ORIGINAL RESEARCH

A Study of Various Histopathological Types of Colorectal Carcinomas

Shashi Kiran K1, V. Hari Shanker2, Triveni Bhopal3, Kandukuri Mahesh Kumar4

ABSTRACT

Introduction: Colorectal cancer is a malignant epithelial tumor of the colon or rectum. Only tumors that have penetrated through muscularis mucosae into sub mucosa are considered malignant at this site. In the developed countries it is the second most common cancer after Lung. Worldwide, the incidence of colorectal cancer is 9.4% and 10.1% in men & women respectively. Objective of the study was to study the various histopathological types of colorectal carcinomas.

Material and Method: The complete details of each specimen like name of the patient, age, sex, and other demographic details were recorded. Gross features of the tumors with respect to the size and site, color, consistency and cut section was recorded. Tumor was sliced to the region of deepest penetration to know the extent of local spread, distance of the tumor from the resected margin was recorded. Depending on the size of tumor, adequate number of blocks was given. At least one block was taken from the region of deepest penetration of tumor. Sections were taken from the proximal and distal resected margins.

Results: The age range was between 25 years to 80 years. Male to female ratio is 1.7:1. The incidence of colorectal carcinomas is more in distal colon & rectum (60%) than proximal colon (40%). Of 30 cases in the present study, there were no cases in Dukes’ A, 17 cases were of Dukes’ B (56.66%) and 13 were of Dukes’ C (43.33%). 10 cases were T2N0Mx, 7 cases were T3N0Mx, 6 were T3N1Mx and 7 cases were of T3N2Mx. 10 cases were of Stage I, 7 cases were stage II, and 13 cases were of stage III.

Conclusion: The median age of incidence of colorectal carcinoma is 52 years with a male predominance, majority of them were of Adenocarcinoma, grade I. Duke’s stage B was prominent, whereas TNM stage was of stage III in majority of cases. Considerable variation was noted in the combined immunoprofile within each stage of Dukes’, TNM, Histological grade, similar to an extent seen even in the literature reviewed.

Keywords: Histopathological types, colorectal carcinoma, immunoprofile

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INTRODUCTION

Colorectal cancer is a malignant epithelial tumour of the colon or rectum. Only tumours that have penetrated through muscularis mucosae into sub mucosa are considered malignant at this site.1 In the developed countries it is the second most common cancer after Lung. Worldwide, the incidence of colorectal cancer is 9.4% and 10.1% in men & women respectively.2 The incidence rate in India is quite low, about 2 to 8 per 100,000,3 with an incidence of 4.3/1,000,000 in males & 3.4/ 1,000,000 in females.4 According to GLOBAL STATISTICS 2008,4 colorectal cancer is the third most commonly diagnosed cancer in males and the second in females, with over 1.2 million new cancer cases and 608,700 deaths estimated to have occurred in 2008. The highest incidence rates are found in Australia and New Zealand, Europe, and North America, whereas the lowest rates are found in Africa and South-Central Asia. Incidence substantially higher in males than in females (M:F = 2.1:1). Incidence of colorectal cancer is highest in developed countries such as the United States and Japan, and lowest in developing countries like Africa and Asia. The Incidence in these regions is slightly higher in men than women, and is highest in African American men.

Bowel cancer is the third most common cancer in the UK (2009), accounting for 13% of all new cases. It is the third most common cancer among men in the UK, accounting for 14% of all new cases of cancer in males. Bowel cancer is the second most common cancer in women in the UK (2009).5 Present study was undertaken to study the various histopathological types of colorectal carcinomas.

MATERIAL AND METHODS

Study settings: Osmania Medical College and General Hospital, Hyderabad
Type of study: Observational study
Study sample: 30 specimens of colorectal carcinoma
Study period: 2 years

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Methods
The complete details of each specimen like name of the patient, age, sex, and other demographic details were recorded. Gross features of the tumours with respect to the size and site, colour, consistency and cut section was recorded. Tumour was sliced to the region of deepest penetration to know the extent of local spread, distance of the tumour from the resected margin was recorded. Depending on the size of tumour, adequate number of blocks was given. At least one block was taken from the region of deepest penetration of tumour. Sections were taken from the proximal and distal resected margins.
To isolate the lymph nodes, a series of thin slices were made through the pericolic fat after palpation. All lymph nodes isolated were subjected for histopathological examination. Sections were processed for one day and later embedded in paraffin which was cut at five micron thickness. Sections were stained with conventional Haematoxylin and Eosin (H&E) stain. Special stains like Periodic Acid Schiff (PAS) were done wherever necessary. Haematoxylin-eosin stained slides of all cases were reviewed and classified using the WHO 2000 classification scheme.

STAINING

Haematoxylin and Eosin (H&E)
Reagents
Harris haematoxylin; Eosin; 1% hydrochloric acid: 0.1ml of reagent grade hydrochloric acid, 99.0ml of tap water; 0.5% ammonium hydroxide in 199.0 ml of tap water.

Procedure
1. Deparaffinise and hydrate the slides to distilled water.
2. Stain in Harris Haematoxylin for 4 minutes.
3. Rinse in tap water for 1 minute.
4. Differentiate in 1% hydrochloric acid solution for 1 minute.
5. Rinse in tap water for 1 minute.
6. Blue in Ammonium hydroxide solution for 1 minute.
7. Rinse in tap water for 1 minute.
8. Dip in 95% alcohol 3 times.
9. Dip in eosin 4 times.
10. Dehydrate in 95% alcohol, absolute alcohol and clear in xylene.
11. Mount with a synthetic resin like D.P.X

Results: Nucleus-blue, Cytoplasm - pink.

PAS STAINING FOR MUCIN
Reagents
Schiff’s reagent; 0.5% periodic acid: 0.5 gm of periodic acid in 100 ml distilled water; Harris haematoxylin; 1% hydrochloric acid: 0.1ml of reagent grade hydrochloric acid, 99.0ml of tap water; 0.5% ammonium hydroxide in 199.0 ml of tap water.

Procedure
1. Deparaffinise and hydrate the slides to distilled water.
2. Oxidize for 5 minutes in 0.5% aqueous periodic acid.
3. Rinse in tap and then in distilled water.
4. Stain in Schiff’s reagent for 15 minutes.
5. Rinse for 2 minutes in each of three changes of freshly made sulphite rinse.
6. Wash 5 to 10 minutes in running tap water.
7. Counter stain with haematoxylin for 1-3 minutes.
8. Differentiate by means of three to five quick dips in 1% acid alcohol wash in tap water and blue in Scott’s tap water substitute.
9. Dehydrate in 95% alcohol, absolute alcohol and clear in xylene.
10. Mount with a synthetic resin like D.P.X

Results: With haematoxylin counter stain nuclei are blue; PAS positive materials are magenta (Purple red).

RESULTS
In the present study, the age range was between 25 years to 80 years. The youngest patient reported was 25 years & the oldest was 80 years. Maximum number of cases is in the range of 51 - 60 yrs.
In the present study male to female ratio is 1.7:1, with a slight
male preponderance. 19 were male accounting for 63.33% and 11 were females accounting for 36.66% (approx. 2:1). In the present study the incidence of colorectal carcinomas is more in distal colon & rectum (60%) than proximal colon (40%). We included Caecum, Ascending Colon, and Proximal 2/3rd of transverse colon in proximal colon & distal 1/3rd of Transverse colon, Descending colon, Sigmoid colon, Recto sigmoid junction in distal colon. Incidence is equal in Caecum, Ascending Colon, Sigmoid Colon, Recto Sigmoid Colon, & Rectum each accounting for 20%.

Clinical presentation
Patients with right sided lesions mainly presented with pain abdomen, mass per abdomen and anaemia. Anaemia in these patients was microcytic hypochromic type. Patients with more distal lesions presented with altered bowel habits. Bleeding per rectum or mass per rectum are main symptoms of patients with rectal lesions.

Morphology
Of the 30 colorectal carcinomas, 27(90%) cases were ulceroproliferative growth, and 1 (3.33%) was papillary & 2 were (6.67%) were ulcerative lesions. (See figures 17, 18, and 19). Sizes of the tumors varied from 2 x 2 cm to 14 x 14 cm. 28 were ulceroproliferative type. Cut surface of these lesions was grey white, with 5 showing areas of hemorrhage and necrosis. Serosa is involved in 6 cases. 6 lesions showed presence of glistening mucin grossly. Lymph nodes were isolated in 22 specimens. Number of lymph node isolated varied from 0 – 20. Isolation of 4 lymph nodes was possible in 14 (46.66%) of the resected specimen. The size varied from 0.5 – 2 cm, Cut section of grossly involved lymph nodes was grey white.

Microscopy
Histologically adenocarcinomas were further divided into 3 groups, based on mucin production and presence of signet ring cells into adenocarcinoma, mucinous adenocarcinoma (if > 50% of the lesion is composed of mucin.) and signet ring cell carcinomas (by the presence of > 50% of tumour cells with prominent intracytoplasmicmucin). Of 30 cases in the present study, 23 were adenocarcinoma, 6 were mucinous adenocarcinomas and 1 was signet ring cell adenocarcinoma. Mucin on H & E stained sections appeared either as extracellular mucin pools or intercytoplasmicmucin pushing the nucleus to periphery. Special stains like PAS demonstrated this.

Grading is based primarily on the proportion of the tumor that is composed of glands & was divided into 4 grades. Well differentiated (≥ 95% gland forming), moderately differentiated (50%-95% gland forming), poorly differentiated (<50% gland forming), & undifferentiated (< 5% gland formation is seen.). Mucinous adenocarcinoma and signet-ring cell carcinoma by convention are considered poorly differentiated. Dukes’ staging was followed for all the 30 resected specimens of adenocarcinoma this includes both mucinous and non mucinous adenocarcinomas. Of 30 cases in the present study, there were no cases in Dukes’ A, 17 cases were of Dukes’ B (56.66%) and 13 were of Dukes’ C (43.33%). In Dukes’ B, 2 cases belong to < 45 years and 15 were above 45 years of age. Among the 13 Cases of Dukes’ C, 6 cases were below 45 years and 7 cases were above 45 years.

TNM STAGING
TNM staging was followed in 30 resected specimens of adenocarcinomas. Staging of the tumor was done mainly on pathological findings.

Local extent of the tumor (T)
Of the 30 resected specimens of colon, 20 showed extension of tumor up to serosa whereas in 10 cases it was confined to muscularispropria.

Nodal metastasis (N)
Regional lymph node metastasis was seen in 13 cases. 6 of these cases showed involvement of 1-3 lymph nodes and 7 showed involvement of ≥ 4 lymph nodes.
Distant metastasis (M)

In all the 30 cases, the distant metastases could not be assessed and were designated as pMx. Of the 30 cases in the present study, 10 cases were T2N0Mx, 7 cases were T3N0Mx, 6 were T3N1Mx and 7 cases were of T3N2Mx. 10 cases were of Stage I, 7 cases were stage II, and 13 cases were of stage III.

DISCUSSION

Colorectal cancer is a malignant epithelial tumour of the colon or rectum. In the developed countries it is the second most common cancer after Lung. Worldwide, the incidence of colorectal cancer is 9.4% and 10.1% men and women respectively. The incidence rate in India is quite low, about 2 to 8 per 100,000. With an incidence of 4.3/1, 00,000 in males, 3.4/1, 00,000 in females.

In colorectal carcinoma, the prediction of prognosis is based mainly on Dukes’ stage at the time of resection. However, Patients, who are apparently at the same surgical stage, often exhibit a different outcome.

In this context, more specific prognostic markers may provide a rational approach to plan adjuvant therapy. Although the role of some cellular oncogenes and tumour suppressor genes in the progression from premalignant to transformed colon lesions has been documented, there are no widely accepted biological parameters that would identify a different clinical aggressiveness.

Our aim was to evaluate the spectrum of histopathological variants of colorectal carcinoma, to assess the relationship between p53 nuclear accumulation and Bcl-2 cytoplasmic expression and to correlate their combined expression with the Dukes’s staging and tumor grade, so as to predict the prognosis of patients in the biopsy specimens.

As mentioned earlier and observed in the review of literature, not all studied used similar parameters. Hence, the present study is compared with different studies having certain similar parameters in comparison.

In the present study, the age range was between 25 to 80 years with mean age of 52 years with male predominance (M: F=2:1). The mean age in the present study is 52 years with a male predominance, majority of them were of Stage II.

The median age of incidence of colorectal carcinoma is 52 years with a male predominance, whereas TNM stage was of stage III in majority of cases.

In the present study the tumor size varied from 2 x 2 cm to 14 x 14 cm with majority being ulceroproliferative type. In the present study, the commonest was adenocarcinoma 23 (76.66%) followed by mucinous adenocarcinomas 6 (20%) and signet ring cell adenocarcinoma 01 (3.33%). Adenocarcinomas are non-mucinous tumors whereas mucinous tumors include both mucinous adenocarcinoma & signet ring cell adenocarcinoma. The present study is similar to that observed by Nicholas FS Watson et al (adenocarcinoma accounting for 85%) & Tollenaar et al (non-mucinous tumors 72.22%).

In the present study we observed more of well differentiated adenocarcinoma constituting for 53.33% (16 cases), followed by moderately differentiated for 6.67% (5 cases) and poorly differentiated for 30% (9 cases) which is in accordance to other studies. Mucinous adenocarcinoma and signet-ring cell carcinoma by convention are considered poorly differentiated (grade 3). Low Grade carcinomas include well and Moderately Differentiated, whereas High Grade includes Poorly and Undifferentiated Adenocarcinomas.

The present study is similar to that observed by Sinicrope et al, AJM Watson et al, Tollenaar et al, Nicholas FS Watson et al, Giatromanolaki et al, Zhao Dan ping et al, Chandrakumarshanmugam et al which had a high incidence of low grade adenocarcinomas than high grade adenocarcinoma.

CONCLUSION

The median age of incidence of colorectal carcinoma is 52 years with a male predominance, majority of them were of Adenocarcinoma, grade 1. Duke’s stage B was prominent, whereas TNM stage was of stage III in majority of cases. Considerable variation was noted in the combined immunoprofile within each stage of Dukes’, TNM, Histological grade, similar to an extent seen even in the literature reviewed.

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