POSITIVE PREDICTIVE VALUE OF E-CADHERIN EXPRESSION IN PREDICTING LYMPH NODE METASTASIS IN COLORECTAL CANCER

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ABSTRACT

Background: Epithelial structure integrity is dependent on cell-cell adhesion property. Cancer cells ability to metastasize is greatly linked with breaks in cell-cell adhesion capacity. Cadherins, catenins and CD44 family are cell-cell adhesion molecules. Epithelial integrity is highly dependent on E cadherin adhesion molecule.

Objective: To determine the positive predictive value of E-cadherin expression in predicting metastasis in lymph nodes in colorectal cancer.

Methodology: In this cross sectional study, we reviewed sixty six resection specimens of well, moderately and poorly differentiated adenocarcinoma of colon and rectum of nodal stage N1, N2 and N3 from Shaukat Khanum Memorial Hospital and Research Center. Study duration: March to December 2010. Poorly preserved and previously treated (surgery, chemotherapy and radiotherapy) samples were excluded. E-cadherin expression was examined immunohistochemically. Expression of E-cadherin in more than 50% of tumor cells was considered as positive staining, while expression in 0-50% of tumor cells was considered as negative staining. Those cases which showed no expression of E-cadherin and positive for metastasis in lymph nodes were considered as true positive while cases which showed no expression of E-cadherin and no metastasis in lymph nodes as false positive. Positive predictive value was determined from true positive and false positive results.

Results: Out of 66 cases in this study, expression of E-cadherin was noted to be positive in 40 cases (60.60%) and negative in 26 cases (39.39%). Out of these 40 cases, 24 cases were negative for metastasis in lymph nodes (true negative) and 16 cases were positive for lymph node metastasis (false positive). Out of 26 negative cases, 20 cases showed lymph node metastasis (true positive) and 6 cases were lymph node negative (false negative). Positive predictive value determined from above data was 76.92%.

Conclusion: These results show that colorectal carcinoma with reduced expression of E-cadherin is greatly associated with metastasis in lymph nodes.

Keywords: Positive Predictive Value, E cadherin expression, Lymph node metastasis, Colorectal cancer

INTRODUCTION

Colorectal cancer (CRC) has epithelial origin and it involves large intestine and rectum.[1] After breast and lung carcinoma, CRC is the 3rd most common cancer. CRC occupies 2nd main reason of cancer related mortality in developed western counries.[2] Large bowel cancer is a cancer of old age and occurs in people of 60 years or above in 86% of cases.[3] The incidence of CRC in Pakistan is similar to other Asian countries but much lower than in the developed countries.[4] Various factors influence CRC prognosis.[5] The prognosis of CRC is influenced by tumor stage (TNM, tumor/nodes/metastases). Tumor stage is dependent on depth of invasion, lymph node involvement and other organ metastasis.[1] With the loss of epithelial integrity, tumor cells permeate into neigh boring stroma. This epithelial integrity is reliant on cell-cell adhesion molecules which are linked to maintenance of intact intercellular junctions. Cadherins, catenins and CD44 family are examples of adhesion molecules, which are important in epithelial structures integrity.[6] Metastatic potential of tumor is directly related with loss of cell adhesion molecules. Decreased tumor differentiation is associated with decreased expression of E cadherin and is also linked with increased metastatic potential.[7] Literature showed that cases positive for E-cadherin loss, lymph node negative cases were 35.3% and lymph node positive cases were 64.7%.[8] Cases which showed > 50% staining of E cadherin in tumor cells had 57.9% negative lymph nodes and 42.1% positive lymph nodes for metastatic tumor.[9] Therefore, positivity of lymph nodes is greatly associated with E-cadherin down regulation.[6] The rationale of this study was to recognize immunohistochemical markers demonstrated by primary tumor which could improve Nodal stage assessment of colon cancer patients. It could be helpful especially in those cases where number of lymph nodes used to be less in number in gross specimens. It has been noted that there is a difference in treatment protocols of stage I/II from stage III CRC. Currently adjuvant

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chemotherapy is given in stage III colorectal tumors and is not given in stage II disease.[10-12] Hence, treatment strategies can be decided in colon cancer biopsies specimens with the help of assessment of E cadherin expression. It is also time and cost effective for surgeons. In this study, we compared protein expression and loss by utilizing antibodies to E-cadherin in tumor cells and took histopathology as a gold standard for this study.

METHODOLOGY

This was a descriptive cross-sectional study carried out at Pathology department of Shaukat Khanum Memorial Cancer Hospital and Research Center (SKMCH&RC). We searched out institutional database for resection specimens from 10th March to 10th December 2010.

Inclusion criteria: Only resection specimens of well, moderately and poorly differentiated adenocarcinoma of colon and rectum showing loss of E-cadherin of nodal stage N0, N1, and N2were included. Cases diagnosed during study period were included.

Exclusion criteria: Poorly preserved and previously treated (surgery, chemotherapy and radiotherapy) samples were excluded.

Data collection procedure and analysis: Cases of CRC (adenocarcinoma) found suitable by inclusion criteria were included in this study. The data regarding age and sex was collected. The 10% buffered formalin was used to fix surgical specimens, grossed and morphology was evaluated after hematoxylin and eosin staining. E-cadherin IHC stain was done on the tumor sections in the same batch according to specification. Appropriate positive and negative internal controls were considered. A consultant pathologist saw histopathology slides and assessed the IHC stain E-cadherin expression and lymph node status by microscope analysis. This information was recorded in a pre-designed performa. The collected data was entered into the SPSS version 19 and interpretation was done. Standard deviation and mean was calculated for quantitative variables like age. Qualitative variables were E cadherin expression and lymph node metastasis, and percentage was calculated for qualitative variables. Positive predictive value (PPV) of E-cadherin expression was calculated and presented as frequency and percentage.

RESULTS

We reviewed sixty six resection specimens of well, moderately and poorly differentiated adenocarcinoma of colon and rectum of nodal stage N1, N2 and N3.

In this study, mean age of participants was 48.92± 17 years (range 20 – 90) and 22 (33.33%) patients were of age range 20 – 40 years, 29 (43.93%) patients were of age range of 41 – 60 years and 15 (22.72%) patients were of age range of 61 – 90 years. (Table 1)

In this study, 40 (60.6%) patients were male and 26 (39.4%) patients were female.

Distribution According To E-Cadherin Expression: Out of 66 patients, E-cadherin expression was found negative in 26 (39.39%) cases and positive in 40 (60.60%) cases.(Table I)

Table I: Comparison of E cadherin expression and Lymph node status(n=66)

E-Cdherin Expression	Lymph node metastasis		Total
	Positive	Negative	
Positive	16 (FN	24 (TN)	40
Negative	20 (TP)	6 (FP)	26
Total	36	30	66

Key: TP = True positive, **FP** = False positive, **FN** = False negative
N = True negative

Distribution According To Lymph node status: Out of 66 patients, lymph nodes were positive in 36(54.54%) cases and negative in 30(45.45%) cases. (Table 1)

Comparison of lymph node status with expression of E-cadherin: Out of 66 patients included in study, expression of E cadherin was negative in 26 patients. Of these, 20 cases showed metastasis in lymph nodes so were labeled as true positive, while rest of the six cases were labelled as false positive. E-cadherin expression was positive in total 40 patients. Out of these 24 cases were negative for lymph nodes metastasis (True negative) and 16 were found to be positive for lymph nodes metastasis (false negative). (Table I)

Positive predictive value (PPV) of E Cadherin in predicting lymph nodes metastasis: Positive predictive value (PPV) of E Cadherin expression in determining lymph node metastasis was 76.92%.

DISCUSSION

Preoperative and pathological staging is important in making decision regarding management of patients. Out of this Nodal staging is important. Hence, studies are being conducted on nodal staging because of its role in prognosis of colorectal carcinoma. Nodal staging also helps in selecting patients eligible for postoperative chemotherapy. Clinical studies require accurate nodal staging in treatment strategies comparative analysis. There are various factors which affect accuracy of nodal staging and cause variations in accuracy of nodal staging.[13] These factors are amount of mesentry fixation, correct sampling of number of lymph nodes and examination of levels of H&E stained slides.[9] Mesenteric nodes used to be small size usually, hence chances of missing the micrometastases are increased in mesenteric nodes.[10,11,12] Recent studies[9,10,13] revealed that reliable nodal staging requires at least 10 to 15 lymph nodes, but this number may be substantially higher to attain greater predictive value.[14,15]

We performed this study to determine the positive predictive value of E cadherin expression loss in determining lymph node metastasis. It was found that the positive predictive value of E cadherin expression was 76.92%.

In literature, there are other clinical trials which have described the direct relationship of reduced expression of E cadherin expression to metastasis in lymph nodes.[14-17]

Our study results shown that in patients showing loss of E-cadherin, 23.07% patients are negative for metastases in lymph nodes while 76.92% cases are positive for metastases in lymph nodes. On other side patients showing E cadherin positivity 40% cases found lymph node positive and 60% were lymph node negative.

A study was conducted by Elzaghrid,[6] showing the comparable results that patients showing loss of E-cadherin, 35.3%were negative for metastases in lymph nodes cases and 64.7% cases were positive for metastases in lymph nodes. While patients showing > 50% staining of E cadherin in tumor were negative for metastases in lymph nodes in 57.9% cases and positive for metastases in lymph nodes in 42.1% cases, with positive predictive value of 64.7%.

In other studies, a decreased expression of E-cadherin in primary tumor showed direct relationship with the frequency of metastases in lymph node and liver.[16-19] In one study, carcinomas were split into three groups and integrated aberrant expression of E-cadherin, α-catenin and β catenin were studied.[17] There results showed that degree of loss of adhesion molecules expression is related with increased chances of metastases in lymph nodes. In addition to that another strange finding was noted when levels of expression of adhesion molecules in primary tumor with the levels in metastatic tumors were compared. There was increased expression of adhesion molecule. It was found 43% in metastatic lymph nodes and 46% in metastatic liver tumors.A study by Takayama et al,[18] noted that decreased expression of E cadherin, α-catenin and β catenin was associated with metastases in CRC.

CONCLUSION

Our study highlights the significance of expression of E cadherin in colorectal tumors. Study suggests that E cadherin expression down regulation is valuable in predicting lymph node status.

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