

The Role of Some Enzymes in the Early Diagnosis of Patients with Colon Cancer

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Abstract:

Colon cancer is considered a malignant neoplasm that ranks third after breast and lung cancer in terms of its prevalence in the world. It is the second cause of death for women, the third for men, and the fourth in the world. Blood samples (100) were collected from the local Baghdad Hospital. they were divided into two groups. The first group includes 30 patients of colon cancer ages ranging from (20 - 40) with 20 healthy people as the control group as the same age and the second group includes 30 patients of colon cancer ages (41 - 60) with 20 healthy as control. Biological evidence to help in the early diagnosis of the disease was measured. The measured level of enzymes (Tyrosine kinase, Ornithine Decarboxylase, 5'-Nucleotidase, Gamma-Glutamyl Transpeptidase) for the two groups with their control also and from the results showed an increase in the concentration of enzymes in the serum of colon cancer patients compare with the control group.

List of Abbreviations ;

TK, Tyrosine kinase; ODC, Ornithine Decarboxylase; 5-NT, 5-Nucleotidase; GGT, Gamma-Glutamyl Trans Peptidase.

Keywords:Colon cancer, Oxidative stress, Enzyme biomarker, Antioxidant enzymes

INTRODUCTION

Colon cancer is one of the widespread health diseases in the world affecting men and women, and men affect 3 or 4 times more than women and it is the fourth cause of death globally (Rawla, Sunkara et al. 2019). The main function of the colon is absorption (water and nutrients) due to the occurrence of genetic or epigenetic changes that lead to cancer, in the beginning, is an adenoma, and then it develops into a malignant tumor that takes 5 to 10 years to turn into a metastatic tumor (Ansa, Coughlin et al. 2018). Colon cancer has three types are either sporadic, familial, or hereditary (Mármol 2017). Increase in the incidence of this disease for people who suffer from inflammatory bowel disease and people who have a genetic history of colon cancer and people who follow an unhealthy diet in addition to frequent smoking and drinking alcohol (Keum and Giovannucci 2019). In developing countries, the incidence of colon cancer increases, and the number of deaths decrease to raise awareness to prevention by reducing the risk factors, correct diagnosis in early-stage, and choose the appropriate treatment for each case,

because colon cancer has 4 stages of tumor development, and survival rate for those infected is five year(Gandomani, Aghajani et al. 2017)].Oxidative stress occurs when there is a disturbance in the balance between oxidizing factors antioxidant factors and it is one of the important reason for the emergence of several diseases, including cancer(Sies 2019).A change in the level of concentrations of these antioxidant enzymes (tyrosine kinase, ornithine decarboxylase, 5-Nucleotidase, Gamma-Glutamyl Transpeptidase) as diagnostic markers for colon cancer

Materials and methods

Study group

60 samples were collected from colon cancer patients from a local hospital in Baghdad, Iraq, and they were divided according to ages into two parts, the first section includes ages (20 - 40) and the second section includes ages (41 - 60).

Control group

40 samples were collected from healthy people from a local hospital in Baghdad, Iraq, and they were divided according to ages into two parts, the first section includes ages (20 - 40) and the second section includes ages (41 - 60).

Prepare blood sample

Withdrawal of 5 ml blood samples from the veins of colon cancer patients are placed in the gelatinous tubes and left to coagulate for 20 minutes at room temperature, then these tubes are placed in a centrifuge for 20 minutes at 1000 xg per minute. Serum was withdrawn with microbes for each sample. Half of each sample is examined immediately, or the other half is stored at -20 ° C or 80 ° C for later use.

Level analysis of serum enzymes in the serum of colon cancer patients and the control group:

The level of enzymes (Tyrosine kinase, Ornithine Decarboxylase, 5'-Nucleotidase, Gamma-Glutamyl Transpeptidase) was measured in patients' serum and control by ELISA group on an ELISA system (Biotechnology / USA). Tyrosine kinase was analyzed by Elabscience group ELISA, USA (catalog numbers: E-EL-H0075). Ornithine Decarboxylase was analyzed by the MyBioSource group ELISA, USA (catalog numbers: MBS2021522). 5'-Nucleotidase was analyzed by Elabscience Group ELISA, USA (Catalog numbers: E-BC-K196-M). Gamma-Glutamyl Transpeptidaes was analyzed by the MyBioSource group ELISA, USA (catalog numbers: MBS704964). All analyzes were conducted according to the manufacturer's instructions.

Statistical analysis

Statistical analysis was performed using Graph Pad Prism version 6 (Graph Pad Software Inc., La Jolla, CA). Results were expressed as mean standard deviation (mean \pm SD). A T-test was

conducted to analyze the statistical significance for both groups. P values 0.05 were considered statistically significant.

Results

Tyrosine kinase Enzyme

The results showed the level of tyrosine kinase in the serum (Mean value \pm SD) of colon cancer patients for the first group that includes ages (20-40) was $(38.06 \pm 201.4\text{pg/ml})$ and for healthy people of the same ages it was $(178.32 \pm 38.06\text{pg/ml})$ as shown in figure (1). As for the level of the enzyme in the second group of colon cancer patients, which includes ages (41 - 60), it was $(370.91 \pm 113.45\text{ng/ml})$ and for healthy people of the same ages it was $(153.65 \pm 33.02\text{ng/ml})$ as shown in Figure (2). The results of the two groups showed a significant increase in enzyme concentration. Tyrosine kinase for colon cancer patients compared to healthy people was for the first group (P) value = <0.0001 and for the second group (P-value = 0.0023).

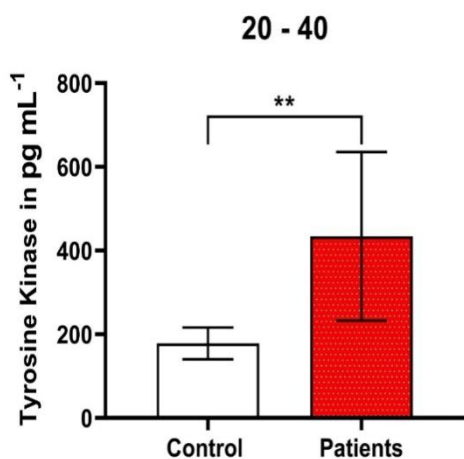


Figure 1. Tyrosine kinase concentration (pg/ml) in control group and colon cancer patients aged (20-40) years.

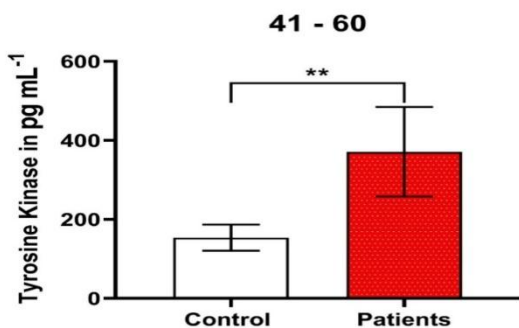


Figure 2. Tyrosine kinase concentration (pg/ml) in control group and colon cancer patients aged (41-60) years.

Ornithine Decarboxylase Enzyme

The increase in the level of the enzyme concentration of ornithine decarboxylase in colon cancer patients of the first group of ages (20 - 40) was (6.23 ± 2.31 ng/ml) compared to the control for the same ages was (2.93 ± 0.58 ng/ml) as shown in Figure (3) and the level of the enzyme in the blood serum of the group The second with ages (41-60) was also increased (11.93 ± 2.61 ng/ml) compared to the control for the same ages also was (3.88 ± 0.45 ng/ml), as shown in Figure (4). The study showed that in both age groups, there was a significant increase in the enzyme level in colon cancer patients compared to control groups where the first group had a (p-value = 0.0003) and the second group (p-value = <0.0001).

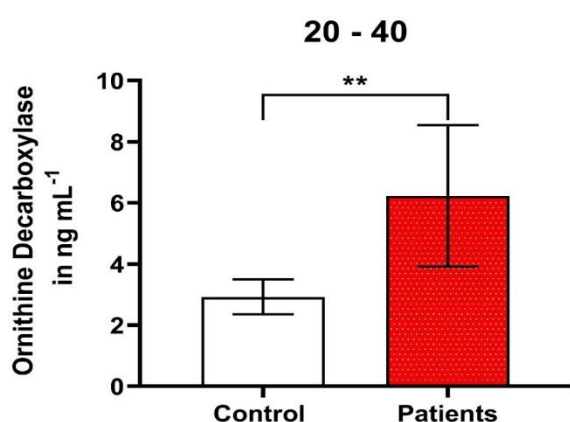


Figure 3.Ornithine decarboxylase concentration (ng/ml) in control group and colon cancer patients aged (20-40) years.

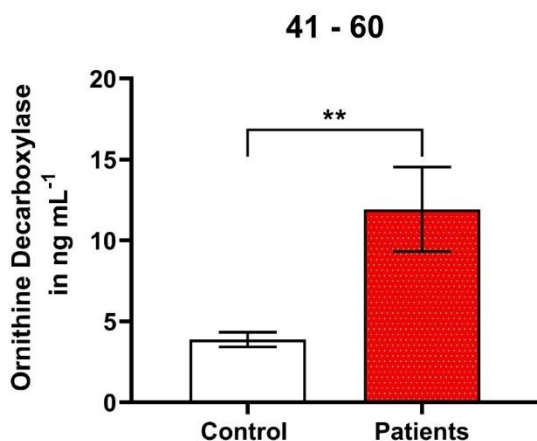


Figure 4.Ornithine decarboxylase concentration (ng/ml) in control group and colon cancer patients aged (41-60) years.

5-Nucleotidase Enzyme

5-Nucleotidase enzyme level for colon cancer patients and control groups ranging in age from (20-40) years were $(9.143 \pm 3.915 \text{ ng/ml})$ and $(3.083 \pm 2.283 \text{ ng/ml})$ respectively as shown in Figure (5). The level of the enzyme was in colon cancer and control groups between the ages of (40-60) years $(9.972 \pm 6.103 \text{ ng/ml})$ and $(5.586 \pm 0.6641 \text{ ng/ml})$, respectively, as shown in Figure (6). (P-value = <0.0001) in the first group and the second group was (P- value = 0.0312).

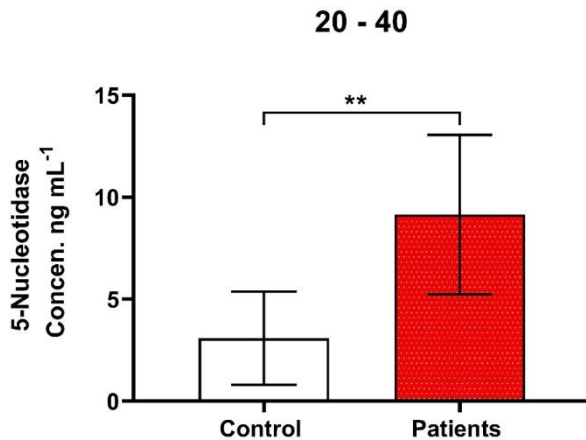


Figure 5.5-nucleotidase concentration (ng/ml) in control group and colon cancer patients aged (20-40) years.

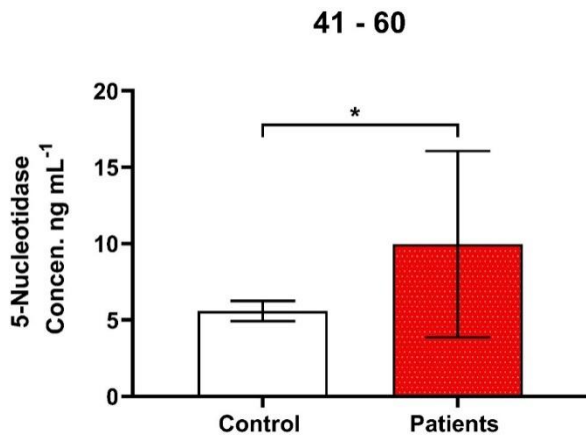


Figure 6.5-nucleotidase concentration (ng/ml) in control group and colon cancer patients aged (41-60) years

Gamma-glutamyl Transpeptidase Enzyme

The results of Gamma-glutamyl Transpeptidase in colon cancer Patients (20-40) was $(2.712 \pm 0.5425 \text{ ng/ml})$ and $(2.603 \pm 2.007 \text{ ng/ml})$ in controls have the same age as shown in the figure (7) whereas the enzyme's level in the colon patients group (41-60) was $(10.49 \pm 4.039$

ng /ml) compared with control as the same age was (4.371 ± 2.416 ng /ml) as shown in the figure (8). This increase was significant with (P-Value =0.0001).

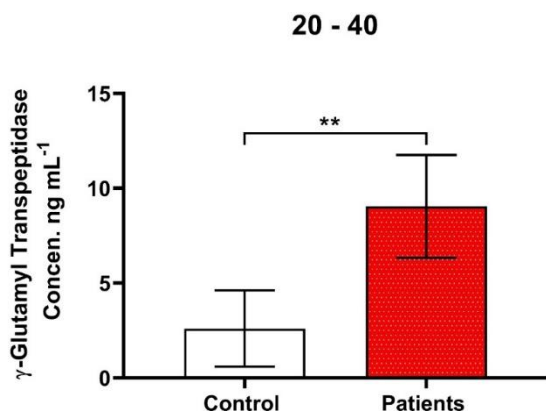


Figure 7.Gamma-glutamyl Transpeptidase concentration (ng/ml) in control group and colon cancer patients aged (20-40) years.

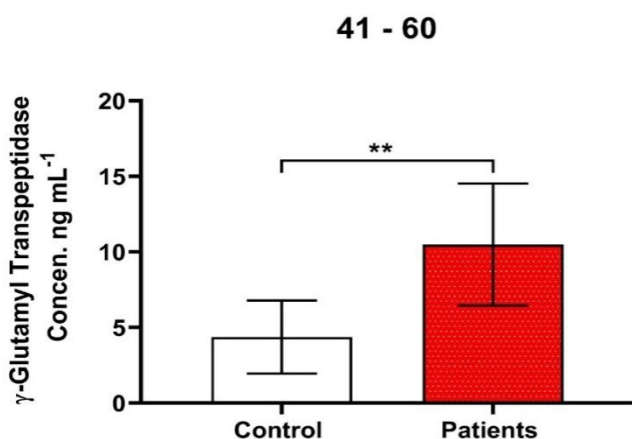


Figure 8. Gamma-glutamyl Transpeptidase concentration (ng/ml) in control group and colon cancer patients aged (41-60) years.

Discussion

Tyrosine Kinase that regulates the signals of most cellular functions such as reproduction, diffusion, migration, differentiation, and apoptosis(Jiao, Bi et al. 2018).The involvement of this enzyme in the formation of tumors and their spread According to the results we obtained, we find a high concentration of the enzyme tyrosine kinase in the serum of colon cancer patients compared to healthy people, and this increase may be caused by a mutation in the enzyme pathway, which leads to a tumor and its rapid growth and regulation of autoclaving and binding of receptors tyrosine kinase with ligands(Esteban-Villarrubia, Soto-Castillo et al. 2020). Our

results are in agreement with a study (Mevizou, Sirvent et al. 2019) that confirmed the increased activity of tyrosine kinase in colon cancer.

The concentration of the enzyme ornithine decarboxylase also increases in colon cancer patients according to our results, as this enzyme affects the cellular functions of the cell, such as the process of cell death and growth signals (Lambertos and Peñafiel 2019). This increase may be due to an increased activity due to response to stimuli of cell growth in cancer, and its activity is also affected by the level of free polyamines and carcinogenic genes (Fan, Feng et al. 2020). A study confirmed increased ODC concentration activity in colon cancer (Jelski and Mroczko 2020).

The 5-Nucleotidase enzyme plays an important role in the carcinogenesis and migration of cancer cells and the development of metastases (Azambuja, Ludwig et al. 2019). Its concentration also increases in colon cancer patients, and this increase may be due to hypoxia and an increase in free radicals and adenosine derived from 5-NT helps to enhance the tumor and its spread and an increase of 5-NT has been linked With poor disease prognosis and short the patient's survival time (Yang, Yao et al. 2021). Our results were consistent with the study (Wang, Fu et al. 2019).

Gamma-glutamyl transpeptidase maintains the balance of oxidative stress and reductions in cell (Castellano and Merlino 2013). Its concentration is also increased in colon cancer patients, and this increase may be due to the enzyme's association with cellular oxidative stress associated with the glutathione pathway, and its activity is also affected by drinking alcohol and smoking and this increase may lead to a genomic alteration For the tumor, which helps the tumor to spread (Hong, Lee et al. 2021). Our results are consistent with a study (He, Fang et al. 2021) that confirmed increased expression of GGT in colon cancer patients. In conclusion The increased level of enzyme tyrosine kinase, Ornithine Decarboxylase, 5- Nucleotidase, and Transpeptidase in the serum of patients with colon cancer, and the results showed a statistically significant increase between the different groups of patients and controls.

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Authors' contributions

Dr.Firas A. Hassan was formulated the current study and responsible for data analysis. Sana was responsible for writing, collecting the samples and the laboratory analyses were conducted at the Research and Development Center of Al-Nahrain University, Baghdad.

Declaration of Conflicting Interests

The authors declared the absence of a potential conflict of interest with regard to research and authoring and / or publication of this article.

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