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Evaluation of kidney function tests and serum levels of carcinoembryonic antigen, polypeptide- specific antigen, and total oxidative stress in Iraqi women colorectal cancer patients with chronic kidney disease

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Abstract

Colorectal cancer is the second leading cause of cancer-related deaths and the third most frequently diagnosed non-skin cancer. Chronic kidney disease can lead to elevated serum carcinoembryonic antigen concentrations. Therefore, this study sought to evaluate how chronic kidney disease affects the prediction accuracy of carcinoembryonic antigen in Iraqi women patients with colorectal cancer. The study aims to measure carcinoembryonic antigen levels, renal function tests, and antioxidant levels in patients with colon cancer. Between February and the end of May 2022, a total of 80 female participants, comprising 40 patients and 40 healthy controls, aged 26 to 82 years—were recruited for the case-control study by the Gastroenterology and Liver Diseases Teaching Hospital A15 Biosystems assays creatinine, uric acid, and urea, while the ELISA assays Tissue peptide-specific antigens, total antioxidant status, carcinoembryonic antigen, and total oxidative status. The results showed that the levels of carcinoembryonic antigen, peptide-specific antigen and total antioxidant status in the patient group were significantly higher *P*-value <0.0001than those in the control group, but their total antioxidant status was significantly lower *P*-value <0.0001 than that in individuals with elevated creatinine, uric acid and urea levels in the control group. This study showed a clear relationship between peptide-specific antigens, total antioxidant status, carcinoembryonic antigen, total oxidative status levels, and renal function (creatinine, uric acid, and urea) and colorectal cancer risk.

Keywords: carcinoembryonic antigen, chronic kidney disease, tissue polypeptide specific antigen, total oxidative status, Colon Cancer, Tumors markers.

تقييم اختبارات وظائف الكلى ومستويات مستضد السرطان الجنيني ومستضد متعدد الببتيد النوعي والإجهاد التأكسدي الكلي في مصل النساء العراقيات المصابات بسرطان القولون والمستقيم وأمراض الكلى المزمنة

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الخلاصة

سرطان القولون والمستقيم يعد ثاني أهم أسباب الوفيات المرتبطة بالسرطان شيوعا، وثالث أكثر أنواع السرطان غير الجلدية تشخيصا . يمكن أن يؤدي مرض الكلى المزمن إلى ارتفاع تركيز مستضد السرطان الجنيني (CEA) في المصل . لذا، سعت هذه الدراسة إلى تقييم كيفية تأثير مرض الكلى المزمن على دقة التنبؤ بمستضد السرطان الجنيني لدى النساء العراقيات المصابات بسرطان القولون والمستقيم .تهدف الدراسة إلى قياس مستويات مستضد السرطان الجنيني، واختبارات وظائف الكلى، ومستويات مضادات الأكسدة لدى مريضات سرطان القولون . بين شهر شباط إلى نهاية شهر ايار 2022، تم اختيار 80 امرأة، تضمنت 40 مريضة و 40 من الاصحاء كحالة ضابطة تتراوح أعمارهن بين 26 و82 عامًا و تم جمع العينات في مستشفى أمراض الجهاز الهضمي والكبد التعليمي– لدراسة الحالات – الاصحاء. استخدم جهاز A15Biosystems لقياس الكرياتينين وحمض اليوريك واليوريا، في حين استخدمت تقنية الاليزا ELISA لقياس مستضدات الببتيد النسيجي النوعي، ومستوى مضادات الأكسدة الكلية، ومستضد السرطان الجنيني، ومستوى الأكسدة الكلية . أظهرت النتائج أن مستويات المستضد السرطاني الجنيني، والمستضد النوعي للببتيد، ومستوى مضادات الأكسدة الكلية في مجموعة المرضى كانت أعلى بشكل معنوي مع قيمة $P<0.0001$ مقارنة بالمجموعة الضابطة، في حين كانت مستوى مضادات الأكسدة الكلية لديهم أقل بشكل ملحوظ مع قيمة $P<0.0001$ لدى المرضى، خصوصا لدى الأفراد الذين يعانون من ارتفاع مستويات الكرياتينين وحامض اليوريك واليوريا في المجموعة الضابطة .

تشير هذه الدراسة الى وجود علاقة واضحة بين مستويات المستضدات النوعية للببتيد، ومستوى مضادات الأكسدة الكلية، والمستضد السرطاني الجنيني، ومستويات حالة الأكسدة الكلية، ووظائف الكلى (الكرياتينين، وحمض اليوريك، واليوريا) وخطر الإصابة بسرطان القولون والمستقيم.

1. Introduction

According to the World Health Organization (WHO), cancer is the leading cause of death globally. In addition, colorectal cancer (CRC) is recognized as one of the leading causes of death in the world. A variety of techniques and approaches have been used to treat CRC and other forms of cancer [1]. In 2020, the World Health Organization reported 1.93 million cases of CRC, with 935,000 confirmed deaths, making CRC the third most common type of cancer [2]. CRC is among the most prevalent cancers globally. It ranks second in second second-highest mortality rate and the fourth highest morbidity rate [3]. CRC occurs in the glandular epithelial cells of the large intestine and is also known as colorectal adenocarcinoma. Cancer arises when a series of epigenetic or genetic changes occurs in specific epithelial cells. Due to abnormally high rates of replication and survival, the over proliferating cells give rise to benign adenomas that can subsequently develop into cancer and spread over decades. [4]. CRC is a genetically heterogeneous malignancy, with approximately 40% of cases harboring KRAS and an additional 10% harboring BRAF oncogenic mutation [5]. Age has a greater impact on the incidence of CRC than other demographic factors. Rarely is sporadic colorectal cancer discovered before the age of 40. 90% of occurrences of this illness occur beyond the age of 50, and its prevalence has grown significantly between those ages. As a result, colorectal cancer mortality starts to climb gradually around the age of five and quickly as people become older [6]. The increased rate of colon cancer patients in industrialised nations is linked to factors that increase the risk of carcinogenesis, including inactivity, an unhealthy diet high in fat and calories, obesity, and a sedentary lifestyle [7]. One conjugated polypeptide chain makes up TPS (tissue polypeptide specific antigen). It is created throughout the molecular cycle's S and G2 phases and released into the cells following mitosis[8]. TPS is used in the diagnosis and management of chemotherapy for gastrointestinal tract cancers, primarily colorectal and pancreatic. The relationship between TPS concentration in blood and neoplasm cell proliferation depends on the rate of cell division[9]. An elevated level of TPS in tumors denotes

tumor hyperplasia before the formation of the mass[10]. To assess the overall level of oxidative stress, total oxidant status (TOS) and total antioxidant status (TAS) are markers of oxidative stress [11]. According to Pere, et al.[12], a variety of endogenous and external variables may have an impact on oxidative state and modify the capacity of gut epithelial cells to withstand harmful metabolic stresses. Although several studies have shown that oxidative stress plays a significant role in carcinogenesis [13, 14], the authors were unsure if oxidative stress is a result of the pathophysiology of CRC. This prompted us to look into the antioxidant content of CRC patient sera. A glycoprotein called CEA is created in the large bowel's cells. A highly useful marker for the therapy and monitoring of the illness, elevated CEA levels are present in 70% of CRC patients at the time of diagnosis [8]. Metabolic byproducts like serum urea, uric acid, or creatinine- commonly used to assess kidney function, may also provide important insights into the etiology of cancer [15]. Comparable differences in blood urea levels, the last metabolite formed after the breakdown of amino acids, indicate that this measure may be utilized as a replacement for other early detection methods for these kinds of malignancies [16]. The breakdown of arginine and glycine produces creatinine, which has been studied in several malignancies and found to have prognostic properties in a number of epithelial tumors [17]. Along with free purine nucleotides, hypoxanthine, and xanthine, uric acid is the last byproduct of the nucleotide metabolic cycle, which denotes the catabolism of nucleic acids. Due to its antioxidant properties, uric acid is the most significant molecule according to our prior understanding [18,19] discovered that uric acid levels were substantially correlated with the prognosis of colon cancer patients. The study aims to measure carcinoembryonic antigen levels, renal function tests, and antioxidant levels in patients with colon cancer.

2. Materials and Methods

The study was conducted from February to the end of May 2022, and the age range of patients and healthy individuals was 26-82 years. Patients were recruited from a Gastroenterology and Liver Diseases Teaching Hospital in Baghdad, Iraq. The symptoms and signs of the disease were diagnosed by specialist doctors at the hospital, and the patient was clinically and histologically diagnosed with colorectal cancer (stage III). The patient's complete medical history was evaluated to exclude any existing systemic disease or ongoing treatment that could affect the biochemical parameters to be measured. Approximately 5 mL of venous blood was collected from each person, allowed to stand for 15 min and centrifuged at 3000 rpm for 10 min. The resulting serum was stored at -20°C for further analysis.

Serum creatinine, uric acid, and urea levels were determined using the protocol of the commercially available A15 biosystems (Spain). Automated ELISA microplate reader analyzer [model IRE 96-SFRI - MedicalExpo (Spain)] using the instructions of the commercially available MyBioSource. (UK) product used to measure TPS, TAS, CEA and TOS concentrations in serum.

3. Statistical analysis

The Statistical Software for the Social Sciences (SPSS) program version 20.0 was used to compare the two groups and analyze all data using descriptive statistics and t tests. Standard errors (SE) are used to express the values obtained. To determine the correlations among all study variables, Pearson correlation analysis was also performed. The statistical test was significant at $p \leq 0.05$ and highly significant at $p \leq 0.01$ with a 95% confidence interval.

Results

All data obtained were statistically analyzed and presented as mean ± standard error. The differences in P values were also included to verify the levels of the studied parameters between

colorectal cancer patients and controls. Table 1 presents the calculated means ± SE for control and patient sera (TPS, TAS, CEA, and TOS).

The results demonstrated that, compared to the control group, patients with colorectal cancer exhibited significantly elevated serum levels of TPS, TAS, CEA, and TOS ($P < 0.0001$).

Table1: Levels of parameter in colorectal CA patients and controls.

Parameter	Mean±S.E		P-value
	Control No.=40	Patient No.=40	
TPS(ng/ml)	33.98±1.22	255.87±3.99	0.0001
TAS(U/ml)	24.73±1.19	6.19±2.38	0.0001
CEA(ng/dl)	68.55±4.08	364.48±12.43	0.0001
TOS(U/mL)	11.5±0.52	102.05±2.62	0.0001

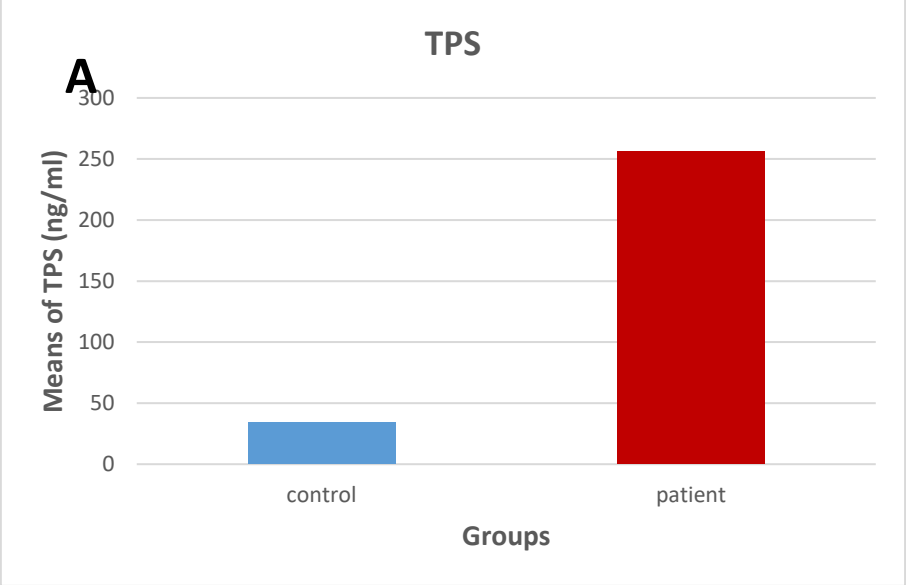

Figure 1: TPS ng/ml in colorectal CA patients and controls

Renal function tests

The results presented in Table 2 indicate significant differences in creatinine, urea and uric acid between the patient group and the control group ($P=0.0001$), with the patient group exhibiting higher concentrations of these markers compared to the healthy group.

Table 2: Levels of Renal function tests in colorectal CA patients and controls

parameter	Mean±S.E		P-value
	Control No.=40	Patient No.=40	
Creatinine(mg/dl)	0.85±0.02	3.08±0.16	0.0001
Urea (mmol/L)	3.38±0.07	7. 41±0.17	0.0001
Uric acid(mg/dl)	4.68±0.11	9.65±0.05	0.0001

Pearson correlation analysis was used to examine the correlations between all parameters considered in this study. Table 3 summarizes the final results.

Table 3: Correlations (r values) between variables in the colorectal CA group of patients

Parameters	TPS	TAS	CEA	TOS	Creatinine	Urea	Uric acid
TPS	**1**	**0.085**	**0.272**	**0.205**	**-0.159**	**-0.120**	**-0.157**
TAS	**0.085**	**1**	**0.113**	**0.086**	**0.432****	**-0.313***	**-0.320***
CEA	**0.272**	**0.113**	**1**	**-0.105**	**-0.171**	**-0.431****	**0.115**
TOS	**0.205**	**0.086**	**-0.105**	**1**	**-0.069**	**0.154**	**-0.163**
Creatinine	**-0.159**	**0.432****	**-0.171**	**-0.069**	**1**	**0.102**	**-0.392***
Urea	**-0.120**	**-0.313***	**-0.431****	**0.154**	**0.102**	**1**	**-0.105**
Uric acid	**-0.157**	**-0.320***	**0.115**	**-0.163**	**-0.392***	**-0.105**	**1**

****. Correlation is significant at the 0.01 level.**
***. Correlation is significant at the 0.05 level.**

This study found that serum TAS was significantly positively correlated with creatinine, and serum CEA was highly significantly positively correlated with urea and uric acid (P>0.01). The final analysis showed that uric acid and creatinine were negatively correlated with TAS (P>0.05).

Discussion

Colon cancer is an abnormal growth of cells in the large intestine. Sometimes growth is restricted for a relatively long time before becoming malignant and then spreading through the intestinal wall to lymph nodes and other parts of the body [20,21]. Colon cancer and rectal cancer are usually grouped because they have many common features [22]. TPS may be particularly suitable for early tumors. Among 60-80 % of CRC patients, the concentration of TPS increases [23]. Seventy-five percent of the patients showed elevated TPS levels, confirming its association with colon cancer and rectal cancer. Among the patients with high levels of TPS, the survival time is significantly short [24], which is consistent with our study. Since numerous studies have demonstrated that oxidative stress plays a significant role in carcinogenesis, the authors are unsure as to whether oxidative stress is a result of the pathogenesis of colorectal cancer [25].

A large number of free radicals, decreased antioxidant activity, increased oxidative stress, interference with data measurement, and experimental errors are all side effects of radiotherapy and chemotherapy, which can inhibit or kill cancer. Additionally, J. F.Feng, etal showed that chemotherapy-treated CRC patients had significantly lower total antioxidant capacity (TAC) than chemotherapy-untreated CRC patients. [26]
Studies that only focus on TAS are ineffective, in addition to experimental influence factors. Independent tests of TAS or TOS may not accurately reflect a subject's OxS status, according to a previous study. The body's oxidation-antioxidant system, whether at the tissue or cellular level, can maintain a dynamic balance when TAS and TOS increase or decrease concurrently, explaining why these values by themselves do not result in oxidative stress. At this point, OSI can more accurately reflect the subject's level of oxidative stress. TAS and TOS should be decided upon simultaneously as a result. [27].

Many epithelial tumors express a glycoprotein called carcinoembryonic antigen (CEA). The first description of this affordable blood test was published by Gold and Freedman in 1965 and is part of most recommended monitoring programs. 70% of CRC patients have elevated CEA levels at diagnosis, making it an excellent marker for post-resection treatment and disease monitoring [8]. Although CEA is generally considered a cancer marker, a study conducted in Taiwan by Chen et al investigated whether elevated CEA has other benefits. In a study of 4841

patients, 999 had elevated CEA levels [28]. According to Yaghoubi et al. [29], which is consistent with this study, CEA has been used for many years as a trustworthy and valid biomarker for differential diagnosis and monitoring purposes in CRC patients. TPS was also more sensitive than CEA in detecting CRC recurrence. In addition, patients with progressive disease had higher levels of CEA and TPS compared with patients without evidence of disease progression. As any decline in renal function will increase these parameters in the blood, evaluation of renal function tests (urea and creatinine) is crucial for the diagnosis and treatment of cancerous diseases. Increased blood levels of urea and creatinine signify a compromised kidney or kidney disease (brackets). Furthermore, renal function parameters (urea and creatinine) are also important diagnostic markers during cancer development. Therefore, careful monitoring of these biochemical parameters plays a crucial role in the prognosis of the disease [30].

Additionally, a study by 33 reported lower urea levels in patients with colon cancer, which contrasts with the findings of the present study.

Dziamn et al. found that uric acid levels were significantly correlated with the prognosis of patients with colon cancer. [31]. However, only a few epidemiological studies have investigated this association. Ko et al did not report evidence of a protective association between SUA and colon cancer. [32], which conflicts with this finding.

Similarly, according to a cohort study of Japanese men conducted in Hawaii, there was no association between SUA levels and the risk of colorectal cancer [33]. On the other hand, in a large Swedish cohort study, [34] reported that SUA levels were positively correlated with colorectal cancer incidence. This is consistent with our findings in this study.

Conclusion

In conclusion, this study showed a clear association between TPS, TAS, CES, TOS levels and renal function markers (creatinine, uric acid, and urea) with the risk of colorectal cancer. CEA is the most commonly examined marker when a gastrointestinal tumor is suspected, and elevated serum CEA levels may be associated with carcinogenesis. For 50% of patients, this is an indicator of tumor recurrence after tumor removal.

Ethical Approval:

On November 14, 2021, the local ethics committee approved this study, which adheres to the ten ethical principles outlined in the Declaration of Helsinki. The Scientific Committee supported this study at the Department of Chemistry/College of Sciences for Women, University of Baghdad.

Conflict of interest

The authors declare that they have no conflict of interest.

- Ethical Clearance: The project was approved by the local ethical committee in University of Baghdad.

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