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Expression of MUC1 & MUC2 in Colorectal Adenocarcinoma

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Abstract

Background: Throughout the world colorectal cancer is a major cause of morbidity and mortality. It is the most common cancer and the fourth most frequent cause of cancer death worldwide. The annual incidence rates (AARs) for colon cancer and rectal cancer in India in men are 4.4 and 4.1 per 100000, respectively. MUC1 is normally expressed on the apical borders of various glandular and luminal epithelial cells. MUC1 represents the best characterized membrane-associated mucin. The expression of MUC1 is up-regulated in colorectal cancer. MUC2 is expressed by intestinal goblet cells. The expression of MUC2 is down-regulated in non-mucinous adenocarcinomas arising within adenomas.

Methods: A descriptive and cross-sectional study was conducted in Department of Pathology, Himalayan Institute of Medical Sciences, Swami Ram Nagar, Dehradun, on 30 surgical specimens and biopsies of colorectal adenocarcinoma after signed informed consent of patients. Immunohistochemical expression of MUC1 and MUC2 were studied.

Results: Maximum numbers of the cases were in fifth decade with male to female ratio of 0.8:1. The most common presenting complaint among the cases of CRC was pain in abdomen (n=26; 86.66%) followed by rectal bleeding (n=18; 60%), constipation (n=11; 36.66%) and weight loss (n=10 ; 33.33%). Maximum number of CRC cases (60%) was in T3N0 TNM stage. Over expression of both MUC1 and MUC2 was observed in 50% of the cases. Over 30% of the cases expressed only MUC1 while 13.3% of the cases expressed only MUC2.

Conclusion: Expression of MUC1 was more than MUC2 in CRC cases. Although data obtained in the present study on prognostic importance of MUC1 and MUC2 is not statistically significant, yet both MUC1 & MUC2 were over expressed in CRC.

Key words: colorectal carcinoma (CRC), MUC1, MUC2.

Introduction

Throughout the world colorectal cancer is a major cause of morbidity and mortality. It accounts for over 9% of all cancer incidences [1]. It is the most common cancer and the fourth most frequent cause of cancer death worldwide. The WHO estimates that 9,45,000 new cases occur yearly, with 4,92,000 deaths. This cancer is more common in developed than developing countries. In developed countries, it is the second most common tumour, with a lifetime incidence of 5%, but its incidence and mortality are now decreasing [2]. The annual incidence rates (AARs) for colon cancer and rectal cancer in India in men are 4.4 and 4.1 per 100000, respectively. The AAR for colon cancer in women is 3.9 per 100000 [3]. Carcinomas of the large bowel may present with

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rectal bleeding, changes in bowel habits (such as diarrhoea alternating with constipation), anemia resulting from chronic blood loss, vague abdominal pain, intestinal obstruction and rarely perforation [4]. The most common location of colorectal cancer is the left side of the colon including the rectum [5]. The survival of colorectal cancer patients depends upon the stage of disease at the time of diagnosis. It ranges from 90% 5-year survival rate for cancers detected at the localized stage; 70% for regional; 10% for people diagnosed for distant metastatic cancer [1]. At present, the adjuvant treatment is based on TNM stage, tumor type and resection margin status. MUC1 represents the best characterized membrane-associated mucin and is the apomucin for epithelial membrane antigen and related structures identified with monoclonal antibodies raised against the membrane component of human milk fat globules. The expression of MUC1 is up-regulated in a variety of cancers including colorectal cancer [6].

Normally MUC1 is expressed on the apical borders of various glandular and luminal epithelial cells in the mammary gland, oesophagus, stomach, duodenum, pancreas, uterus, prostate and lungs, providing protections to the underlying epithelia and playing a role in cell signalling [7]. MUC2 is expressed by intestinal goblet cell. It is expressed in adenomas and mucinous carcinomas. MUC2 down-regulation is seen in non-mucinous adenocarcinomas arising within adenomas [6]. Hospital based and population based data show that the incidence rates for rectal cancer is higher than colon cancer in all parts of India [8].

We therefore aim to study the MUC1 and MUC2 expression pattern and their correlation with histological type, grade, and stage of colorectal carcinoma in a tertiary hospital of sub Himalayan region of India.

Material and Methods

This study was performed retrospectively in Department of Pathology, Himalayan Institute of Medical Sciences, Swami Ram Nagar, Dehradun, on 30 surgical specimens and biopsies of colorectal adenocarcinoma after signed informed consent of patients in past 1 year from January 2016 to December 2016. Immunohistochemical expression of MUC1 and MUC2 were studied and correlation was done with clinicopathological features of colorectal adenocarcinoma. MUC1 & MUC2 staining included steps in accordance with the protocol given by “Biogenex” (literature with the kit). Patient who received neoadjuvant chemotherapy or radiotherapy prior to surgery were excluded. After detailed histological examination of tumor, Grading and staging was done according to the WHO classification and TNM staging of colorectal cancer [9]. Expression of MUC1 and MUC2 was scored semi quantitatively; based upon the percentage of positive cells and intensity of staining which was classified on a scale as follows [10].

CRC was regarded as MUC1 or MUC2 positive when the score was 3 and 4, according to previous reports.

MUC1 and MUC2 Expression	Score
<5% cells	0,1
5-30% cells	2
30-60% cells	3
>60% cells	4

Results

The mean age of presentation was 50 years and male to female ratio was 0.8:1. The most common presenting complaint among the cases of CRC was pain in abdomen (86.66%) followed by rectal bleeding (60%). On endoscopic examination 80% cases of CRC showed presence of exophytic growth at various sites in the colon and rectum while 20% of the cases (n=6) had ulcerated lesion. CRC were more common in left side of colon (60.00%) than right side of colon (26.67%). Rectum was the most common site (n=12; 40.0%), followed by ascending colon (n=5; 16.67%) and sigmoid colon (n=4; 13.34%). About 93.33% of the cases (n=28) were moderately differentiated

Table 1: Profile of MUC1 and MUC2 expression in Colorectal carcinoma (n=30)

Expression of MUC1 and MUC2	No. of Cases (n)	Percentage (%)
MUC1+/MUC2+	15	50.00
MUC1-/MUC2-	2	6.66
MUC1+/MUC2-	9	30.00
MUC1-/MUC2+	4	13.33
Total	30	100

adenocarcinoma while 6.66% of the cases (n=2) were poorly differentiated adenocarcinoma. Maximum number of the cases were in T3 stage (n=18; 60.00%) and 63.33% (n=19) did not show lymph node metastasis. Positive expression of MUC1 was seen in 80% (n=24) while only 63% (n=19) showed positive expression of MUC2. Over expression of both MUC1 and MUC2 was observed in 50% of the cases. 6.7% of the cases did not express any of these. 30% of the cases expressed only MUC1 while 13.3% of the cases expressed only MUC2 (Table 1).

Out of total 30 cases of CRC 50% (n=15) expressed both MUC1 and MUC2, 30% (n=9) expressed MUC1 and 13.3% (n=4) expressed MUC2 only. 6.7% (n=2) did not expressed any of these two. Out of 15 cases which expressed both MUC1 and MUC2 10 (66.66%) were in rectum and sigmoid colon and 3 (20%) were in caecum and ascending colon. Only 2 (13.33%) cases were in transverse colon. Total 9 cases expressed MUC1 only, out of which 3 were in rectum, 2 each in descending colon and ascending colon and 1 case each was present in caecum and splenic flexure. 4 cases expressed MUC2 only, out of which 2 (50.00%) were in rectum and 1 (25%) of each was in caecum and ascending colon. Only 2 cases did not express MUC1 and MUC2. Out of which 1 (50%) was in rectum and other (50%) was in splenic flexure.

Although, rectum was the most common anatomical site for the colorectal carcinoma (Table 2) Yet, there was no statistical significant difference found between MUC1 and MUC2 over expression and other anatomical sites (p=0.3257).

Cases which expressed both MUC1 and MUC2 or only MUC1 or neither of these had maximum no. of cases of T3. Only MUC2 + cases, had maximum no. of cases of T2 (Table 3; p=0.081). Cases which were both MUC1/MUC2 + or only MUC2 + had maximum number of cases of N0 lymph node status. While cases which were only MUC1 + had

Table 2: Correlation of MUC1 and MUC2 expression with anatomical site of colorectal cancer (n=30)

	MUC1+/MUC2+	MUC1-/MUC2-	MUC1+/MUC2-	MUC1-/MUC2+	Total
Caecum (n)	1	0	1	1	3
Ascending colon	2	0	2	1	5
Transverse colon (n)	2	0	0	0	2
Splenic flexure (n)	0	1	1	0	2
Descending colon (n)	0	0	2	0	2
Sigmoid colon (n)	4	0	0	0	4
Rectum	6	1	3	2	12
Total n (%)	15 (50%)	2 (6.7%)	9 (30%)	4 (13.3%)	30 (100%)

Table 3: Correlation of expression of MUC1 and MUC2 with tumor size (n=30)

Expression of MUC1 and MUC2	Tumor Size				Pearson Chi-square
	T2 n(%)	T3 n (%)	T4 n (%)	Total n	
MUC1+/MUC2+	2 (13.3%)	12 (80%)	1 (6.7%)	15	
MUC1-/MUC2-	0 (0.0%)	1 (50%)	1 (50%)	2	
MUC1+/MUC2-	3 (33.3%)	5 (55.6%)	1 (11.1%)	9	
MUC1-/MUC2+	2 (50%)	0 (0.0%)	2 (50%)	4	
Total	7 (23.3%)	18 (60%)	5 (16.7%)	30	p=0.081

maximum number of cases had N1 lymph node stage (Table 4). There was no statistical significant difference found between MUC1 and MUC2 expression and lymph node status of the cases (p=0.126).

Discussion

In the present study, maximum number of the cases (n=10; 33.3%) were in the age group of 41-50 years, followed by 51-60 years (n=7; 23.33%). The youngest patient was 31 years old and the eldest was 73 years old. Chalya PL et al., Khayal AK et al., found that maximum number of cases were in 5th decade similar to our results [11, 12]. In the present study, 53.33% of the cases (n=16) were females and 46.66% of the cases (n=14) were males with male to female ratio of 0.8:1.

These results were concordant with previous study done by Manne U et al., in which 58 cases of CRC were included, out of which 53% were female and 47% were males with male to female ratio of 0.8:1 [13]. These results were discordant with the results of Veruttipong D et al., where males outnumbered females [14].In the present study the commonest presenting complaint of the cases was pain in abdomen (86.66%) followed by rectal bleeding (60%), constipation (36.66%) and weight loss (33.33%). Similar results were found by others [15]. Rectal bleeding was observed as that the most common complaint [16,17]. These results were discordant with the present study where abdominal pain was the most common presenting symptom followed by rectal bleeding.

In the current study on radiological examination 63.33% of the cases showed thickening of the colonic wall similar with Yadav RR et al., [17]. On endoscopic examination 80% of the cases showed presence of exophytic growth at various sites in the colon while 20% cases had ulcerated lesion. Rectum was the commonest site (40.0%), followed by ascending colon (16.67%) and

Table 4: Correlation of MUC1 and MUC2 expression with Lymph node status (n=30)

Expression of MUC1 and MUC2	Lymph node status				Pearson Chi-square
	N0 n (%)	N1 n (%)	N2 n (%)	Total N	
MUC1+/MUC2+	10 (66.7%)	2 (13.3%)	3 (20%)	15	p=0.126
MUC1-/MUC2-	1 (50.0%)	1 (50.0%)	0 (0.0%)	2	
MUC1+/MUC2-	3 (33.3%)	5 (55.6%)	1 (11.1%)	9	
MUC1-/MUC2+	4 (100%)	0 (0.0%)	0 (0.0%)	4	
Total	18 (63.3%)	8 (26.6%)	4 (13.3%)	30	

sigmoid colon (34 %), similar to earlier studies [18, 19]. Previous study by Duncan TJ *et al* observed that colon was the most common site followed by rectum [20]. This study was discordant with present study.

In the present study, 93.33 % of the cases were moderately differentiated adenocarcinoma (MDA) while 6.66 % cases were poorly differentiated adenocarcinoma (PDA). These results were concordant with previous study by Fleming M *et al*. [21]. In the present study maximum number of the cases showed T3 tumor stage of TNM staging. This was in accordance with study by Khayal KA *et al*., [12].While Duncan TJ *et al*., found that maximum number of CRC cases were in T2 tumor stage [20].

In our study maximum number of cases had no lymph node invasion. This is in concordant with study done by Manne U *et al.*, [13].

In present study 80.00% of the cases showed positive expression of MUC1 while 63.00% of the cases showed positive expression for MUC2 in the study population. Some of the earlier studies found maximum number of cases showed positive expression of MUC1, while others found maximum cases with MUC2 positive expression [12,18, 22]. Ajijoka Y *et al.*, found that maximum number of cases were MUC1+ve, MUC2–ve, discordant with our study [10]. Out of cases which expressed both MUC1 and MUC2, maximum numbers of cases (66.7%) were in sigmoid colon and rectum. Similar results found by Duncan TJ et al., [20]. Maximum number of cases which were either MUC1+/ MUC2+ or MUC1+/MUC2-, had T3 tumor stage; similar results found by Jang KT et al., [18]. Maximum number of cases which where both MUC1/MUC2 + or only MUC2 + had no lymph node status. This was concordant with study done by Ajioka Y *et al.*, [10].

Limitation of our study is small sample size due to which the results may be biased for expression of MUC1 and MUC2 in CRC. Therefore we suggest that larger study should be conducted to see significant correlation between expression of MUC1, MUC2 and clinicopathological features.

Conclusions

The study concluded that colorectal adenocarcinoma was slightly more common in females, mostly presenting in fifth decades. Pain in abdomen was the most common symptom followed by rectal bleeding. Hence patients with these symptoms need proper screening and workup for CRC. Radiological investigations and endoscopy plays an important role in early diagnosis of CRC. Left side of the large bowel was more affected in CRC. Over expression of mucin has been implicated in tumorigenesis and progression of CRC. Maximum number of cases expressed both MUC1 and MUC2. Expression of MUC1 was more than MUC2 in CRC cases. Only 6.6% CRC cases did not express any of these.

However, the data obtained in the present study on prognostic importance of MUC1 and MUC2 is not statistically significant as suggested by previous literature, may be due to small sample size. This needs to be studied further in larger study population.

Authors' contributions

All authors have read the manuscript and agree to its publication in World Journal of Pathology.

Conflict of Interest

The authors declare that there is no conflict of interest.

Ethical Consideration

The study was approved by the Ethics Committee of the Institute and written informed consent was obtained from all participants. The copy of consent is available with the authors.

Data Availability

Raw data is available with the authors.

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