Original Article

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Expression of Cathepsin D in Pancreatic Carcinoma

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Abstract

Background: Carcinoma of the Pancreas is one of the leading causes of mortality due to malignancy. Most of the cases present with advanced stage because of subtle clinical manifestation. With a curative resection, the five year survival rate reaches to 25-30%. *Aims and Objective:* Aim of this study is to correlate Cathepsin D expression with pancreatic adenocarcinoma grade, perineural and lymphovascular invasion. Materials and method: Total of 50 Whipple pancreato duodenectomy specimens were selected for this study. Sections from pancreatic carcinoma were graded according to their degree of differentiation. Perineural invasion and lymphovascular invasion was noted. Immunohistochemical study using Cathepsin D antibody was performed in 50 cases and degree of antibody expression was scored in each case. *Results:* Tumor grading based on Histopathological examination and Cathepsin D expression was done. 12% cases were well differentiated, 42% were moderately differentiated and 46% cases were poorly differentiated carcinoma. Cathepsin D expression was scored. Score 1 was given to 8% cases, 2 to 44% cases and 3 to 48% cases. Lymph node metastases were present in 70% of cases and absent in 30% of cases. Perineural invasion was present in 56% of cases and absent in 44% of cases. Conclusion: Our study shows that increased expression of Cathepsin D was associated with higher grade of pancreatic adenocarcinoma and poor survival rate.

Keywords: Pancreatic Carcinoma; Cathepsin D; Perineural Invasion.

Introduction

Pancreatic ductal adenocarcinoma is the fourth most common cause of cancer-related deaths in the developed world [1]. The disease usually remains undiagnosed until the late stage, has a dismal prognosis and a 5-year survival rate of <5% [2,3]. Tumor recurrence is one of the main causes of poor survival after curative resection of pancreatic adenocarcinoma. For early stage disease following resection, many patients develop local recurrence and distant metastasis [4,5]. Proteases are secreted and activated at invadopodia sites that corresponds to areas of extracellular matrix degradation [6]. Several classes of proteases are involved in the proteolytic events which occurs during extracellular matrix degradation and tumour invasion [7,8], including serine proteases such as urokinase or tissue-type plasminogen activators plasmin, and elastase [9], cysteine proteases such as cathepsins B and L [10], aspartyl proteases such as cathepsin D [11], and the matrix metalloproteinases [12]. Cathepsin B is a lysosomal protease that has been shown to promote tumor invasion and distant metastasis of pancreatic carcinoma [13]. Upregulation of cathepsin D expression is seen in several cancer types, including pancreatic carcinoma and plays a crucial role in extracellular matrix proteolytic degradation , which can be completely inhibited following treatment with the inhibitor pepstatin [14-17]. In recent years, the expression of various factors like MicroVessel Density, Vascular Endothelial Growth Factor and Cathepsin were assessed using biological markers in terms of prognostic evaluation in pancreatic adenocarcinoma. Studies have shown that after curative resection

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Cathepsin is essential predictor for tumor recurrence [18]. In case of solid tumors, local tumor invasion is promoted by lysosomal proteases- Cathepsin.

Materials & Methods

The study was carried out in the Department of Pathology, after obtaining the approval from Institutional Human Ethical Committee of Govt. Stanley Medical College, Chennai, from February 2010 to December 2012. A total of 50 Whipple pancreato duodenectomy specimens were selected for this study.

All specimens of pancreatectomy and Whipple pancreatico duodenectomy were processed routinely and stained with Hematoxylin and Eosin. Sections from pancreatic carcinoma were graded according to their degree of differentiation into well,moderate and poorly differentiated. Perineural invasion and lymphovascular invasion was noted. Immunohistochemical study using Cathepsin D antibody was done in 50 cases and degree of antibody expression was scored in each case. Each slide was evaluated with cathepsin D immunostaining for percentage of positive cells staining. If immunopositivity was noticed in cytoplasm or membrane of > 10% epithelial cells, slides were considered as positive.

Scoring of cathepsin D expression was based on percentage of positive cells staining - 0 - no staining , 1- If the tumor tissue consists of less than 10% immunoreactive cells, 2 - 10–50% immunoreactive cells, 3- more than 50% immunoreactive cells .

Results

Age group ranges from 30-70 years, with mean age of 53 years. Pancreatic carcinoma peaks in the age group of 50-60 years. Out of 50 cases of pancreatic adenocarcinoma , males were 68% and females were 32 % with a M : F of 2.1:1. 12% cases were well differentiated, 42% were moderately differentiated and 46% cases were poorly differentiated carcinoma (Table 1 , Figure 1). In our study, most of the cases were moderately to poorly differentiated. 82% were located in head of the pancreas and 18% in body and tail of pancreas. Lymph node metastases were present in 70% of cases (Table 2). Perineural invasion was present in 56% of cases (Table 3). Scoring of Cathepsin D expression was done. score 1 was given for 8% cases, 2 for 44% cases and 3 for 48% cases (Figure 1). Our study shows that increased expression of Cathepsin D was associated with higher grading of pancreatic adenocarcinoma.

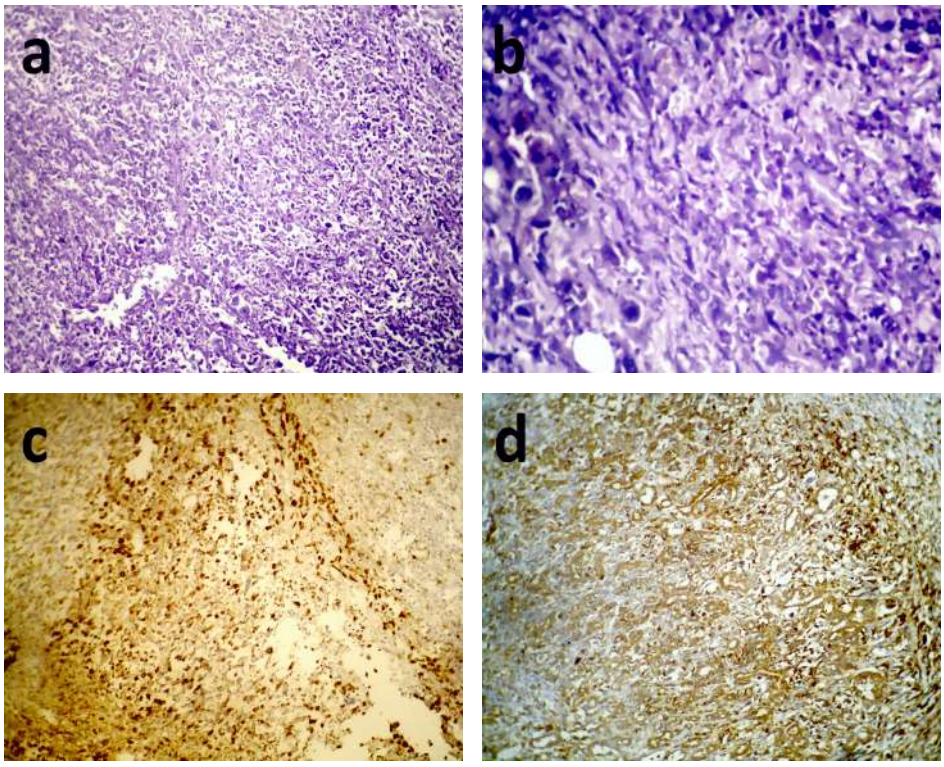

Fig. 1: 10 x view –Poorly differentiated adenocarcinoma. 10 X view- Perineural invasion. Cathepsin D Expression –Score 2. Cathepsin D Expression –Score3

Table 1: Tumor grading based on Histopathology and cathepsin D expression.

Grading	No. of cases in HPE	No. of Cases Expressing Cathepsin D
1	6	4
2	21	22
3	23	24

Table 2: Lymph node metastasis

Lymph node Mets	No. of cases	% of cases
Present	35	70
absent	15	30

Table 3: Perineural invasion

Perineural invasion	No. of cases	% of Cases
Present	28	56
Absent	22	44

Discussion

Up-regulated Cathepsin D expression has been associated with poor prognosis in many malignancies including breast [18,19], prostate [20], and cervical cancer.[21] In pancreatic cancer there are many studies suggesting association of Cathepsin expression with local invasion by extracellular matrix degradation and metastasis, resulting in poor survival rate. In our study, males are affected more than females with a M : F of 2.1:1. 82% of tumors were located in head of the pancreas and 18% in body and tail of pancreas. In our study, most of the cases were (42%) moderately to (46%) poorly differentiated. Our study shows that increased expression of cathepsin D was associated with higher grading of pancreatic adenocarcinoma. This correlates with the study by Niedergethmann et al., where they have evaluated prognostic significance of Cathepsin B following resection in 70 patients with pancreatic carcinoma [22]. During the 3-year follow-up period, 58.6% of patients had disease recurrence, with early recurrence within 6 months of resection occurring in 18.6% of patients. 95.7% of tumor cells were immunoreactive for cathepsin B . They observed that, in comparison to absent or weak immunoreactivity, moderate and strong immunoreactivity was associated with early postoperative recurrence, within 6 months of surgery, shorter survival and associated with perineural invasion. In our study, perineural invasion was present in 56% of cases. In a study by Hannah J. Whiteman et al , increased expression of Cathepsin D was noted in the pancreatic cancer with higher grade and perineural invasion [23]. Many studies have shown the association of Cathepsin D with invasion and short survival rate. Castilla et al. reported association between increased expression of cathepsin D in meningiomas and recurrence [24]. Cathepsin D expression evaluation may be useful in identifying more aggressive forms of CNS neoplasms [25-27]. Among 64 patients of laryngeal squamous cell carcinoma, Lazaris et al. demonstrated those with lymph node metastasis expressing Cathepsin D, were at higher risk of relapse [28]. Kawasaki et al. observed in 78 patients with oral squamous cell carcinoma, association between the Cathepsin D expression and clinical stage, nodal status, pattern of invasion, nuclear proliferating antigen and shorter survival time [29]. Saku et al observed strong and diffuse staining for Cathepsin D was seen in poorly differentiated adenocarcinomas and in signet ring cell carcinoma [30].

In our study, lymph node metastasis and perineural invasion was seen especially in cases of moderately to poorly differentiated adenocarcinoma when compared with well differentiated adeno carcinoma. This finding correlates with the study done by Niedegethmann et al, where they had observed that perineural and lymphatic invasion was associated with higher grading of pancreatic adenocarcinoma. Our study shows that increased expression of cathepsin D was associated with higher grading of pancreatic adenocarcinoma. In a study by Ohta and Terada et al, Cathepsin expression was noted increased with higher grade of pancreatic adenocarcinoma [31].

Conclusion

In patients with pancreatic adenocarcinoma, complete surgical resection is the main modality of treatment. These patients have poor survival rate because of early local recurrence and distant metastasis. This study showed that Cathepsin D can be used as a prognostic predictor in pancreatic carcinoma, especially in early stages.

Conflict of Interest: Nil

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