

**Ministry of Higher Education
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College of Science**



Useing Some Biochemical and Biomolecular Markers for Detection of Colon Cancer

**A Thesis Submitted to the Council of College of Science/
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in
Biochemistry**

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Abstract

Study includes (104) patients diagnosed with colon cancer proved by colonoscopy and histopathology. Blood and tissue samples have been selected consecutively over the period from March 2013 to April 2015 from patients treated in Oncology and Nuclear Medicine hospital, Al-zahrawi teaching hospital/Mosul and Azadi Teaching Hospital/Kirkuk. All cases and controls were aged (15-85 years). Normal (100) blood donor individuals have been used as control. The demographic distribution of colon cancer according to age: 73.1% older than 50 year-old and 26.9% were younger than 50 year-old. The incidence of males was higher than females in all age groups. In addition, the distribution according to residence: colon cancer patients who live in rural areas that comprise 70% of all patients were significantly higher than patients in urban areas. Also the results showed that the majority of colon cancer patients were smokers and the anatomical site of the tumor localized in rectosigmoid was in 71 patients. Biochemical markers were classified into four groups which are: enzymatic tumor markers, Antioxidant enzymes, Trace elements and some biochemical parameters. The results showed that patients with early significantly ($p < 0.05$) increase activity of Cyclooxygenase 2 and Arginase for colon cancer patients compared with controls. This study also includes the measurement of the serum alkaline sphingomyelinase level which significantly decreased ($p < 0.05$) in colon cancer patients in stage A compared with controls and this decrease was independent of Dukes' stage, thus strengthening the hypothesized validity of these assays to be used as serum test for the early detection of colon cancer. A significant increase ($p < 0.05$) was observed in the activities of (Alkaline phosphatase, Lactate dehydrogenases and α -amylases), in comparison with control. In addition, significant increase ($p < 0.05$) in concentration of triglyceride and total proteins in comparison with control and a significant decrease ($p < 0.05$) in Cholesterol, High density lipoprotein, Low density lipoprotein and albumin concentration compared with control. A significant increase ($p < 0.05$) is also seen in the sialic acid and the early stages B. So it can be considered as markers for the diagnosis of colon cancer associated with early stages. In addition, the results demonstrated a significant decrease ($p < 0.05$) in the activities of antioxidant enzymes paraoxonase and arylesterase in patients when compared with controls. Trace elements revealed that there was a significant decrease ($p < 0.05$) in concentration of Zn when compared with controls, while a significant increase ($p < 0.05$) in concentration of Cu and Mg in patients when compared with controls. All these results reflect was seen the initial changes occurring in neoplastic

conditions. Antigenic tumor markers include carcino-embryonic antigen and Carbohydrate antigen 19-9. Measurement in blood serum from 104 patients with colon cancer indicate significant elevation ($p < 0.05$) in levels of CEA and C19-9 only in patients with advanced stage C and metastatic stage D compared with control. Study showed a significant increase ($p < 0.05$) in free DNA in all stages of colon cancer patients compared with control group. Tissue samples have been obtained, from colon cancer patients for each duke's stage. Genomic instability was detected: By Random amplified polymorphic DNA method by using five primers (OPE-07, OPA00, OPA-13, OPD-20 and OPA-10). Three primers only show genetic instability OPE-07, OPA-00 and OPA-13 based on the separation of amplified products on agarose gel for each mass and serum samples in early stage (A and B), advance stage C and metastasis stage D compared with normal tissues. Therefore, OPE-07, OPA-00 and OPA-13 appears to be useful as genetic markers. Genetic instability was also observed in serum which was revealed by the deletion or insertion of amplified DNA fragments from seru DNA patient compared with normal tissue. This serum sample might be used for diagnosis colon cancer patient when it was difficult to get tumor tissue from patients. On the other hand, the highest genetic instability is represented by samples stage A and B of tumor tissues which reflects a lot of DNA damages or changes happened during the development of cancer. Moreover, a deletion of the same fragment has been found in the serum samples using the same primers OPE-07 and OPA-13. The most important finding was the association between the level of genomic instability and colon cancers Duckes stages and the utilization of RAPD analysis could be an important method for detecting genetic instability in colon cancer tissues and patient's serum especially in the early stages of the disease and is considered as a useful diagnostic method for early detecting of colon cancer.