

Estimation and Assessment of Plasma Reduced Glutathione Status among Sudanese Females with Breast Cancer Undergoing Chemotherapy

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Abstract

Original Research Article

Glutathione is considered as a major molecule that plays an essential role in the antioxidant defense system. The mechanism of chemotherapeutic agents are act based on oxidative stress to kill the cancer cell, so the antioxidant activity of glutathione is thought to be interfering with chemotherapeutic action. The aim of the recent study is Estimation of Plasma Reduced Glutathione among Sudanese Females with Breast Cancer Undergoing Chemotherapy. The current study was conducted on 100 Sudanese females with Breast Cancer who attended the Institute of Nuclear Medicine – Madani – SUDAN between August to October / 2018. ELISA 96 well plates (OD 412 nm) and quantiChrom Glutathione assay kit were used to measure the level of reduced glutathione in Plasma. The outcome of the recent study, the level of plasma reduced glutathione in patients was significantly higher than control (MEAN 92.03±46.6; 13.87±9.27 respectively; P-value 0.001). Our finding suggests that the status of reduced glutathione levels might be play a role as a prognostic factor for poor response of chemotherapy in Sudanese female with Breast Cancer.

Keywords: Glutathione, Breast cancer, chemotherapy, Antioxidant, Poor response.**Copyright © 2020:** This is an open-access article distributed under the terms of the Creative Commons Attribution license which permits unrestricted use, distribution, and reproduction in any medium for non-commercial use (NonCommercial, or CC-BY-NC) provided the original author and source are credited.

INTRODUCTION

The main objective of chemotherapy is to eliminate only the tumorous cells (however, most of the antineoplastic agents act in a non-specific way, harming as many malignant cells as normal) [1]. Nevertheless, some of the Breast Cancer Patients (BCP) appears no improvement toward chemotherapeutics agents and their clinical situation becomes worse even after taken many doses.

Drug resistance in Breast cancer chemotherapy represents a major challenge to the successful treatment of patients [2, 3]. Chemotherapy acts against cancer cells based on an increase in the rate of oxidative stress to through raise the level of reactive oxygen species up to the toxic level to make serious damage in large molecules such as DNA and also cause loss of malignant cellular membrane integrity and subsequently cell death [4-8]. But cancer cell creates a mechanism to increase its antioxidant rate as response to the high oxidation rate of chemotherapy [9].

Some of the previous studies have demonstrated that there are many factors interfere with the action of chemotherapeutics drugs. One of the reasons is attributed to the changeable level of antioxidants in cancer cells. It is thought one of the most common substances that cancer cells are use as antioxidants is Glutathione because it has been proven that glutathione has a high antioxidant activity and it is considered as “master antioxidant “molecules inside the body [10]. The cancer cell can benefit from a high rate of glutathione as an anti-oxidant to discourage the effectiveness of chemotherapeutic agents [11, 12]. The reduced form of glutathione (GSH) is prevalent form in mammalian cells [13, 14] main protective roles of reduced glutathione, it represents in many functions such as amino acid transporting through the plasma membrane; GSH scavenges hydroxyl radical and hydrogen peroxide; also glutathione is able to regenerate the most important antioxidants molecules like vitamins through back them to their active form [15, 16].

The strength of glutathione as powerful antioxidant molecules attributed to cysteine sulfhydryl group which has ability to react directly with toxicants molecules or indirectly through activation of glutathione S-transferase group of enzymes, and based on most of the antineoplastic agents kill the malignant cells through binding with electrophilic sites of DNA to kill cells so when this binding is interrupted by glutathione, cancer cell could escape from killing by chemotherapeutic agents and it becomes more progressive[17].

The aim of conducting this study is Estimation of Plasma Reduced Glutathione among Sudanese Females with Breast Cancer Undergoing Chemotherapy because it has been observed most of the patients with chemotherapy do not appear to satisfy improvement response toward their treatment doses.

Study design

Cross-sectional, case control study design was conducted on 100 Breast Cancer Patients (BCP) Who attended to Institute of the nuclear Medicine, AL Gezira State –SUDAN during August – October / 2018, and 100 healthy females (control). A well-constructed -questionnaire was used for Data Collection. Ethical approval was granted by the Ethical Committees in the Ministry of Health – Al Gezira State - SUDAN and the Institute of the nuclear Medicine –Al Gezira University – Madani.

The study population was divided into 50 % patients and 50% control see figure (1) for distribution.

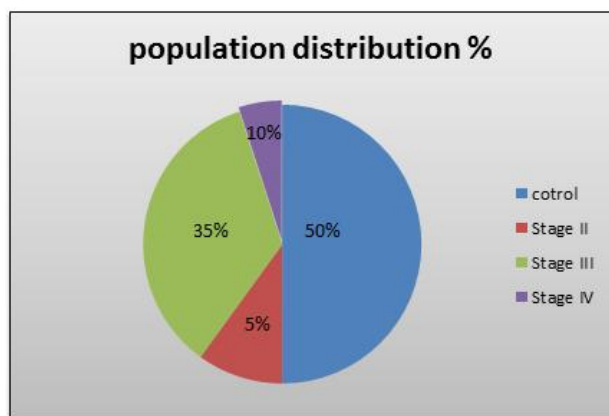


Fig-1: Illustrate distribution of population

MATERIAL AND METHOD

ELISA 96 well plates (OD 412 nm), were used. Whole EDTA Blood samples were collected and centrifuged at 3000 rpm for 10 minutes. Plasma was separated and aliquots in Eppendorf tubes and kept at -20 °C for 2 weeks later. To measure reduced glutathione the quantiChrom Glutathione assay kit (DIGT-250) was used (it is simple and direct automation-ready procedures for measuring in addition to low interference [18], the kits composed of two reagents;

reagent A and B (volume 30 ml for each bottle) and calibrator (10 ml volume and equivalent to 100µM). All reagents were equilibrated to Room Temperature. Reagent A (for deproteination) 120 µL of plasma sample mixed with 120 µL reagent A in 1.5 ml centrifuge tubes mixed and vortex well. 200 µL of the mixture (sample + reagent A) was transferred into 96 well plates and 100 µL of reagent B was added. The plate was lightly tapped to mix. Incubated 25 min at Room Temperature (RT) and read at Optical Density (OD) 412 nm. For calculations follow the manufacture assay kit (DIGT-250) instructions.

RESULT

Data were statistically analyzed using the Statistical Package of Social Science (SPSS) program version 21. Statistics used were the mean, standard deviation (SD), Confident Interval (CI) 0.05, Pearson correlation (r) and P-value. P-Value of <0.05 was considered significant and > 0.05 was considered insignificant. For mean and Stander Division of age and level of reduced Glutathione of study subjects see table (1).

Table-1: Age and plasma reduced glutathione in patients and control

Parameters	patients (Mean/SD)	control (Mean/SD)
Age	47.2 ±3.41	44.6 ± 2.25
Level of Reduced Glutathione (µM)	92.03 ±46.06	13.87 ±9.27
Number of Subjects	100	100

- P-value < 0.001 was obtained when compared level of reduced Glutathione between patients and control.

DISCUSSION

We observed that the reduced glutathione level is significantly higher (P-value < 0.001) in patients compared with control. This is in agreement with the results of earlier studies which have demonstrated that reduced glutathione level increased in Breast Cancer Patients [19-22]. The increased level of GSH in our participants' (patients) might be related to cancer because we obtained a normal level in healthy participants (control). So it could be there is a relationship between a high level of GSH and poor response to chemotherapy treatment especially most of the patients who participated in this study were in late stages and they have been given many doses of treatment but they didn't appear any improvement based on their clinical situation. Nevertheless, for more approval the result of this study we need further studies including a large sample size and long term follow-up of patients in future researches.

The deep Knowledge and study the role of glutathione interference as an antioxidant with chemotherapeutic agents in Sudanese females with Breast Cancer may give a clear picture of the relationship between poor response to chemotherapy and the generation of the high level of antioxidants in patients.

CONCLUSION

Our finding suggests that the reduction in glutathione level may have a play an important role as a prognostic factor for poor response of patients to chemotherapy in Sudanese female with Breast Cancer.

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