ORIGINAL ARTICLE

Assessment of Biomarkers of Oxidative Stress among Patients with Lymphoma

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ABSTRACT: The role of oxidative stress is one of the most important factors in the development of cancer. In this study, the catalase activity and total antioxidant capacity in NHL patients were measures and compared with those in healthy subjects. The population of this study consisted of 25 patients with non-Hodgkin's lymphoma, diagnosed based on pathological findings. Total antioxidant capacity (TAC) and catalase catalytic activity (CAT) were determined by spectrophotometry. Based on the results, the CAT activity was significantly lower and TAC was lower (0.29 ± 0.04 vs. 2.69 ± 0.15; P-value=0.01) in NHL patients than in healthy subjects. The findings showed that these markers could be used as a prognosis in non-Hodgkin's lymphoma that oxidative stress may also be related to the NHL or increase the risk of disease.

INTRODUCTION

A group of diseases characterized by uncontrolled cell growth and proliferation is called cancer [1, 2]. The angiogenic process results from the growth of a cancerous tumor, which provides sources of oxygen and nutrients for the tumor tissues and metastasizes to other parts of the body [3, 4]. Non-Hodgkin's lymphoma (NHL) is a malignant lymphoproliferative disease. Risk factors, such as autoimmune disorders, specific infections, genetic factors, and history of lymphoma, can predispose to the disease. An imbalance between the production of free radicals and reactive metabolites leads to the production of reactive oxygen species leading to damage to vital cells in the body [5, 6]. In high concentrations, free radicals can cause oxidative stress, which is a harmful process damaging severely all biological molecules in the body, disrupting cell function, and even leading to necrosis and disease. Oxidative stress leads to cancer, cardiovascular disorders, and neurological diseases over time [7, 8, 9]. To combat this process, the body uses the immune system of antioxidants, such as peroxidase and glutathione reductase, catalase, and superoxide dismutase. Additionally, such antioxidants as vitamin C, vitamin E, and carotenoids, which have high antioxidant power, are taken to minimize the harmful effects of reactive oxygen species [10, 11].

Many studies have been performed to investigate the total antioxidant capacity (TCA) and lipid and catalase peroxidation. Serum levels (e.g., catalase catalytic activity) are an essential component of the antioxidant system [12]. This study aimed to compare the levels of TCA and catalase catalytic activity (CAT) in the serum of NHL patients from those of healthy subjects and investigate whether these variables may be considered necessary to predict NHL patient survival.

MATERIALS AND METHODS

This case-control study was conducted on 25 cases in Zahedan city, Sistan and Baluchestan Province, Iran.

Study population

Patients group

To conduct the study, the blood samples of 25 patients with lymphoma were collected. The subjects did not have any other known cancers or systemic diseases and were under the treatment process. The inclusion criteria were having lymphoma (non-Hodgkin's disease) being confirmed by a pathologist and lacking secondary malignancies and congenital chromosomal abnormalities. The patients' demographic information, including age, gender, height, weight, location, and body mass index were obtained and calculated.

Control group

The blood samples of 25 healthy subjects with no known cancer or systemic disease were collected in Zahedan city.

Samples collection

The cases in both groups were homogenous regarding age, gender, and ethnicity. A volume of 7 ml of blood samples was collected in non-coagulated tubes and stored at 25°C for 25 min. Subsequently, the blood samples were centrifuged at 3,000 rpm for 10 min in a refrigerated centrifuge and the serum was carefully collected and stored at -70°C until oxidative stress enzymes were measured.

Oxidative stress enzymes were measured according to the guidelines of standard protocols.

Assay of catalase activity

To evaluate the activity of CAT, a colorimetric enzyme assay kit was used at 405 nm (ZellBio GmbH, Ulm, Germany). Based on the kit protocols, the CAT activity unit was considered micromoles. This method can determine the activity of CAT with a 0.5 U/ml sensitivity. The coefficient of variation was calculated within and between the measurements rendering for 6.3% and 7.9%, respectively.

Measurement of total antioxidant capacity

Serum TAC levels were measured using a kit and following the protocol of the manufacturer (ZellBio GmbH, Germany) by colorimetric method and reduction of oxidant at 490 nm. Serum TAC levels were considered to be superior to the function of vitamin C due to the presence of antioxidants in the sample. This method can determine the serum TAC level with a 0.1 mm sensitivity (100 μmol/L). It is claimed that the variability values within and between assays are 4.4% and 4.2%, respectively.

Statistical analysis

The collected data were analyzed in SPSS software (version 21) using the mean score, as well as the student t-test to compare the groups. A p-value of < 0.05 was considered significant.

RESULTS

Blood serum samples from diagnosed NHL patients and healthy volunteers (n=25 each group) were analyzed to measure the TCA level and CAT activity. According to the results, CAT activity and TCA levels were significantly lower in NHL patients than in the control group (Table 1). Therefore, the findings showed a decrease in the antioxidant system and an increase in lipid peroxidation resembling oxidative stress among ovarian cancer patients.

Table 1. Comparison of total antioxidant capacity levels and catalase catalytic activity in study and control groups

Parameter	Control (n=25)	Cases (n=25)	P-value
TCA	2.69±0.15	1.29±0.04	0.01
CAT	4.62±0.4	3.45±0.2	0.01

TCA: total antioxidant capacity, CAT: catalase catalytic activity

DISCUSSION

Many clinical disorders originate from an imbalance between the production of oxidative molecules and the ability of biological antioxidant systems to detoxify these oxidative stress markers [13, 14]. Oxidative stress plays an important role in changing metabolic pathways in tumor cells, resulting in changes that can affect tumor progression and the adaptation of cancer cells to oxidative stress, as well as increasing the treatment resistance, angiogenesis, and risk of metastasis [15, 16, 17, 18]. Consequently, oxidative stress can be particularly important in the prognosis of cancer. In this study, the CAT activity and TAC level were measured in blood samples of patients with lymphoma and compared with the results of a study of healthy individuals. The Findings of the present research indicated that the TAC level in patients with non-Hodgkin's lymphoma was significantly lower than that in the control group. Additionally, the results of CAT enzyme activity showed a low significant relationship between patients and healthy individuals. The reason for this change can be attributed to an increase in metabolism as a result of increased production of free radicals [19, 20, 21]. One of the natural metabolites in living cells is hydrogen peroxide. defense against oxidative stress, antioxidant enzyme catalase to break down hydrogen peroxide into water and oxygen, which is not a radical [22, 23, 24, 25]. Removal of hydrogen peroxide is essential since it leads to the oxidation of cellular components. Liver and kidney cells also have catalase activity and the lowest catalase activity is in connective tissues [26, 27, 28]. Although serum TAC and CAT levels are good markers for oxidative and antioxidant status, they are still not definitively indicative of changes in NHL patients, due to the involvement of many factors [29, 30, 31, 32].

CONCLUSIONS

The results of our study showed a decrease in antioxidant activity of TCA and CAT indicated a decrease in the activity of NHL antioxidant enzymes. This means that oxidant/antioxidant imbalance may also be associated with an increased risk for NHL patients. Low serum levels of TCA and CAT develop this hypothesis that some disturbances in the oxidant-antioxidant balance in favor of oxidation may contribute to NHL formation. It is recommended to perform further studies to determine the role of oxidative stress among NHL patients.

ACKNOWLEDGMENTS

Conflicts of interest

The authors declare that there is no conflict of interest.

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