Original Article

Immunohistochemical Expression of E- and N-Cadherin in Nodular Prostatic Hyperplasia and Prostatic Carcinoma

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Abstract

Background: Different theories have been postulated to explain the development of nodular prostatic hyperplasia (NPH). Epithelial to mesenchymal transition (EMT) is a physiologic process in which the epithelial cells lose their polarity and cell-cell adhesion and acquire a mesenchymal phenotype. **Aim:** The aim of the present study is to investigate the potential role of E- and N-cadherin in the induction of EMT in NPH and prostatic carcinoma. **Methods:** This study was carried out on 55 cases of NPH and 20 cases prostatic carcinoma for evaluation of immunohistochemical expression of E and N cadherins. **Results:** Most NPH (54/55 cases, 98.2%) and all cases of prostatic carcinoma showed positive N-cadherin expression in prostatic glands and stroma. High percentage of N-cadherin expression by stromal cells was significantly in favor of prostatic carcinoma compared to NPH. High percentage of N-cadherin expression by epithelial cells of carcinoma group was significantly associated with young age while its high expression by stromal cells was significantly associated with multicentricity. About 96.4% of NPH and 75% of prostatic carcinoma showed positive E-cadherin expression with a significant difference. No significant association between E-cadherin and N-cadherins in both NPH and prostatic carcinoma was identified. **Conclusions:** The prominent expression of N-cadherin in large numbers of NPH and prostate carcinoma cases in the epithelial and stromal components could point to the occurrence of EMT in those diseases. It also opens a new gate for treatment of those patients by targeting N-cadherin molecule. The absence of inverse association between E-cadherin and N-cadherins in NPH and prostatic carcinoma may indicate that cadherin switch is not an essential step for the development of EMT.

Keywords: E-cadherin, epithelial-to-mesenchymal transition, N-cadherin, nodular prostatic hyperplasia, prostatic carcinoma

Introduction

Nodular prostatic hyperplasia (NPH) is a common chronic proliferative disease of the male genital system characterized by excessive growth of prostatic tissue, and its incidence is increasing with age.[1] In Egypt, NPH represents 41.31% of all prostatic lesions.[2]

Prostate cancer (PCa) is a common malignant tumor of the male genital system, and it ranks as the most common noncutaneous cancer in men in the United States.[3] In Egypt, PCa represents 48.01% of all prostatic lesions and 61.63% of all male genital tract malignancies.[2]

Cadherins are calcium-dependent molecules responsible for cell-cell junctions and include more than 80 members.[4] Classic cadherins is a subfamily of cadherins that mediate adherence junction between epithelial cells maintaining the tissue integrity and cellular polarity in addition to their pivotal role during embryogenesis.[4,5] E- and N-cadherins belong to classic cadherins family.[6] E-cadherin is expressed on the cell surface of all epithelial cells where N-cadherin is found in neural tissue, fibroblasts, skeletal, and cardiac muscles together with endothelial cells.[7]

Although NPH is a common disease, its exact pathogenesis remains a mystery and thus, there is no definitive effective treatment for this disease.[8,9] Different theories have been postulated with a great overlap in between them trying to explain NPH including inflammatory mediators effect,[10,11] defects in stem cells,[12] embryonic reawakening with alteration of interaction between prostatic epithelial cells and stroma,[13] hormonal imbalance,[14] and increased transforming

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growth factor-β (TGF-β).[15] Some studies have linked NPH development with the process of epithelial-mesenchymal transition (EMT).[16]

EMT is a physiologic process in which the epithelial cells lose their polarity and cell-cell adhesion and acquire a mesenchymal phenotype increasing their motility, resistance to apoptosis together with excessive extracellular matrix deposition.[17,18] During this process, reprogramming of epithelial cells occurs by losing the epithelial markers as E-cadherin and keratins and acquiring the mesenchymal markers as vimentin, α-smooth muscle actin , and N-cadherin.[17,19] This process has a role in embryogenesis, tissue healing, and fibrosis in addition to cancer metastasis.[19]

The aim of this study is to evaluate the immunohistochemical expression of E- and N-cadherins, members of the classical cadherin family, in NPH and prostatic carcinoma to investigate their potential role in the induction of EMT in these diseases.

Materials and Methods

This retrospective study investigated 75 prostatic specimens from Egyptian patients, retrieved from the Pathology Department, Faculty of Medicine, Menoufia University, during the period from January 2014 to October 2016. They were randomly selected, based on the availability of paraffin-embedded blocks for serial cutting and examination.

The studied cases included

- 55 cases of NPH that were surgically removed as transurethral resection of the prostate (TURP) (44 cases), open prostatectomy (8 cases) and ultrasound-guided core biopsy from prostate (3 cases)
- 20 cases of prostatic adenocarcinoma that were surgically removed as ultrasound-guided core biopsy from prostate (17 cases) and TURP (3 cases).

From each representative paraffin-embedded block, multiple contagious 4-μm-thick sections were cut and mounted on:

- Glass slides for routine hematoxylin and eosin (H and E) staining
- Positively charged slides for immunostaining procedure.

Histopathological examination

Histopathological examination of H and E stained sections was performed, to confirm the diagnosis and to determine the following:

In benign prostatic hyperplasia group

Gland to stroma ratio – The cases were divided into equal gland to stroma ratio (1:1), gland predominance, and stroma predominance.[20] Basal cell hyperplasia, squamous metaplasia, ectatic blood vessels, acute inflammation, chronic inflammation, and infarction were also assessed as present or absent.

In prostatic adenocarcinoma group

- Centricity: Multicentric or unicentric
- The presence or absence of necrosis or perineural invasion.

Gleason's Score

Cases were classified according to the latest modification of Gleason's scoring system.[21] For statistical purposes, cases with Gleason 6 and 7 were lumped together against cases presented with Gleason 8, 9, and 10.

Immunohistochemistry

Multiple paraffin sections 4 μm in thickness from each case were stained by immunohistochemical method (one for N-cadherin and the other for E-cadherin). The method used for immunostaining was the streptavidin-biotin amplified system.

- Two primary antibodies have been used
  - N-cadherin is a mouse antihuman cadherin antibody antibody (Neural cadherin, 13B154, US Biological). It is received as 0.1 concentrated and diluted by phosphate buffer saline (PBS) in a dilution 1:150
  - E-cadherin is a mouse monoclonal antibody (MS-9470-R7, Thermo scientific, USA). It is received as 7.0 ml (ready to use for immunohistochemistry).

In this system, two reagents were utilized, the biotinylated secondary anti-immunoglobulin which is a purified monoclonal anti-mouse immunoglobulin G (Thermo scientific, NOS-3F7-B11 B5) capable of binding to the primary antibody and the streptavidin-biotin enzyme complex.

Immunohistochemical staining was performed using the Universal Dakocytomation Labeled streptavidin–Biotin-2 system, Horseradish Peroxidase (LSAB-2 System, HRP Kit, Catalog No. K0679). All slides were deparaffinized using xylene and then rehydrated in decreasing concentrations of ethanol. Antigen retrieval using microwave heating (20 min; 10 mmol/citrate buffer, pH 6.0) followed by inhibition of endogenous peroxidase activity (hydrogen peroxidase for 15 min) were used. The primary antibodies were applied on the slides and incubated overnight at room temperature in humidity chamber. Finally, the detection of bound antibody was accomplished using a modified labeled avidin-biotin reagent for 20 min then PBS wash. A 0.1% solution of diaminobenzidine was used for 5 min as a chromogen. Slides were counter-stained with Mayer's hematoxylin for 5–10 min. Negative control slides were prepared, by omitting the primary antibodies from the staining procedure. Tissue sections prepared from colon carcinomas and melanoma were used as a positive control for E-cadherin and N-cadherin, respectively.

Interpretation of N-cadherin and E-cadherin expression

The studied cases were designated as positive for N-cadherin when brown staining was seen either in the cell membrane or cytoplasm in any number of cells.[22] N-cadherin expression was assessed in both epithelial and stromal cells, separately. Only membranous brown staining was considered as a positive expression for E-cadherin in any number of cells.[22] The expression was evaluated in epithelial cells only because it was difficult to assess membranous expression in stromal cells.

For both N-cadherin and E-cadherin, the percentage of expression was assessed in epithelial and stromal cells for N-cadherin and

in epithelial cells only for E-cadherin. The median value was then calculated and used as cutoff point for dividing the cases into high (>median) and low (≤ median) expression.

Statistical analysis

Data were collected, tabulated, and statistically analyzed using a personal computer with "Statistical Package for the Social Sciences (SPSS) version 20 program (IBM corporation, Armonk, NY, USA). Chi-square and Fisher's exact tests were used for evaluation of qualitative data, whereas the Mann–Whitney test was used for evaluation of quantitative data. Value of $P < 0.05$ was considered statistically significant.

Results

The data of NPH and prostatic carcinoma are presented Tables 1 and 2, respectively.

Immunohistochemical results of N-cadherin expression

N-cadherin expression in nodular prostatic hyperplasia

- Most of the studied cases (54/55 cases, 98.2%) showed positive N-cadherin expression in prostatic glands and stroma [Figure 1a] with only one case (1.8%) exhibited neither epithelial nor stromal expression [Figure 1b]
- Most of the studied NPH cases showed predominance of N-cadherin cytoplasmic pattern of expression in epithelial cells [Figure 1a and c]; however, membranous pattern was also appreciated [Figure 1d]. The stromal cells showed only cytoplasmic N-cadherin staining
- The percentage of N-cadherin expression in epithelial cells ranged between 10.0% and 95.0%, with a mean ± standard deviation (SD) of 75.93 ± 17.78 and a median of 80. Twenty-three cases (42.6%) showed high expression using 80% as a cutoff point
- The percentage of N-cadherin expression in stroma ranged between 20.0% and 85.0%, with a mean ± SD of 64.26 ± 15 and a median of 70. Sixteen cases (29.6%) showed high expression using 70% as a cutoff point

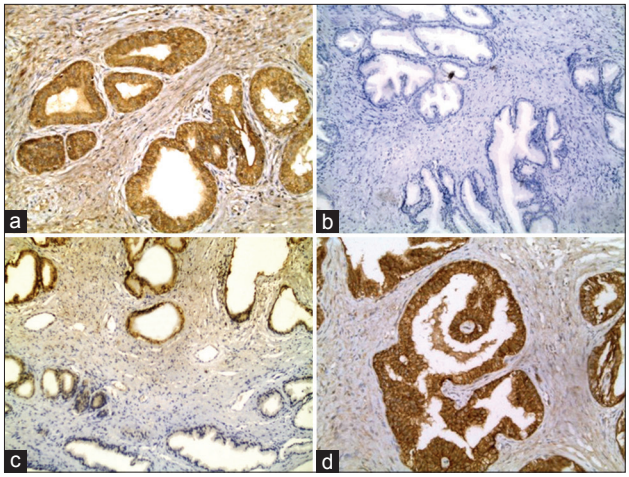

Figure 1: Nodular prostatic hyperplasia with high cytoplasmic expression of N-cadherin in epithelial and stromal cells (a), negative N-cadherin (b), low cytoplasmic N-cadherin (c) and membranous expression in epithelial component (immunohistochemical staining ×200 for [a, b and d], ×100 for [c]) (d)

- An intense cytoplasmic N-cadherin staining pattern was noticed within endothelial cells lining ectatic blood vessels together with proliferated smooth muscle bundles within NPH stroma.

The association between percentage of N-cadherin expression and clinicopathological parameters in nodular prostatic hyperplasia group

- There was no significant association between percentage of N-cadherin epithelial expression and clinicopathological parameters in the NPH group

Table 1: Clinical and histopathological data of nodular prostatic hyperplasia cases

Variables	*n*=55, *n* (%)
Age (years)	
Minimum-maximum	35.0-80.0
Mean±SD	65.76±7.89
Median	65.0
PSA (ng/dl) (10 cases)	
Minimum-maximum	1.5-8.8
Mean±SD	2.93±2.12
Median	2.35
Gland to stroma ratio	
Equal gland to stroma ratio (1:1)	12 (21.8)
Gland predominance	34 (61.8)
Stroma predominance	9 (16.4)
Basal cell hyperplasia	5 (9.1)
Squamous metaplasia	1 (1.8)
Ectatic BVs	43 (78.2)
Acute inflammation	6 (10.9)
Chronic inflammation	10 (18.2)
Infarction	4 (7.3)

SD: Standard deviation, BVs: Blood vessels, PSA: Prostate specific antigen

Table 2: Clinical and histopathological data of prostatic adenocarcinoma cases

Variables	*n*=20, *n* (%)
Age (years)	
Range	59.0-82.0
Mean±SD	6.29±72.65
Median	73.50
Centricity	
Multi centric	12 (60.0)
Unicentric	8 (40.0)
Gleason's score	
6	2 (10.0)
7	8 (40.0)
8	3 (15.0)
9	5 (25.0)
10	2 (10.0)
6+7	10 (50.0)
8+9+10	10 (50.0)
Range	6.0-10.0
Mean±SD	7.85±1.23
Median	7.50

SD: Standard deviation

- However, NPH cases that showed ectatic stromal blood vessels were significantly associated with lower N-cadherin stromal expression ($P = 0.028$) [Figure 2a]
- There was a statistically significant association between percentage of N-cadherin expression by both epithelial cells and stromal cells in NPH group, since cases that showed low epithelial expression exhibited also low stromal expression and vice versa ($P = 0.002$) [Figure 2b].

N-cadherin expression in prostatic adenocarcinoma

All cases of prostatic adenocarcinoma (20 cases) showed positive expression of N-cadherin in both malignant epithelial cells and stroma (100%) [Figure 3]. Similar to NPH cases, cytoplasmic staining pattern takes the upper hand in malignant cancer cells together with adjacent reactive stroma, however, membranous pattern of N-cadherin was still appreciated in some malignant cells.

- The percentage of N-cadherin expression in malignant epithelial cells ranged between 40.05% and 95.0%, with a mean ± SD of 81.75 ± 12.90 and a median of 85. Nine cases (45%) showed high expression using 85% as a cutoff point
- The percentage of N-cadherin expression in stroma ranged between 60.0% and 80.0%, with a mean ± SD of 71.50 ± 6.09 and a median of 75. Eleven cases (55%) showed high expression using 75% as a cutoff point.

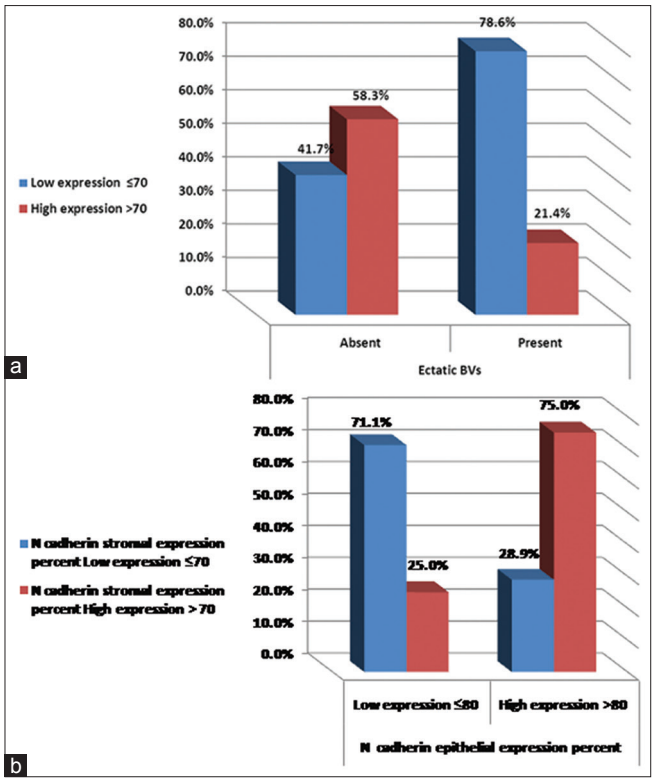

Figure 2: (a) Nodular prostatic hyperplasia that showed ecstatic blood vessels was associated with lower stromal N-cadherin expression. (b) the relationship between the percentage of N-cadherin expression by epithelial and stromal cells in nodular prostatic hyperplasia group

The association between percentage of N-cadherein expression and clinicopathological parameters in prostatic carcinoma group

a. N-cadherin expression in epithelial cells:
   - Prostatic carcinoma cases that showed high N-cadherin expression in epithelial cells were significantly younger (mean age = 73 years) than those exhibiting low N-cadherin (mean age = 76 years) ($P = 0.029$) [Figure 4a].

b. N-cadherin expression in stroma:
   - There was a tendency of prostatic carcinoma cases with multicentric infiltration of the received cores to be associated with higher percentage of N-cadherin by stromal cells (9 cases, 75%) in comparison to unicentric cases that exhibited lower expression (6 cases, 75%), ($P = 0.065$) [Figure 4b].

There was no statistical significance between epithelial cells and stroma in the carcinoma group as regards N-cadherin expression percent.

Comparison between nodular prostatic hyperplasia and carcinoma groups regarding N-cadherin expression

There was no significant difference between NPH and prostatic carcinoma groups regarding N-cadherin expression by epithelial cells. On the other hand, carcinoma cases tended to have higher percentage of stromal N-cadherin expression (median value = 75) in comparison to NPH cases (median value = 70), a relation that showed near significance ($P = 0.063$). High percentage of N-cadherin expression by stromal cells was significantly associated with carcinoma group compared to NPH group ($P = 0.044$) [Table 3].

E cadherin expression in nodular prostatic hyperplasia:

- Fifty-three cases (96.4%) out of 55 NPH cases showed positive expression of E cadherin [Figure 5a and b] whereas only two cases were negative (3.6%) [Figure 5c]

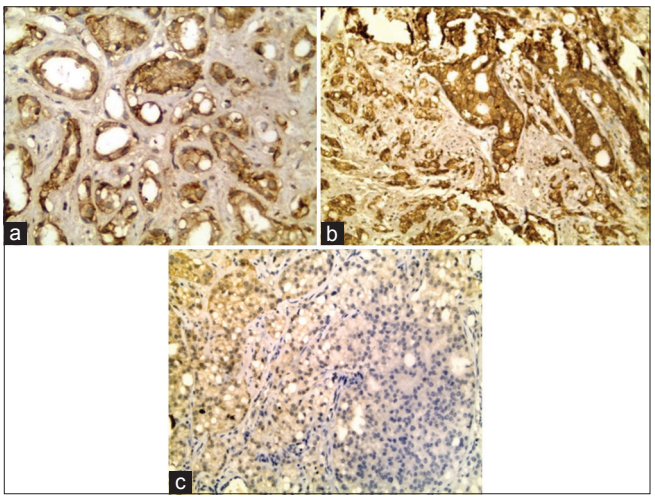

Figure 3: Prostatic adenocarcinoma showed high cytoplasmic and membranous expression of N-cadherin in malignant acini with positive stroma (a and b) and low expression in another case (c). (immunohistochemical staining ×400 for [a] and ×200 for [b and c])

Table 3: Comparison between the two studied groups (nodular prostatic hyperplasia and prostatic carcinoma) as regards N-cadherin expression

N-cadherin	NPH, *n* (%)	Carcinoma, *n* (%)	Test of Significant	*P*
Epithelial cells expression	*n*=55	*n*=20		
Negative	1 (1.8)	0	χ^2=0.369	1.000[FE]
Positive	54 (98.2)	20 (100.0)		
Epithelial cells percent	*n*=54	*n*=20		
Minimum-maximum	10.0-95.0	40.0-95.0	U=423.50	0.147
Mean±SD	75.93±17.78	81.75±12.90		
Median	80.0	85.0		
Low expression	31 (57.4)	11 (55)	χ^2=0.03	0.85
High expression	23 (42.6)	9 (45)		
Stroma expression	*n*=55	*n*=20		
Negative	1 (1.8)	0 (0.0)	χ^2=0.369	1.000[FE]
Positive	54 (98.2)	20 (100.0)		
Stroma percent	*n*=54	*n*=20		
Minimum-maximum	20.0-85.0	60.0-80.0	U=389.50	0.063
Mean±SD	64.26±15.0	71.50±6.09		
Median	70.0	75.0		
Low expression	38 (70.4)	9 (45)	χ^2=4.05	0.044*
High expression	16 (29.6)	11 (55)		

χ^2 for Chi-square test, [FE]*P* for Fisher's exact, U for Mann-Whitney test, *Statistically significant at $P \leq 0.05$. SD: Standard deviation, NPH: Nodular prostatic hyperplasia

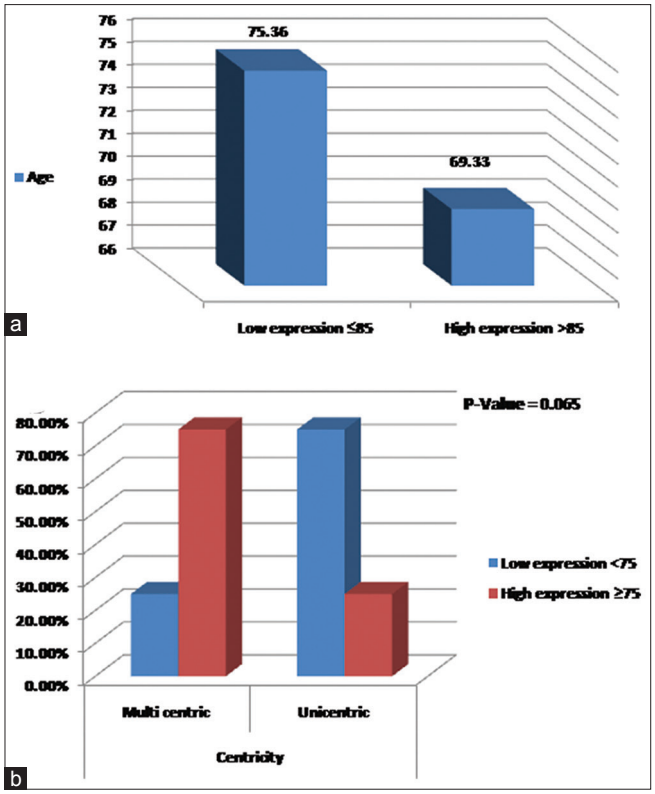

Figure 4: (a) The relationship between N-cadherin expression by epithelial cells and age of prostatic carcinoma group. (b) The relationship between N-cadherin expression by stromal cells and centricity of prostatic carcinoma group

- The percentage of E-cadherin expression ranged between 5.0% and 50.0% with a mean ± SD of 29.81 ± 11.48 and a median of 30. High percentage of E cadherin was identified in 30 cases (56.6%) of NPH using 30% as a cut-off point.

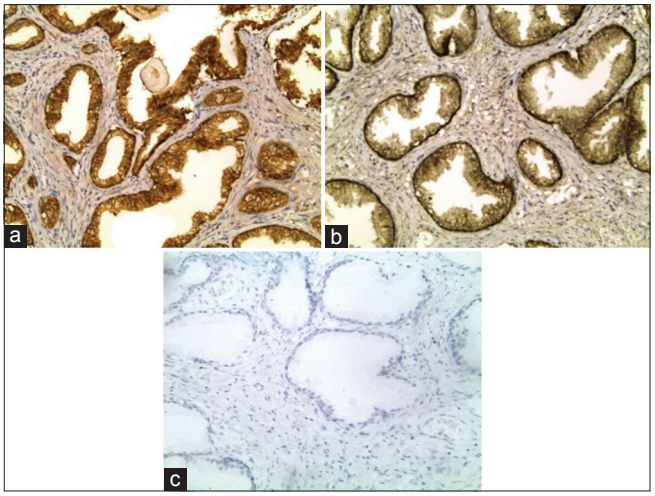

Figure 5: E cadherin membranous expression in nodular prostatic hyperplasia with high expression in (a), low expression in (b) and absent expression in (c) (immunohistochemical staining ×200 for [a and b], ×100 for [c])

The association between percentage of E cadherin expression and clinicopathological parameters in nodular prostatic hyperplasia group

There was no significant association between E-cadherin expression percent and different studied parameters in NPH cases.

E-cadherin expression in prostatic adenocarcinoma:

Fifteen cases of prostatic adenocarcinoma out of 20 showed positive E-cadherin expression (75%) [Figure 6a] and 5 cases

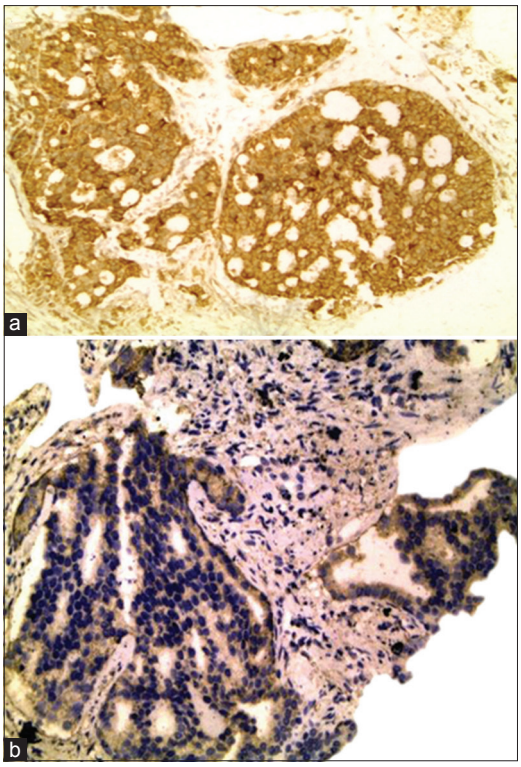

Figure 6: High membranous expression of E cadherin in prostatic carcinoma (a) with negative expression in (b) (IHC, ×200)

were negative [Figure 6b]. The percentage of E cadherin expression ranged between 10.0% and 70.0% with a mean of 32.67 ± 20.95 and a median of 30. Eight prostatic carcinoma cases showed high E cadherin expression using 30% as a cutoff point.

The association between percentage of E cadherin expression and clinicopathological parameters in prostatic carcinoma group

There was an absence of significant association between E-cadherin expression and different studied clinicopathological parameters in prostatic carcinoma group.

Comparison between nodular prostatic hyperplasia and prostatic carcinoma groups as regards E-cadherin expression

NPH cases showed more E-cadherin positivity (96.4%) in comparison to prostatic carcinoma cases (75%) (P = 0.013). However, there was no significant difference between the two groups as regards the percentage of E-cadherin expression [Table 4].

The relationship between N-cadherin and E-cadherin expression in nodular prostatic hyperplasia and prostatic carcinoma groups

There was no significant correlation between E-cadherin expression and N-cadherin expression in both epithelial and stromal cells in the NPH group. Furthermore, there was no significant relationship between N-cadherin and E-cadherin expression percent in both epithelial cells and stromal cells in the carcinoma group.

Discussion

In the current study, 54/55 NPH cases (98.2%) and 20/20 PCa cases (100%) expressed N-cadherin in epithelial and stromal cells. Our results were in agreement with the study of Kolijn *et al*.[23] who reported that N-cadherin was expressed in nearly equal number of NPH and PCa cases causing no statistical difference between the two tested groups but with higher expression percent in carcinoma cases.

It has been reported that endothelial cells lining the blood vessels can change to mesenchymal fibroblast-like cells through a process named as endothelial-to-mesenchymal transition (EndMT).[24] This process has an important role during heart development and also shares in the pathogenesis of several diseases as postischemic cardiac fibrosis.[25] During EndMT, the endothelial cells lose the endothelial markers and acquire mesenchymal ones.[24] The endmt process was also suggested to play a role in the accumulation of mesenchymal cells in the stroma of NPH cases in the study of Alonso-Magdalena *et al*.[26] in which the NPH stroma exhibited thick-walled blood vessels. Similarly, in our study, N-cadherin being a mesenchymal marker was seen to be expressed in the endothelial cell lining of ectatic stromal blood vessels as a feature of EndMT, although this feature predominated in cases exhibited low N-cadherin expression percent (≤70%). Kalluri and Weinberg[19] reported that the proportion of EndMT greatly varies among organs and also depends on the degree of fibrosis.

The idea that NPH is a proliferative stromal disease is not accepted nowadays since stromal cells lacked the expression of proliferative markers as Ki 67. Instead, It has been proposed that myofibroblasts and smooth muscles cells that accumulate in the prostatic stroma in this disease were derived from epithelial cells via the process of EMT.[19] This could explain the observation of intense N-cadherin staining of proliferated muscular stroma in the studied NPH cases in addition to the significant association between N-cadherin expression in prostate epithelial and stromal cells in those cases.

The predictive value of age on the course of PCa is a matter of controversy. Some reported that the onset of PCa in the younger patient was associated with poor survival.[27] Others found that advanced age was associated with higher grade and stage thus having high mortality rates.[28] This means that the age alone has a weak effect on PCa prognosis and its effect is usually influenced by the presence of other adverse histologic factors.[28,29] Adding to that, several studies demonstrated that N-cadherin expression in PCa is considered as an aggressive biomarker of PCa being linked to adverse prognostic factors and poor outcome in this tumor.[22,30] Thus, the appearance of a significant relationship between high N-cadherin epithelial expression percent and patients with lower mean age in the studied PCa cases may be related to the association of those cases with other poor prognostic factors.

Ahn *et al*.[31] demonstrated an association between PCa involving multiple cores and the development of high Gleason's grade and positive surgical margins. Furthermore,

Table 4: Comparison between nodular prostatic hyperplasia and prostatic carcinoma groups as regards E cadherin expression

E cadherin	NPH ($n=55$), *n* (%)	Carcinoma ($n=20$), *n* (%)	Test of Significant	*P*
Expression				
Negative	2 (3.6)	5 (25.0)	$\chi^2=7.910$	0.013*,FE
Positive	53 (96.4)	15 (75.0)		
Percent (%)				
Minimum-maximum	5.0-50.0	10.0-70.0	U=392.0	0.935
Mean±SD	29.81±11.48	32.67±20.95		
Median	30.0	30.0		
Low expression	23 (43.4)	7 (46.7)	$\chi^2=0.051$	0.822
High expression	30 (56.6)	8 (53.3)		

χ^2 for Chi-square test, ^{FE}P for Fisher exact, U for Mann-Whitney test, *Statistically significant at $P\leq0.05$. SD: Standard deviation, NPH: Nodular prostatic hyperplasia

the survival of PCa patients has been decreased with increased the fraction of affected cores.[32] It was found that N-cadherin expression in the stromal cells surrounding the malignant PCa cells is critical for tumor cells invasion and metastasis. This is related to the ability of N-cadherin in mediating homotypic adhesion between PCa cells and stromal fibroblasts since it was expressed in both types of cells.[33] These data could explain the significant relationship between high N-cadherin stromal expression and multicentric cases in our study since they were considered as adverse prognostic factors in PCa.

In addition to their role in prostate development and function, prostate stromal cells were found to share in the pathogenesis of prostatic lesions.[34,35] This is related to the difference in the expression of several transcription factors in tissues of different prostatic pathology.[36] This could explain the statistical difference between the studied NPH and PCa cases regarding N-cadherin expression in stromal cells. Higher percentage of stromal expression predominated in PCa cases owing to the previously mentioned critical role of prostate stromal cells in facilitating tumor cell migration and metastasis.[33]

E-cadherin is constantly expressed in different normal epithelial cells including prostate epithelium.[7] However, in prostatic diseases, decreased E-cadherin expression has been reported. The pronounced immunohistochemical decline of E-cadherin expression has been recorded in PCa tissues and to lesser degree in NPH cases[37,38] which could occur due to disruption of E-cadherin molecule at different cellular levels.[39] This recapitulates the significant decrease in E-cadherin positivity in PCa compared to NPH cases although no statistical difference in expression percent between the studied groups has been appreciated agreeing with others.[40]

TGF-β1 has a pivotal role in the induction of EMT in different physiologic and pathologic conditions.[41] During the process of EMT; TGF-β1 induces disruption of epithelial cell-cell junction (main morphologic changes) in addition to the enhancement of cellular motility.[42] EMT is characterized by up-regulation of N-cadherin and down-regulation of E-cadherin (cadherin switch) which usually occurs in response to TGF-β1.[43] However, it was found that E cadherin down-regulation is not always an essential step for EMT since morphological changes of EMT induced via TGF-β1 could precede E cadherin down-regulation. This could be confirmed by the observation that E cadherin could be still detected on the cell membrane of the morphologically changed cells by immunostaining in the first few days of EMT occurrence. An explanation for this observation is that rapid up-regulation of N-cadherin in response to TGF-β1 activation causing inhibition of E cadherin function before its down-regulation takes place, thus EMT occurs.[42]

Furthermore, morphological changes of EMT can occur in response to TGF-β1 independent of N-cadherin increase, but increased motility of the affected cells of mammary cell lines, which is an integral part of EMT cannot occur without N-cadherin up-regulation.[42] On the same line, several studies observed that N-cadherin expression represents a cornerstone in the process of EMT mediated tumor metastasis rather than E-cadherin.[6,44] This could explain the absence of a significant inverse relationship between E- and N-cadherins in NPH and PCa groups in the current study. However, high median values of N-cadherin (80 in NPH and 85 in PCa) were associated with the low median value of E cadherin (30 in both groups) but with an absence of statistical significance.

In this study, although membranous N-cadherin staining was noticed in epithelial cells of both groups, the cytoplasmic staining pattern predominated and was the only pattern expressed in stromal cells. This was in accordance with the study of Nakajima *et al*.[6] who observed the predominance of cytoplasmic staining pattern of N-cadherin in pancreatic cancer cells. In the same line, epithelioid cells of mesothelioma showed membranous N-cadherin staining where cytoplasmic staining appeared in malignant cells with spindle cell morphology.[45] N-cadherin structure was formed of two domains; extracellular and cytoplasmic ones in addition to the transmembrane part.[46] The extracellular domain is responsible for homotypic cellular adhesion while the cytoplasmic tail binds to catenin and in turn to actin cytoskeleton activating the motility behavior of the cells in addition to augmentation of cellular adhesion.[6]

Thus predominance of N-cadherin cytoplasmic pattern of staining in epithelial cells of NPH cases supports the notion

that EMT plays a role in the development of NPH as the epithelial cells undergo disruption of cellular polarity with activation of their motility so they can break down the cell membrane and accumulate in the adjacent stroma. The link between this pattern of N-cadherin staining together with metastasis and survival in PCa cases needed to be clarified on larger scale study.

Conclusion

The prominent expression of N-cadherin in large numbers of NPH and PCa cases in the epithelial and stromal components could point to the occurrence of EMT in those diseases. The absence of inverse association between E- and N-cadherins in NPH and prostatic carcinoma may indicate that cadherin switch is not an essential step for the development of EMT. It also opens a new gate for treatment of those patients by targeting N-cadherin molecule.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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