in 5 (20.8%) patients after last CT with ANT. GPBB increased above the cut-off in 5 (21.7%) patients after HD-CT and remained elevated in 5 (21.7%) patients after HCT. CTnI became elevated (above 0.40 μg/L) in 2 (8.3%) patients after first and last CT with ANT. Both patients with cTnI positivity had elevated GPBB. Other tested biomarkers remained below the cut-offs during the study.** ***Conclusion:*** **Our results suggest that GPBB could become a sensitive biomarker for detection of acute cardiotoxicity associated with conventional CT containing ANT and HD-CT followed by HCT. The predictive value for development of cardiomyopathy in the future is not known and should be evaluated during a prospective follow-up. Based on our data, a larger prospective and multicenter study would be most desirable to define the potential role of new circulating biomarkers in the assessment of cardiotoxicity in oncology.**

Key Words: **cardiotoxicity, biomarkers, glycogen phosphorylase BB, chemotherapy, anthracyclines, high-dose.**

Cardiotoxicity is a well-known and potentially serious complication of anticancer therapy that can significantly impair patient's quality of life and also substantially increase health care costs. The greatest risk for development of cardiotoxicity represent anthracyclines (ANT) [1–3] and high-dose chemotherapy (HD-CT) especially regimens containing high-dose cyclophosphamide [4–6]. Various methods have been recommended for monitoring of cardiotoxicity in oncology [7, 8]. In our conditions, echocardiography and electrocardiography are routinely used [9, 10]. Recently, biochemical markers of cardiac injury, especially cardiac troponins and natriuretic peptides, have been investigated in the assessment of cancer therapy-induced cardiotoxicity [11–13].

Cardiac troponins — cardiac troponin T (cTnT), cardiac troponin I (cTnI) — and myocardial izoenzyme of creatine kinase (CK-MB) are cardiospecific markers that show structural injury of cardiomyocytes from various causes, including cardiotoxic effect of anticancer therapy [14–17].

Heart-type fatty acid binding protein (H-FABP) and glycogen phosphorylase isoenzyme BB (GPBB) are new perspective markers of myocardial ischemia and necrosis, recently evaluated in the diagnostics and risk stratification of acute coronary syndromes [18–22]. H-FABP is a relatively small cytoplasmic protein for the oxidation of fatty acids that is quite specific for cardiac muscle. H-FABP is rapidly released from the myocardium into the bloodstream after ischemic injury. Plasma H-FABP increases above the reference limit within 2–3 h of the onset of myocardial injury and returns to normal within 18–30 h. GPBB is a glycogenolytic enzyme providing glucose for heart muscle tissue. After glycogenolysis in ischemic tissue, GPBB is released from the sarcoplasmic reticulum into the cytoplasm and then into the circulation through the damaged cell membrane. GPBB is released into the circulation 2–4 h after myocardial injury, returning to normal values within 24–36 h of damage occurring. In the acute coronary syndrome setting, both markers are regarded early markers of cardiac injury due to acute myocardial ischemia. However, the main mechanism of cardiac injury caused by anticancer therapy is

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Abbreviations used: ALL – acute lymphoblastic leukemia; AML – acute myeloid leukemia; ANT – anthracyclines; CK-MB – creatine kinase MB; CT – chemotherapy; cTnI – cardiac troponin I; cTnT – cardiac troponin T; GPBB – glycogen phosphorylase BB; HCT – hematopoietic cell transplantation; HD-CT – high-dose chemotherapy; H-FABP – heart-type fatty acid binding protein.

mainly non-ischemic and prior cyclic exposition to ANT agents may play a role (chronic and late cardiotoxicity). Therefore, it is difficult to estimate the kinetics of release of these biomarkers from cardiomyocytes in this setting. At present, new perspective biomarkers of cardiotoxicity have been evaluated inadequately in patients treated for oncological diseases.

The aim of our study was to evaluate cardiac toxicity of conventional CT containing ANT and HD-CT followed by hematopoietic cell transplantation (HCT) with multiple biomarkers of cardiac injury: glycogen phosphorylase BB (GPBB), heart-type fatty acid binding protein (H-FABP), cardiac troponins (cTnT, cTnI), creatine kinase MB (CK-MB mass), myoglobin. Experience with GPBB and H-FABP in this context is very limited.

PATIENTS AND METHODS

A total of 47 patients treated for acute leukemia were included in the study. The study population was divided into 2 groups — (1) 24 patients treated with conventional CT containing ANT (3–6 cycles of CT, mean total cumulative ANT dose 463.2 ± 114.3 mg/m^2), mean age 48.1 ± 10.9 years, 13 males. All patients had de novo acute myeloid leukemia (AML): 2 — AML M1, 16 — AML M2, 5 — AML M4, 1 — AML M5 according to FAB classification; (2) 23 patients treated with HD-CT (ablative preparative regimen containing high-dose cyclophosphamide in the total dose of 120 mg/kg in combination with peroral busulfan 16 mg/kg /BuCy2/ or fractionated total body irradiation 12 Gy /CyTBI/) followed by HCT, mean age of 44.5 ± 10.6 years, 15 males. Twenty patients had AML, from that 2 — AML M1, 14 — AML M2, 4 — AML M4 according to FAB classification, and 3 patients had acute lymphoblastic leukemia (ALL), from that 2 — B-ALL, 1 — T-ALL. All patients had normal liver and renal functions during the study. The study was carried out with approval from the ethics committee. All patients gave a written consent before they were included in the study.

In the first group, biochemical analysis was performed at the baseline (before CT), the day after first CT with ANT (after first CT; mean cumulative ANT dose 130.6 ± 29.8 mg/m^2), the day after last CT with ANT (after last CT; mean cumulative dose 463.2 ± 114.3 mg/m^2). In the second group, cardiac biomarkers were assessed the day before administration of HD-CT (before HD-CT), the day after completion of HD-CT (after HD-CT) and the day after HCT (after HCT).

Circulating biomarkers of cardiac injury were measured according to the manufacturer's guidelines as follows: cTnT, CK-MB mass, myoglobin (Roche Diagnostics; Elecsys analyzer), GPBB, H-FABP, cTnI (Randox; Evidence Investigator analyzer).

Concentrations of cardiac biomarkers diagnostic for cardiotoxicity of oncology treatment have not been established yet. On that ground, values above the reference range based on a number of cardiology studies and recommended by the manufacturers were considered elevated. The cut-off values for cardiac injury were as follows: 7.30 μg/L for GPBB, 4.50 μg/L for H-FABP, 0.40 μg/L for cTnI, 0.01 μg/L for cTnT, 4.80 μg/L for CK-MB mass and 76.0 μg/L for myoglobin.

RESULTS

The cut-off values of all biomarkers and the number of patients with elevated values are shown in Table 1 and 2.

Table 1. Elevated biomarkers of cardiac injury in association with anthracycline-based chemotherapy (n = 24)

Cardiac biomarkers	Before CT	After first CT	After last CT
Myoglobin above 76.0 μg/L	0	0	0
CK-MB mass above 4.80 μg/L	0	0	0
cTnT above 0.01 μg/L	0	0	0
cTnI above 0.40 μg/L	0	2 (8.3%)	2 (8.3%)
H-FABP above 4.50 μg/L	0	0	0
GPBB above 7.30 μg/L	0	4 (16.7%)	5 (20.8%)

In Table 1 and 2: CT – chemotherapy; HD-CT – high-dose chemotherapy; HCT – hematopoietic cell transplantation; CK-MB mass – creatine kinase MB; cTnT – cardiac troponin T; cTnI – cardiac troponin I; H-FABP – heart-type fatty acid binding protein; GPBB – glycogen phosphorylase BB.

Table 2. Elevated biomarkers of cardiac injury in association with high-dose chemotherapy and hematopoietic cell transplantation (n = 23)

Cardiac biomarkers	Before HD-CT	After HD-CT	After HCT
Myoglobin above 76.0 μg/L	0	0	0
CK-MB mass above 4.80 μg/L	0	0	0
cTnT above 0.01 μg/L	0	0	0
cTnI above 0.40 μg/L	0	0	0
H-FABP above 4.50 μg/L	0	0	0
GPBB above 7.30 μg/L	0	5 (21.7%)	5 (21.7%)

Before CT/HD-CT, all biomarkers of cardiac injury were below the cut-off values in all patients. GPBB concentrations increased above the cut-off in 4 (16.7%) patients after first CT and in 5 (20.8%) patients after last CT with ANT. In the second group, GPBB increased above the cut-off in 5 (21.7%) patients after HD-CT and remained elevated in 5 (21.7%) patients after HCT. CTnI concentrations became elevated in 2 (8.3%) patients after first and last CT with ANT. Both patients with cTnI positivity had elevated GPBB. CTnI remained negative after HD-CT and HCT in all patients. Other tested biomarkers (H-FABP, cTnT, CK-MB mass, myoglobin) remained below the cut-offs during conventional CT containing ANT and HD-CT followed by HCT.

DISCUSSION

GPBB and H-FABP have been recently investigated in the early detection of cardiac injury. They have been suggested sensitive markers of myocardial ischemia and necrosis in patients with acute coronary syndromes [18–22]. Moreover, GPBB could be associated with pulmonary artery wedge pressure and left ventricle mass index in patients with hypertrophic cardiomyopathy. This finding raises an important issue concerning the role of cardiac biomarkers in the diagnosis of hypertrophic cardiomyopathy and could potentially provide a useful tool to stratify these patients [23]. However, in the field of cardiotoxicity of anticancer therapy, these perspective biomarkers of cardiac injury have been evaluated inadequately. In 2008, we published our pilot study on usability of multiple biomarkers, including GPBB and H-FABP, for evaluation of ANT-induced cardiotoxicity in patients with acute myeloid leukemia [24]. Recently, we applied multiple biomarkers, including GPBB and H-FABP, in the assessment of cardiotoxicity associated with administration of preparative regimen and HCT for

various hematological malignancies [25]. ElGhandour *et al.* [26] studied H-FABP in 40 non-Hodgkin's lymphoma patients treated with 6 cycles of CT containing doxorubicin (cumulative dose 300 mg/m^2). The authors concluded that H-FABP may serve as a reliable early marker for prediction of cardiomyopathy induced by doxorubicin. Otherwise, no studies have been published so far and experience with these perspective biomarkers in the assessment of cardiotoxicity of anticancer therapy is very limited.

In our present study on 47 acute leukemia patients, we found significant elevations in GPBB after CT with ANT (in 16.7% and 20.8% patients, respectively) and after HD-CT followed by HCT (in 21.7% patients). Increased release of GPBB from cardiomyocytes after administration of CT could be considered a sign of acute subclinical cardiotoxicity of this treatment. Positivity of GPBB in patients with negativity of other biomarkers (cTnI, cTnT, H-FABP, CK-MB mass, myoglobin) suggests that GPBB could be a more sensitive marker for detection of acute cardiac injury caused by anticancer therapy, both conventional and HD-CT. Whether these acute changes will have predictive value for development of CT-associated cardiomyopathy in the future is not clear and should be evaluated during a prospective follow-up. Based on our data, a larger prospective and multicenter study would be most desirable to define the potential role of new circulating biomarkers in the assessment of cardiotoxicity in oncology.

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