Elevated Serum Levels of CEA in Egyptian Female Breast Cancer Patients as Standing Tumor Biomarker

Samar A. Ghonem, Rasha F. Zahran, Menha M. Swellam, Camelia A. Abdel-Malak

Abstract- Tumor markers are substances produced by the tumors or by other cells of the body in response to cancer or certain benign conditions. Although most of these markers are made by the normal cells as well as by cancer cells, they are produced at much higher levels in cancerous conditions. These markers are used to evaluate the patient's response to treatment and to detect the presence of metastasis or recurrence. Breast cancer is one of the most common malignancies in females worldwide. The Carcinoembryonic antigen CEA is tumor marker that is often expressed in people with breast cancer. It plays a crucial role in diagnosis, monitoring response to therapy, early detection of metastasis and determination of recurrence in patients with breast cancer.

Keywords- Breast cancer, Carcinoembryonic antigen, Diagnosis, Screening, Tumor marker

1 INTRODUCTION

A tumor marker is a biomarker that is found in blood, urine or body tissues that can be elevated by the presence of one or more types of cancer. It is produced either by the tumor itself or by the host in the response to a tumor [1]. The ideal tumor marker should be both specific and sensitive to detect small tumors to allow early diagnosis or help in screening. Few markers are specific for a single tumor. Most markers are produced by different tumors of the same tissue type. They are present in higher quantities in cancer tissue or in blood from cancer patients more than in the blood of normal subjects. Tumor markers are mostly useful in evaluating the progression of the disease status after initial chemotherapy and radiotherapy to monitor subsequent treatment strategies [2]. Breast cancer is the second most common type of cancer after lung cancer (10.4% of all cancer incidence, both sexes counted) and the fifth most common cause of cancer death [3]. It is a disease caused by a combination of genetic and environmental factors. Numerous risk factors that may be associated with breast cancer have been recognized. Not all breast cancer patients have the same clinical picture. Some factors increase a woman's risk of breast cancer more than others [4].

In recent decades, the serum concentration of tumor markers has been used to detect tumor activity. Tumor markers provide a minimally invasive cost-effective source of data valuable for monitoring disease course, determining prognosis, and helping in treatment planning. An understanding of the individual test characteristics and limitations is important for optimal use and accurate interpretation of results [5]. The real usefulness of tumor markers in the management of breast cancer has been questioned because of the low diagnostic sensitivity for early disease [6].

Carcinoembryonic antigen (CEA), which belongs to a family of related cell surface glycoproteins, is the most widely used tumor marker in the clinical practice. It is a tumor marker for colorectal, gastrointestinal, lung and breast cancer [7]. CEA was first identified as a tumor specific antigen found in extracts of tumor tissue. It is also found in normal foetal gastrointestinal tract epithelial cells. It is a glycoprotein that contains 45-50% carbohydrates. It is a single polypeptide chain consisting of 641 amino acids, with lysine at its N-terminal position [8].

The human carcinoembryonic antigen (CEA) family is composed of 29 genes. These genes are classified into two major subfamilies, the CEA cellular adhesion molecule (CEACAM) and the pregnancy specific glycoprotein subgroups. The CEACAM family belongs to the immunoglobulin superfamily. CEA might act as an adhesion molecule. Because alternations in cell adhesions are involved in cancer invasion and metastasis, it was further suggested that CEA may play a crucial role in these processes [9].

Continuous rising level of CEA in breast cancer may explain either cancer not responding to treatment, or recurrence after treatment. As

steadily rising CEA may be the first sign of cancer recurrence after treatment [10]. Also, patients with advanced cancer or metastatic cancer may have higher CEA levels rather than in patients with localized diseases [7]. CEA can be used to help diagnosis, clinical staging, to detect recurrence in patients who have undergone surgery, and to monitor the therapeutic response in patients undergoing chemotherapy or radiotherapy [11].

In breast cancer, elevated CEA is associated with metastatic disease. Circulating levels of CEA in breast cancer patients are directly dependable on the size of both primary and metastatic tumor. For breast cancer, CEA is being replaced by other more specific markers, such as CA 15-3 [12] B. Geng et al [13] suggested that there should be an association between CEA, CA 15- 3 and the clinicopathological parameters for proper diagnosis in patients with metastatic breast cancer.

2 MATERIALS AND METHODS

2.1 Enrolled individuals and collection of samples

After obtaining ethical approval from the Scientific Medical Ethical Committee of National Research Centre, blood samples were withdrawn from enrolled individuals after they signed their informed consent. The study was conducted on individuals who were divided into: group I was healthy females (n=20) and they were considered as control group, group II (n=30) consists of female patients with benign breast diseases and group III (n=100) consists of female patients with primary breast cancer. According to the study strategy, inclusion criteria were patients of primary breast cancer and exclusion criteria were patients that undergo mastectomy or lumpectomy, patients who received radio- or chemotherapy and patients with other types of malignancies or distant metastasis.

Blood samples were collected from all joined individuals then for 30 minutes they allowed to clot at room temperature. After that, for 10 minutes; all samples were centrifuged at 10.000 g at 4°C. The separated result sera were aliquoted and stored at -80°C for tumor marker assessments.

2.2 Tumor markers assessment

By enzyme linked immunesorbent assay (ELISA), CEA was detected in serum samples using available commercial ELISA kit (Catalog No.E1-207, Immunospec Corporation, Canoga Park, CA, USA), then their concentration was detected.

The wells are coated with anti-CEA antibodies. The samples, Standards and Controls are incubated in the wells with enzyme conjugate which is another antibody directed toward a different region of CEA molecules and chemically conjugated with horseradish peroxidase. Unbound enzyme conjugate is washed off and the amount of bound peroxidase is proportional to the concentration of the CEA present in the Samples, Standards and Controls. Upon addition of the TMB substrate, the intensity of color developed is proportional to the concentration of CEA in the serum. The optical density of the colored samples is read with a microplate reader at 450 nm.

The CEA values of the samples were obtained by plotting the concentration (X) of each reference standard against its absorbance (Y) on log-log standard curve as illustrated in table (1) and the mean absorbance values for each specimen were used to determine the corresponding concentration of CEA in ng/mL from the standard curve as shown in figure (1).

Table (1): The mean absorbance value for each specimen and it correspondent concentration of CEA.

CEA (ng/mL)	Absorbance (450 nm)
0	0.000
1.5	0.113
3	0.236
6	0.493
15	1.080
30	2.005

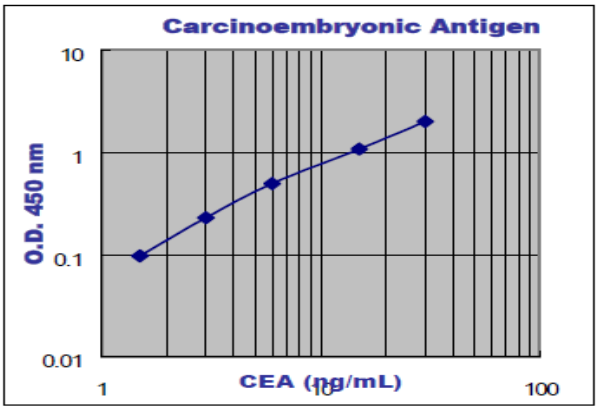

Figure 1: A standard curve constructed for CEA by plotting the absorbance values on the vertical (or Y) axis and the concentrations on the horizontal (or X) axis

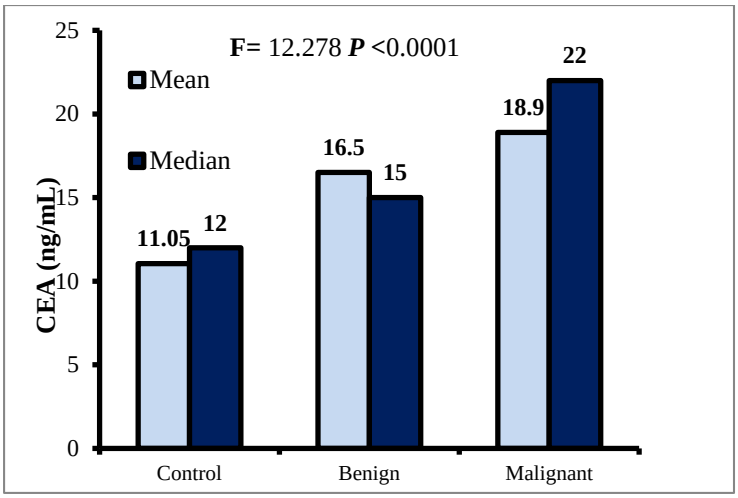

Figure 2: Levels of CEA in the three investigated groups

3 RESULTS

In the current study the tumor marker CEA was measured using the commercial kit as reported earlier in the Materials and Methods section of the present study among three investigated groups; healthy control individuals (n=20), benign breast lesion (n=30) and primary breast cancer (n=100).

3.1 Level of CEA in the three investigated groups

The levels of CEA were reported for all groups. Significant difference was detected (F=12.278, P < 0.0001) among the three investigated groups as CEA level was highly-detected in breast cancer patients followed by the benign, then the control individuals as illustrated in figure (2).

Statistical analysis was carried out by Chi-Square test and P > 0.05 is considered non-significant; P < 0.05 is considered significant.

3.2 The percentage of CEA in the different groups of the study

As illustrated in figure (3), percentage of individuals with CEA level > cutoff value significantly higher in malignant group (77%) compared to benign group (40%) and control group (5.0%) while percentage of individuals with CEA level ≤ cutoff value significantly higher in control group (95%) and benign group (60%) compared to malignant group (23%) (X^2 = 42.25, P < 0.0001).

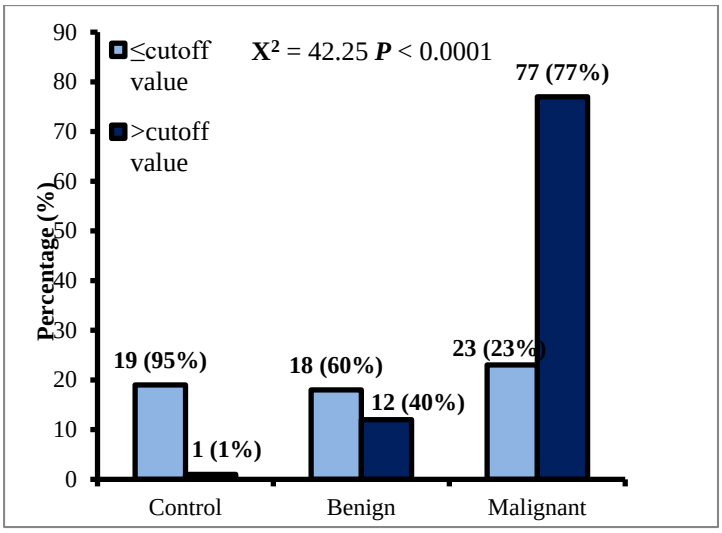

Figure 3: The percentage of CEA in the three investigated groups.

4 DISCUSSION

Breast cancer is the most commonly diagnosed cancer and the leading cause of cancer death among females around the world [14] and is also considered as the most frequent cancer among Egyptian females [15].

A biomarker is a substance in a person's blood, urine, or other body fluids. It can also be found in or on the tumor. A biomarker, sometimes called a tumor marker, is made by the tumor or by the body in response to the cancer. Biomarkers help doctors learn more about each person's cancer so they can recommend the best treatment options for each patient as it represents a measurable characteristic which should be characterized by high specificity and sensitivity, reliability and should be easy to measure. Cancer biomarkers have tremendously increased the efficacy of treatment and efficacy of detection [16].

In this current study, the level of CEA , the most commonly used biomarkers in breast cancer, was studied in serum samples from three groups, all were females and categorized according to their pathological classification into primary breast cancer, benign breast lesions and healthy controls.

The present study revealed that serum levels of CEA were significantly increased in breast cancer group as compared with the other two studied groups; benign and control groups. Significant difference was detected with value ($P < 0.0001$) among them as CEA was highly-detected in breast cancer patients followed by the benign, then the control individuals.

Shao et al [7] reported that elevated serum levels of CEA were identified in breast cancer patients compared to healthy controls. Thus, that study elucidates their common use as routine tumor biomarkers for breast cancer. Other studies presented by Lee et al [17] and He et al [18] showed that significant elevated levels of serum CEA in patients with malignant breast tumors, but not in the patients with benign tumors which could be a potential biomarker for breast cancer monitoring, these results were in agreement with the findings of the current study.

Without more powerful serum markers, although imperfect, CEA remains the most commonly used biomarkers in breast cancer and are recommended for practical use by the American Society of Clinical Oncology (ASCO) [19]. A recent study by Bayo et al [20] concluded that significant differences were found in patients with higher levels of the conventional markers, CEA and CA15.3 in breast cancer patients than healthy control individuals, these results were in agreement with the findings of the current study.

5 CONCLUSION

CEA remains the most commonly used biomarkers in breast cancer and still keeps its diagnostic role in practical use for breast cancer patients. With appearance of many new tumor markers, CEA cannot be completely replaced by another biomarker even it cannot be used alone so, association of the CEA use with other tumor markers is better for proper prognosis, diagnosis and screening of breast cancer.

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