SUST
Journal of Natural and Medical Sciences
Journal home page:
http://Scientific-journal.sustech.edu/

The Impact of Colorectal Carcinoma Chemotherapy on Serum Levels of Carcinoembryonic Antigen and Carbohydrate Antigen 19-9 among Sudanese Patients

Waleed A. Salim, [1] A. H. Khatab, [2] Abdelgadir Elmugadam, [3] Omer Fadl Idris [4]

[1] Preparatory year, Al Jouf University. [2] Faculty of medicine University of Khartoum. [3] College of Medical laboratory Science. Sudan University of Science and Technology, [4] Faculty of Science and Technology. Al-Neelain University.

*Corresponding author: Email: **ahmed.wali@ymail.com**

Received: Septmber 2018 **Accepted: November 2018**

Abstract:

Colorectal cancer (CRC) is one of the most common cancer related causes of morbidity and mortality in developed countries. Tumor markers are substances synthesized and excreted by tumor tissues that can be detected in abnormal concentrations in serum, urine, and other body fluids. This study attempted to investigate whether CEA and CA 19-9 levels can be used to monitor the response of chemotherapy in patients with colorectal cancer (CRC). This study included 100 patients with CRC admitted to National Cancer Institute (NCI-Sudan) to receive chemotherapy, the concentration of CEA and CA 19-9 levels were measured pre and post chemotherapy in which all patients with CRC received chemotherapy with a 5-fluorouracil (5-FU). Patients with CRC showed a highly significant increase ($p < 0.01$) of CEA levels between pre and post chemotherapy in early stage (Stage I; $p=0.009$ and stage II; $p=0.001$). And also there was a significant increase ($p < 0.05$) between pre and post chemotherapy of CA 19-9 levels in early stages (Stage I; $p=0.045$ and stage II; $p=0.017$), but there was no significant difference found between pre and post chemotherapy in the levels of CEA and CA 19-9 in more advanced tumor stage. These data suggest that early stages of CRC patients are of more response to chemotherapy than late stage and also CEA is better than CA 19-9 in flow of patients with CRC especially in advanced stage.

Keywords: Colorectal cancer, CEA, CA 19-9, follow up.

المستخلص

سرطان القولون والمستقيم هو واحد من أكثر الأسباب الشائعة المرتبطة بالوفيات في البلدان النامية. واسمات الاورام هي المواد التي يتم انتاجها وإفرازها بواسطة أنسجة الورم التي يمكن اكتشافها بتركيزات غير طبيعية في مصل الدم والبول وسوائل الجسم الأخرى. الهدف من هذه الدراسة معرفة ما إذا كان من الممكن استخدام مستويات CEA و CA 19-9 لمراقبة استجابة العلاج الكيميائي في مرضى سرطان القولون والمستقيم. شملت هذه الدراسة 100 مريض مصابين بسرطان القولون والمستقيم يترددون علي المعهد القومي للسرطان – السودان لتلقي العلاج الكيميائي ، حيث تم قياس تركيز مستويات CEA و CA 19-9 قبل وبعد العلاج الكيميائي حيث تلقي جميع المرضى الذين يعانون من سرطان القولون والمستقيم العلاج الكيميائي من النوع 5-fluorouracil (5-FU). كشفت هذه الدراسة وجود ارتفاع ملحوظ ذو دلالة احصائية معنوية حيث كان الاحتمال الاحصائي للمقارنة اقل من 0.01 لمستويات CEA بين ما قبل وبعد العلاج الكيميائي للمراحل المبكرة للمرض (المرحلة الاولي ؛ P = 0.009 والمرحلة الثانية ؛ P = 0.001). وكذلك كانت هنالك

زيادة معنوية ذات دلالة حيث كان الاحتمال الاحصائي للمقارنة اقل من 0.05 لمستويات CA 19-9 بين ما قبل وبعد العلاج الكيميائي للمراحل المبكرة للمرض (المرحلة الاولي ؛ P = 0.045 والمرحلة الثانية ؛ P = 0.017). لكن لم يكن هناك فرق كبير ذو دلالة قبل وبعد العلاج الكيميائي في مستويات CEA و CA 19-9 في مراحل الورم الأكثر تقدما. خلصت هذه الدراسة الي ان المراحل المبكرة من مرضي سرطان القولون والمستقيم هم اكثر استجابة للعلاج الكيميائي من المراحل المتاخرة. وكذلك CEA افضل من CA 19-9 في متابعة المرضي المصابين بسرطان القولون والمستقيم خاصة في المراحل المتقدمة من المرض.

Introduction

Colorectal cancer (CRC) is the second most common cancer in women and the third most common cancer in men worldwide (Siegel et al. 2012). CRC is one of the most significant health problems throughout the world, with an estimated 1.2 million new cases and 0.6 million deaths each year (Jemal and Bray. 2011). Also colorectal cancer (CRC) is the fourth most common solid carcinoma diagnosed in the United States (Wang, et al. 2014).

Tumor markers (TMs) play an important role in cancer diagnosis, prognosis, treatment, and monitoring (Carpelan-Holmstro¨m. et al, 1996; Levy et al. 2008; Du et al. 2010). They are proteins released from dying tumor cells or produced by neoplastic cells. There are two subcategories of these proteins, specific and non-specific. The specific proteins are expressed only in the tumor cells and are very useful for the detection and diagnosis of specific malignant tumors. Non-specific proteins or markers related to malignant cells are oncofetal or carcinogenic antigens, such as carcinoembryonic antigen (CEA), alphafetoprotein (AFP), prostate specific antigen (PSA), carbohydrate antigens CA15.3 and CA19-9 (Osaka et al. 2009).

Carcinoembryonic antigen (CEA) is the most common tumor marker used in patients with colorectal cancer (Filella., et al. 1992) CEA is a high molecular-weight glycoprotein belonging to the immunoglobulin superfamily of molecules. It functions as an intercellular adhesion molecule promoting the aggregation of human colorectal carcinoma cells (Duffy et al. 2003; Hanke B et al. 2001).

CEA is increased in approximately 60% to 85% of the patients with colorectal cancer (Filella et al. 1992; Hanke et al. 2001). CEA has a specificity for colorectal cancer of 90%, but a sensitivity of only 40% to 75%. (Duffy et al. 2003; Hanke B et al. 2001).

An additional marker to monitor colorectal cancer is carcinoma antigen (CA) 19-9. CA 19-9 was described by Koprowski et al in 1979 as a monoclonal antibody, raised against a human colorectal cancer cell line (Jolanda Stiksma et al. 2014). Increased serum levels of CA19-9 have been described in association with a range of gastrointestinal malignancies including colorectal carcinoma (Knopf et al. 2001). CA 19-9 is commonly used in the management of pancreatic cancer. CA 19-9 is also increased in approximately 35% to 40% of patients with advanced colorectal cancer (Hanke et al. 2001).

In the Sudan little is known about the predictor values of CEA and CA 19-9 as monitoring and prognostic markers among colorectal cancer patients especially pre and post operation, in addition to monitoring chemotherapy treatment. Therefore the present study aimed to assess whether these tumor markers CEA, CA 19-9 are of value in monitoring the progress of patients being treated with systemic chemotherapy for colorectal cancer.

Materials and Methods

A cross-sectional hospital based study was performed between June 2016 and February 2017. A total of 100 patients with colorectal carcinoma (55 males and 45 female) admitted to National Cancer Institute (NCI-Sudan) to receive chemotherapy, the median age was 52

years (range 11–80 years) were enrolled in this study.

Interview with CRC patients was done before starting chemotherapy which included full history (age, sex, exposed to surgery, site of tumor, onset of disease and stage of tumor).

A questionnaire was specifically designed to obtain information which helped in either including (patients of colorectal cancer admitted to National Cancer Institute to receive chemotherapy) or excluding (patients of colorectal cancer admitted to National Cancer Institute had jaundice, anemia (HB less than 9gm/dl, TWBCs less than 3000/cumm, platelet less than 70, vomiting and diarrhea) certain individuals in or from the study respectively.

The ethical clearance was approved by the Research Ethical Committee of the Ministry of Health (Sudan). Data and samples were collected after informing and agreement of colorectal cancer patients about the purpose and importance of the study.

The specimen was serum; 5 ml venous blood was collected in plain container (without anticoagulant) before chemotherapy and after completion of the dose of chemotherapy. After had been allowed to stand at room temperature for one hour to obtain serum.

Tumor markers Carcinoembryonic antigen (CEA) and CA 19-9 were determined by electrochemiluminescence Immunoassay (ECLIA) using commercial kits from Roche Company (Germany) and Cobas e 411Immunoassay Analyzer.

CEA and CA 19-9 present in the test sample bound with a biotinylated monoclonal specific antibody, and a monoclonal specific antibody labeled with a ruthenium complex form a sandwich complex. After addition of streptavidin-coated microparticles, the complex became bound to the solid phase via interaction of biotin and streptavidin. The reaction mixture was aspirated into the measuring cell where the microparticles were magnetically captured onto the surface of the electrode. Results were determined via a calibration curve which was instrument specifically generated by 2-point calibration and a master curve provided via the reagent barcode.

Each tumor was histopathologically classified according to the American Committee Cancer Classification and Staging System. Results were tabulated, analyzed and compared using paired sample T test with p value being considered significant at $p < 0.05$.

Results

One hundred patients (55 males and 45 females) aged 11–80 years (median 52) were studied. 50% of patients had been exposed to surgery before the start of chemotherapy (adjuvant therapy) and 50% of patients had not been exposed to surgery before the start of chemotherapy.

The data collected in this study analyzed using SPSS computer program. The mean and standard deviation of the serum concentration of CEA and CA 19-9 were measured pre and post chemotherapy of CRC patients. Results were tabulated, analyzed and compared using paired sample T test with p value being considered significant at $p < 0.05$.

Results of CEA and CA 19-9 pre and post chemotherapy in different stages showed a significant increase ($p < 0.05$) in serum level of CEA in cancer patient's stages I, II and III for CEA and stage I and II only for CA 19-9. But only stage IV for CEA and stage III and IV was not significant in case of CA 19-9. ***(Table 1&2).***

Table 1: Results of CEA pre and post chemotherapy in different stages:

Stage		**Mean ± SD (ng/ml)**	**P value**
I	Pre chemotherapy	19.7±8.2	0.009
	Post chemotherapy	10.7±7.3	
II	Pre chemotherapy	25.4±28.6	0.001
	Post chemotherapy	13.3±13.0	
III	Pre chemotherapy	26.3±17.5	0.048
	Post chemotherapy	18.0±27.3	
IV	Pre chemotherapy	106.1±115.8	0.288
	Post chemotherapy	127.9±159.1	

- The table shows the mean ± SD and probability (P).
- t- Test was used for comparison.
- P < 0.05 considered significant.

Table 2: Results of CA 19-9 pre and post chemotherapy in different stages:

Stage		Mean ± SD (U/ml)	P value
I	Pre chemotherapy	32.9 ±20.8	0.045
	Post chemotherapy	27.7±19.1	
II	Pre chemotherapy	27.4±40.4	0.017
	Post chemotherapy	19.8±24.3	
III	Pre chemotherapy	22.4±19.1	0.423
	Post chemotherapy	29.1±65.6	
IV	Pre chemotherapy	74.3±83.0	0.197
	Post chemotherapy	91.0±109.0	

- The table shows the mean ± SD and probability (P).
- t- Test was used for comparison.
- P < 0.05 considered significant.

Table 3 show the comparison between serum levels of tested markers and exposed to surgery. There was a significant increase in CEA and CA 19-9 in post chemotherapy compared to pre chemotherapy in stage I, II& III, but there was no difference in CEA and CA 19-9 in stage IV for patients with CRC who were exposed to surgery before chemotherapy.

Table 4 show the comparison between serum levels of tested markers and patients who were not exposed to surgery. There was a significant increase in CEA in post chemotherapy compared to pre chemotherapy in stages I & II only. There were no differences in stage III & IV for CEA and also for CA 19-9 in all stages

Table (3): show that the relation between levels of serum CEA and CA 19-9 in four stage group among all patients of CRC who exposed to surgery (adjuvant therapy).

Stage		CEA (ng/ml)		CA 19-9 (U/ml)	
		Mean ± SD	P value	Mean ± SD	P value
I	Pre chemotherapy Post chemotherapy	14.2 ± 4.6 9.3 ± 2.9	0.012	30.7 ± 17.8 27.0 ± 19.7	0.042
II	Pre chemotherapy Post chemotherapy	19.4 ± 10.8 11.4 ± 9.7	0.000	34.1 ± 49 23.4 ± 29.2	0.027
III	Pre chemotherapy Post chemotherapy	22.1 ± 14.9 8.3 ± 3.7	0.017	15.9 ± 6.6 8.14 ± 10	0.017
IV	Pre chemotherapy Post chemotherapy	43.8 ± 35.2 58.2 ± 84.5	0.494	50.7 ± 33.4 54 ± 46.7	0.667

- The table shows the mean ± SD and probability (P).
- t- Test was used for comparison.
- P < 0.05 considered significant.

Table (4): show that the relation between serum level of CEA and CA 19-9 in four stage group among all patients of CRC who did not exposed to surgery.

Stage		CEA (ng/ml)		CA 19-9 (U/ml)	
		Mean ± SD	P value	Mean ± SD	P value
I	Pre chemotherapy Post chemotherapy	25.5± 6.7 12.0±10.3	0.047	35.1±24.9 28.3±20.4	0.190
II	Pre chemotherapy Post chemotherapy	36.3±45.1 16.8±17.5	0.046	15.0±7.4 13.1±8.8	0.162
III	Pre chemotherapy Post chemotherapy	28.3±18.6 22.6±32.3	0.312	25.5±22.3 37.1±78	0.348
IV	Pre chemotherapy Post chemotherapy	195.2±134.9 227.0±192	0.455	108±120.3 144.0±151	0.245

- The table shows the mean ± SD and probability (P).
- t- Test was used for comparison.
- $P < 0.05$ considered significant.

Discussion

Colorectal cancer (CRC) represents the second highest cancer mortality rate with an estimated 447,000 new cases diagnosed in 2012 and approximately 215,000 deaths in Europe (Ferlay et al. 2013) Surgical resection remains the primary method of treatment for localized disease with a curative rate of approximately 50% (Yukawa et al. 2001)

Cancer embryonic antigen (CEA) and carbohydrate antigen (CA19-9) are well-known tumor markers that are used in the diagnosis of colorectal cancer. They are also used in preoperative staging and postoperative follow-up of patients, especially patients who are treated with chemotherapy (Reiter et al. 2000).

This study was carried out in 100 colorectal cancer patients being admitted to National Cancer Institute (NCI-Sudan) to receive chemotherapy from June 2016 to February 2017 in Al Jazeera State. And the study aimed to monitor the response of chemotherapy of colorectal carcinoma by measurements of carcino-embryonic antigen (CEA) and carbohydrate antigen 19-9 (CA 19-9) pre and post chemotherapy in which all patients received chemotherapy with a 5-fluorouracil (5-FU).

In the present study serum level of CEA was found to be significant in stage I, II, and III (p=0.009, p=0.001p=0.048 p=0.016 respectively) but only stage IV was insignificant (p=0.288) the CRC patients in the post chemotherapy compared to the pre chemotherapy.

Our results showed a significant results for CA 19-9 in stage I and II (p=0.045, p=0.017) with insignificant results in stage III and IV (p=0.045 and p=0.017) of the CRC patients in the post chemotherapy compared to the pre chemotherapy.

These results agree with Gina Brown 2015 who reported that the systemic treatment of patients with colorectal cancer was regarded as palliative when their disease was found to be metastatic, or to be locally advanced and inoperable. In this setting, the aim of treatment was to achieve control of disease in order to prolong survival, with a particular emphasis on treating or preventing cancer-related symptoms and on maintaining quality of life (Gina Brown. 2015). Despite advances in systemic therapy, treatment is not on its own likely to cure patients. Another study done by U. Ward. et al 1993, showed the partial response (27%) of tumor markers (CEA, CA-195 and CA-242) in CRC patients undergoing chemotherapy for advanced colorectal cancer.

In the current study, the serum levels of CEA and CA 19-9 significantly increased in stages I, II, III (p=0.012, p=0.000, p=0.017 for CEA respectively and (p=0.042, p=0.027, p=0.017) for CA 19-9 respectively, but there was no significant difference in stage IV (p=0.494 for CEA and p=0.667 for CA 19-9 respectively) of

CRC patients undergo surgery before the start chemotherapy (Adjuvant therapy).

These results agree with other researchers who found the levels of these marker in adjuvant therapy to be significant in early stage of CRC ($p<0.01$) and recurrence in the advance stage is 80% after the curative resections for CRC (Takao Ohtsuka. Et al 2008).

Results from the present study indicated that CEA is better than CA 19-9 in flow of patients with CRC. This finding was in agreement with Clemens Giessen-Jung et al 2015 who claimed that CEA remains the only recommended biomarker for follow-up care of colorectal cancer (CRC). Locker et al. 2006 and Duffy et al. 2007 reported that circulating CEA is only recommended to monitor therapy in advanced CRC and for prognostic information. Another study done by Yu et al. 2013 reported that CEA is the most commonly used tumor marker in surveillance and monitoring of CRC disease.

In conclusion, the measurement of tumor markers might be useful in the monitoring of response, and in the prediction of prognosis in patients treated with chemotherapy especially in early stages of colorectal cancer. Also the study concluded that CEA was a tumor marker of choice for CRC management. Further studies are required to confirm these findings.

Acknowledgement

Special thanks to all the staff members of National Cancer Institute (Al Jazeera University) for their support, guidance and encouragement in making this research possible.

References

1. Carpelan-Holmstro¨m M, Haglund C, Lundin J, Alfthan H, Stenman UH, Roberts PJ. (1996). Independent prognostic value of preoperative serum markers CA 242, specific tissue polypeptide antigen and human chorionic gonadotrophin beta, but not of carcinoembryonic antigen or tissue polypeptide antigen in colorectal cancer. Br J Cancer 74(6):925–9.

2. Du J, Li Y and Li J, (2010). Zheng J. Polycomb group protein Bmi1 expression in colon cancers predicts the survival. Med Oncol 27(4):1273–6.

3. Duffy MJ, Dalen van A, Haglund C, Hanson L, Klapdor R, Lamez R, Sturgeon C, Topolcan, (2003). Clinical utility of biochemical markers in colorectal cancer. European Group on Tumor Markers (EGTM) guidelines. Eur J Cancer 39:718-27.

4. Duffy MJ, van Dalen A, Haglund C, Hanson L, Holinski-Feder E, Llapdor R, Lamezer R, Peltomaki P, Sturgeonc, Topolcan O, (2007). Tumor markers in colorectal cancer: European Group on Tumour Markers (EGTM) guidelines for clinical use. Eur J Cancer 43:1348–60.

5. Filella X, Molina R, Grau JJ, JM Pique, JC Garcia-Valdecases, E Astudillo, A Biete, JM Bordas, A Novell, E Campo, (1992). Prognostic value of CA 19-9 levels in colorectal cancer. Ann Surg 216:55-9.

6. Ferlay J, Steliarova-Foucher E, Lortet-Tieulent J, Rosso S, Coebergh JW, Comber H, Forman D, Bray F, (2013). Cancer incidence and mortality patterns in Europe: estimates for 40 countries in 2012. Eur J Cancer 49 (6):1374–403.

7. Giessen-Jung, Dorothea Nagel, Maria Glas, Fritz Spelsberg, Ulla Lau-Werner, Dominik Paul Modest, Marlies Michl, Volker Heinemann, Petra Stieber, Christoph Schulz, (2015). Preoperative serum markers for individual patient prognosis in stage I–III colon cancer. Tumor Biol. 36:7897–7906.

8. Gina Brown. (2015). Colorectal cancer. Cambridge Books Online Chapter 6 - Chemotherapy. pp. 97-109.

9. Hanke B, Riedel C, Lampert S, Happich K, Martus P, Parasch, Himmier B, Hohenberger W, Hahn EG, Wein A, (2001). CEA and CA 19-9 measurement as a monitoring parameter in metastatic colorectal cancer (CRC) under palliative fist line chemotherapy with weekly 24-hour infusion of high-dose 5-fluoracil (5-

FU) and folinic acid (FA). Ann Oncol 12:221-6.
10. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. (2011). Global cancer statistics. CA Cancer J Clin. 61(2):69–90.
11. Jolanda Stiksma, Diana C, Grootenorst, Peter Willem G, vander linder, (2014). CA 19-9 as a Marker in Addition to CEA to Monitor Colorectal Cancer. Clinical Colorectal Cancer, Vol. 13, No. 4, 239-44.
12. Knopf K, Warren JI, Feuer E J, Brown ML. (2001). Bowel surveillance pattern after a diagnosis of colorectal cancer in Medicare beneficiaries. American Society for Gastroenterology endoscopy; vol. 54: 5.
13. Levy M, Visokai V, Lipska L, Topolcan O. (2008). Tumor markers in staging and prognosis of colorectal carcinoma. Neoplasma 55(2):138–42.
14. Locker GY, Hamilton S, Harris J, Jessup JM, Kemeny N, Macdonald JS, Somerifield MR, Hayes DF, Bast RC JR, (2006). ASCO 2006 update of recommendations for the use of tumor markers in gastrointestinal cancer. J Clin Oncol 24:5313–27.
15. Osaka A, Hasegawa H, Yamada Y, Yanagihara K, Hayashi T, Mine M, Aoyama M, Sawada T. (2009). A novel role of serum cytochrome c as a tumor marker in patients with operable cancer. J Cancer Res Clin Oncol, 135:371-377.
16. Reiter W, Stieber P, Reuter C, Nagel D, Lau-Werner U, lamerz R. (2000). Multivariate analysis of the prognostic value of CEA and CA19-9 serum levels in colorectal cancer 20(6D): 5195-5198.
17. Siegel R, Desantis C, Virgo K, Marotto A, Smith T, Cooper D, Gansler T, Lerro C, Fedewa S, lin C, Cannady RS, Cho H, Scoppa S, Hachey M, Kirch R, Jemal A, Ward E, (2012). Cancer treatment and survivorship statistics. CA Cancer J Clin 62:220–41.
18. Takao Ohtsuka. Nakafuusa Y, Sato S, Kitajima Y, Tanaka M, Miyazaki K, (2008). Different Roles of Tumor Marker Monitoring after Curative Resections of Gastric and Colorectal Cancers. Dig Dis Sci 53:1537–1543.
19. U. Ward, JN Primrose, PJ Finan, TJ Perren, P Selby, DA Purres, EH Copper, (1993). The use of tumour markers CEA, CA-195 and CA-242 in evaluating the response to chemotherapy in patients with advanced colorectal cancer. Br. J. Cancer 67: 1132-1135.
20. Yan-Rong Wang, Jian-Xia Yan, LI-Na Wang, (2014). The diagnostic value of serum carcino-embryonic antigen, alpha fetoprotein and carbohydrate antigen 19-9 for colorectal cancer Journal of Cancer Research and Therapeutics - Volume 10 - Special Issue 3.
21. Yu H, Son GM, Joh YG. (2013). The clinical significance of preoperative serum levels of carbohydrate antigen 19–9 in colorectal cancer. Journal of Korean Surgical Society 52:231–7.
22. Yukawa N, Yoshikawa T, Akaike M, Sugimasa Y, Takemiya S, Yanoma S, Noguchi T, Takanshi Y, (2001). Plasma concentration of tissue inhibitor of matrix metalloproteinase 1in patients with colorectal carcinoma. Br.J of Surgery 88: 1590-1601.