Immunohistochemical Study of Stromal CD10 Expression in Mammary Duct Carcinoma

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Abstract

Breast cancer is the most common cancer among women worldwide. The present study was conducted on 50 female cases of mammary in situ and invasive duct carcinoma. All studied cases of invasive duct carcinoma were graded as grade I, II and III according to Elston/Nottingham modification of Bloom-Richardson system. All cases diagnosed as duct carcinoma in situ, with or without focal invasion, as well as invasive duct carcinoma grade II and most of invasive duct carcinoma grade I cases showed weak immunostaining as opposed to invasive duct carcinoma grade III in which 71.4% of its cases showed strong immunostaining. There was a highly significant correlation between CD10 immunostaining intensity and tumor grade. No significant correlation could be achieved between CD10 immunostaining intensity and either patients age, tumor size, lymph node metastasis or hormone profile.

Key Words: CD10 – Immunohistochemical – Duct carcinoma.

Introduction

BREAST cancer is the most common cancer among women worldwide, and accounts for 18% of all female malignancies. Infiltrating duct carcinoma is the most frequent histological type of breast cancer, accounting for approximately 68% of cases [1]. In Egypt, breast cancer constitutes 33% of all female cancers in Egyptian National Cancer Institute (NCI) [2].

Breast cancer is an important cause of morbidity and mortality among women. It represents the second leading cause of cancer deaths in females (after skin cancer). The estimated annual incidence of breast cancer worldwide is about one million cases (22% of all cancers in the world in 2000) [3].

CD 10 is a zinc-dependent peptidase (metalloproteinase), which degrades a variety of bioactive peptides. Earlier studies suggested that CD10 expression in tumor stroma is associated with biological aggressiveness of the tumor. Expression of CD 10 in the stroma of invasive breast carcinoma is associated with ER-negativity, higher tumor grade, decreased patient survival, most significant in the node-negative subset. So CD10 constitutes a clinically important prognostic marker and a potential target for development of novel therapies [4].

The aim of the study is to detect the grade of studied cases of mammary in situ and invasive duct carcinoma according to Elston/Nottingham modification of Bloom-Richardson system, to evaluate immunohistochemically the expression of stromal CD10 in all the studied cases and to correlate the obtained statistically analyzed results with the available previously done hormone profile of all the studied cases.

Material and Methods

The material of this study included 50 paraffin blocks of female cases of mammary in situ and invasive duct carcinoma, obtained either by breast lumpectomy or radical mastectomy. All cases were studied histologically and graded according to Elston/Nottingham modification of Bloom-Richardson system, then studied immunohistochemically using monoclonal antibodies against CD10.

These blocks were obtained from private laboratories within the period from January 2000 to December 2009.

The clinical data were obtained from the pathology reports of the patients. These data included age of the patients, all clinical data, the maximal diameter of the tumor and available previously done hormone profile.
The previously done hormone profile included the immunohistochemical expression of estrogen, progesterone and Her2/neu receptors. Estrogen and progesterone receptor immunostaining were scored as negative, weak, moderate and strong. Her2/neu receptor immunostaining was scored as negative, weak and strong. It is worth mentioning that Her2/neu expression in duct carcinoma in situ cases has no clinicopathological indication and that Her2/neu expression in cases of extensive duct carcinoma in situ with focal invasion was considered for the areas showing stromal invasion only [6].

Each paraffin block was re-cut by rotatory microtome at 5 microns thickness then mounted on glass slides to be stained by hematoxylin and eosin (H&E) for histopathological examination and on charged slides for CD10 immunostaining.

Histopathological Evaluation (Elston and Ellis, 2002):

According to Elston/Nottingham modification of Bloom-Richardson system, all studied cases of invasive duct carcinoma were graded as grade I, II and III. Elston/Nottingham modification of Bloom-Richardson system depends on:

A- Tumor tubule formation:
1 point: >75% of tumor.
2 points: 10-75% of tumor.
3 points: <10% of tumor.

B- Number of mitotic figures in most active area.
This is done by counting 10 high power fields:

\[
\begin{align*}
&\text{(a) } 1 \text{ point: } 0-5 \\
&\text{(b) } 2 \text{ points: } 6-11 \\
&\text{(c) } 3 \text{ points: } 12-22 \\
\end{align*}
\]

C- Nuclear pleomorphism:
1 point: Minimal nuclear variation in size and shape; small regular uniform cells.
2 points: Moderate nuclear variation in size and shape.
3 points: Marked nuclear variation in size and shape.

This should be done by evaluating areas with greatest atypia.

Scoring:
3-5 points: Well differentiated (grade I).
6-7 points: Moderately differentiated (grade II).
8-9 points: Poorly differentiated (grade III).

Immunohistochemical staining:

The blocks were assessed immunohistochemically using standard immunoperoxidase method (monoclonal CD10, Novocastra, dilution 1:50, pretreatment with Decloak 5 minutes in TRS, Novocastra, Newcastle upon Tyne, United Kingdom) was achieved by a labeled streptavidin-biotin peroxidase complex method (Zymed Histostain Plus Kit, Zymed Laboratories, South San Francisco, Calif). The formed complexes were visualized with aminoethyl carbazole chromogen/substrate. CD 10 immunostaining was recorded as negative, weakly positive, and strongly positive.

The negative control was carried out by omitting the primary antibody in each case.

As positive control for cases stained for CD 10, a film obtained from patient with acute lymphoblastic leukemia, which exhibited strong intensity of CD 10 immunostaining, was employed.

Evaluation of CD10 Immunostaining:

Every section was carefully examined at power magnification (x100) for the presence of tumor stromal immunostaining using Olympus microscope CX21.

The CD 10 immunostaining was scored quantitatively as Negative when there is no tumor stromal staining, as weak positive when there is either diffuse weak staining or weak or strong focal staining in less than 30% of tumor stromal cells and Strong positive when there is strong staining in 30% or more of tumor stromal cells [4].

Statistical analysis:

The significance of the results was assessed by determining the probability factor “p” value using the chi-square and one way ANOVA tests.

\[ p<0.05 = \text{Significant.} \]
\[ p<0.01 = \text{Highly significant.} \]
\[ p<0.001 = \text{Very highly significant.} \]
\[ p>0.05 = \text{Not significant.} \]

Results

The present study included 50 female cases of mammary duct carcinoma, 26 (52%) cases were removed by lumpectomy while 24 (48%) cases were removed by radical mastectomy.

The majority of cases studied were invasive duct carcinoma, as they represented 36 cases (72%), while only 2 cases (4%) were duct carcinoma in situ. The rest of cases showed extensive duct carcinoma in situ with focal invasion (24%).
In Invasive duct carcinoma cases, grade II represented the majority of cases studied (52.8%), followed by grade III (30.6%) while grade I was only (16.6%) of cases.

The majority of the studied cases of mammary duct carcinoma were above the age of 40 years (82%), while cases below the age of 40 years represented only 18%. The mean age of all mammary duct carcinoma cases studied was 49.12.

Only some cases of invasive duct carcinoma grade II (37%) and few cases of invasive duct carcinoma grade III (18%) were seen in the age group below 40 years. These results were insignificant.

The least mean age detected in invasive duct carcinoma grade III was (49.7), while the highest mean age seen in invasive duct carcinoma grade II was (67). Therefore, tumor grade was inversely proportionate with the mean age.

All cases of duct carcinoma in situ as well as 58.3% of cases of extensive duct carcinoma in situ with focal invasion and the majority of all grades of invasive mammary duct carcinoma cases showed tumor sizes less than 5cm in maximal diameters. However these results were insignificant. The mean tumor size of all mammary duct carcinoma cases studied was 3.7.

As regard to tumor grade, it showed inverse proportion with mean tumor size. The highest mean tumor size was seen in cases of extensive duct carcinoma in situ with focal invasion, while the lowest mean tumor size was seen in invasive duct carcinoma grade II cases.

Cases subjected to radical mastectomy operation showed positive lymph node metastasis in all of the invasive duct carcinoma Grade III cases and only 46.1% of grade II cases. A high statistical significance for these results was detected ($p<0.05$).

Most of the invasive duct carcinoma cases grades I and II and all cases of duct carcinoma in situ showed positive expression of estrogen receptor immunostaining. On the other hand, most of grade III invasive duct carcinoma cases showed negative estrogen and progesterone receptor expression. Her2/neu was expressed in less than half of all cases of invasive duct carcinoma. These results were statistically insignificant.

Most of the studied cases of extensive duct carcinoma in situ with focal invasion and invasive duct carcinoma grades II and III showed positive CD10 immunostaining. Meanwhile half of the cases of duct carcinoma in situ and invasive duct carcinoma grade I were CD 10 positive which was statistically insignificant.

Strong stromal CD10 immunostaining was only seen in 71.4% of invasive duct carcinoma grade III cases, while all duct carcinoma in situ cases were weakly stained stromal CD 10 immunostaining. There was a highly significant correlation between the intensity of stromal CD10 immunostaining and the tumor grade ($p<0.0002$).

There was no difference between stromal CD 10 immunostaining and different age groups of the studied cases. Moreover, these results were not statistically significant ($p>0.05$).

Also, there was no statistically significant difference between stromal CD 10 immunostaining intensity and tumor sizes in the studied cases ($p>0.05$).

Strong stromal CD10 immunostaining was seen in only 25% of cases with metastatic lymph nodes, with negative CD 10 immunostaining in node negative cases. No statistical significance for these results was attained ($p>0.05$).

Most of the estrogen, progesterone and Her2/neu positive cases showed weak CD 10 immunostaining, furthermore no statistically significant correlation between CD 10 immunostaining intensity and hormone profile expression in the studied cases was noted ($p>0.05$).

Cases that were negative for stromal CD10 immunostaining were also negative for estrogen receptor staining in 38.4% of cases, while cases showing strong CD 10 immunostaining showed equal distribution between negative and moderate estrogen receptor staining intensity (33.3% each). Nevertheless, these results showed no statistical significance ($p>0.05$).

Negative progesterone receptor immunostaining was seen in most of CD 10 negative cases (61.5%). However, no statistical significance was reached ($p>0.05$).

Her2/neu negative immunostaining was seen in the majority of cases whether negatively stained for stromal CD10 (92.3%), weakly stained for CD 10 (77.4%) or strongly stained for stromal CD 10 (83.3%). These results were not statistically significant ($p>0.05$).
Fig. (1): (A) Extensive duct carcinoma in situ with focal invasion showing diffuse weak CD10 immunostaining. X100. (B) Duct carcinoma in situ showing diffuse weak CD10 immunostaining. (C) Invasive duct carcinoma grade I, showing weak focal CD10 immunostaining in less than 30% of stromal cells X200. (D) Invasive duct carcinoma, grade III showing strong CD10 immunostaining in more than 30% of the stromal cells. X200. (E) Invasive duct carcinoma grade III, showing weak CD10 immunostaining in less than 30% of the stromal cells. X400. (F) Invasive duct carcinoma grade III, showing strong CD10 immunostaining in more than 30% of the stromal cells X200.

Table (1): Expression of CD10 immunostaining in mammary duct carcinoma cases.

<table>
<thead>
<tr>
<th>Tumor type</th>
<th>CD10 expression</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Positive</td>
<td>Negative</td>
</tr>
<tr>
<td>Duct carcinoma in situ</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Extensive DCIS with focal invasion</td>
<td>8</td>
<td>4</td>
</tr>
<tr>
<td>Invasive duct carcinoma Grade I</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Invasive duct carcinoma Grade II</td>
<td>18</td>
<td>1</td>
</tr>
<tr>
<td>Invasive duct carcinoma Grade III</td>
<td>7</td>
<td>4</td>
</tr>
<tr>
<td>Total</td>
<td>37</td>
<td>13</td>
</tr>
</tbody>
</table>

Table (2): Correlation between the intensity of CD10 immunostaining expression and hormonal profile expression positivity in mammary carcinoma cases.

<table>
<thead>
<tr>
<th>Hormone profile expression</th>
<th>CD10 immunostaining</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Negative</td>
<td>Weak</td>
</tr>
<tr>
<td>Positive Estrogen receptor</td>
<td>8 (25%)</td>
<td>20 (62.5%)</td>
</tr>
<tr>
<td>Positive progesterone receptor</td>
<td>5 (20.8%)</td>
<td>16 (66.7%)</td>
</tr>
<tr>
<td>Positive Her2/neu</td>
<td>1 (11.1%)</td>
<td>7 (77.8%)</td>
</tr>
</tbody>
</table>
Discussion

Breast cancer is a major concern worldwide and is one of the leading causes of death in women [7]. The estimated annual incidence of breast cancer worldwide is about one million cases (22% of all cancers in the world in 2000) [3]. According to the Egyptian National Cancer Institute, breast cancer constitutes about 33% of all Egyptian female cancers [2]. It is considered the most common cancer among women worldwide, of which infiltrating duct carcinoma is the most frequent histological type, accounting for approximately 68% of breast cancer cases [1].

Seventy two percent (72%) of our studied cases were diagnosed as invasive duct carcinoma, and (24%) were diagnosed as extensive duct carcinoma in situ with focal invasion, while the remaining (4%) of were diagnosed as duct carcinoma in situ. Furthermore, the invasive duct carcinoma cases, grade II represented the majority of cases studied (52.8%) of cases, followed by grade III which was diagnosed in (30.6%) of cases while grade I was diagnosed in only (16.6%) of cases.

Our results are similar to those reported by Makretsov et al. [4] which was conducted on 452 cases of mammary duct carcinoma, in which invasive duct carcinoma comprised 258 cases. Grade II represented the majority of invasive duct carcinoma cases as it was diagnosed in 53.8% of cases (139 cases), followed by grade III which was diagnosed in 26.4% of cases (68 cases), while grade I was noted in 19.8% of cases (51 cases). Correspondingly, in the study done by Yamaguchi et al., 2008 [8], it was observed that the most prevalent grade was invasive duct carcinoma grade II, representing 40% of studied cases.

Our results were conflicting to those reported by Putti et al. [7], whose most prevalent grade of invasive duct carcinoma cases was grade III which accounted for 94% of their studied cases followed by grade II which was found in 5% of their cases, while grade I was the least prevalent accounting for only 1% of their cases. Moreover, Lu et al. [9], stated that the majority of their studied cases were diagnosed as invasive duct carcinoma grade III in (75%), followed by grade II which constituted 24% of their cases, while grade I represented only 1% of their studied cases.

In the present work, 82% of the studied cases were patients above the age of 40 years. The patients’ age in the present work ranged from 25 up to 70 years, with mean age of 50. Also, somewhat similar results were obtained from Yamaguchi et al. [8], in which the patients’ age ranged from 27 up to 87 years with mean age of 52.

Dissimilar to our results, a higher mean patients’ age of 60.2, 61, and 62 was recorded in the work done by Van der Vegt et al. [10], Makretsov et al., 2007 [4] and Lu et al. [9], respectively.

Seventy eight percent of our studied cases showed tumor size less than 5cm in maximal diameter. They ranged from 1.5 up to 13.5cm in maximal diameters with mean tumor size of 3.7±2.57. Correspondingly, Lu et al., 2002 [9] observed in their study that 89% of their cases showed tumor sizes less than 5cm in maximal diameters. Tumor size in their study ranged from 0.6 up to 11 cm in maximal diameters, with mean tumor size of 3.2. Van der Vegt et al., 2010 [10], also reported that 93.5% of their studied cases showed tumor sizes less than 5cm in maximal diameters.

On the other hand much smaller tumor sizes were observed in the study done by Makretsov et al. [4], as 46.1% of their studied cases showed tumor sizes 2cm or less in maximal diameters, keeping in mind that 10.9% of their total cases were of unknown tumor sizes.

Contrary to our results the study done by Putti et al. [7], recorded that the majority (53%) of their studied cases showed tumor size greater than 5cm in maximal diameter.

In the current study, positive lymph node metastasis was seen in all cases of invasive duct carcinoma grade III, and was negative in all of the invasive duct carcinoma grade I cases. As regards to invasive duct carcinoma grade II cases, there was a slight predominance of lymph node negativity (53.9%). These results showed a highly significant correlation between tumor grade and lymph node metastasis (p<0.01).

Contrary to our results, Xiaojin et al. [11], revealed that 85% of their studied cases diagnosed as invasive duct carcinoma grades II and III showed positive axillary lymph node metastasis, compared to 67% of invasive duct carcinoma cases grade I and the difference was not statistically significant.

In the present study, we observed positive estrogen receptor expression in all cases of duct carcinoma in situ and most of invasive duct carcinoma cases grades I (83.3%) and II (84.2%). However, only 36.4% of cases of invasive duct carci-
noma grade III and 41.7% of cases showing extensive duct carcinoma in situ with focal invasion showed estrogen receptor positivity. No significant correlation was found between tumor grade and estrogen receptor expression ($p>0.05$).

Arafah [12], similarly noted that the majority of invasive duct carcinoma grade I cases (88.9%) showed estrogen receptor positivity and most of invasive duct carcinoma grade II cases (70.6%) were immunopositive to estrogen receptor compared to only 54.5% of invasive duct carcinoma grade III cases showing estrogen receptor positivity.

In our study, progesterone receptor positivity in all duct carcinoma in situ cases and the majority of invasive duct carcinoma grade II cases (63.2%) was observed. Additionally, only 33.3% of invasive duct carcinoma cases grade I, 36.4% of invasive duct carcinoma cases grade III and 33.3% of extensive duct carcinoma in situ with focal invasion showed progesterone receptor positivity. There was no statistically significant correlation found between tumor grade and progesterone receptor expression ($p>0.05$).

Contradictory to our results, Arafah [12], noticed positive progesterone receptor immunohistochemical staining in most of invasive duct carcinoma grade I cases (77.8%), 63.5% of invasive duct carcinoma grade II cases and about half of invasive duct carcinoma grade III cases (48.8%). Moreover her study stated a statistically significant correlation between tumor grade and progesterone receptors immunohistochemical expression. This disparity between her results & ours may be attributed to the larger sample size (164 cases of invasive duct carcinoma in comparison to 36 in ours).

As regards Her2/neu receptor, positive immunohistochemical expression was seen in less than half of all cases of invasive duct carcinoma grades II (42.1%) and III (9%), but none of the cases of invasive duct carcinoma grade I showed Her2/neu immunostaining. No significant correlation was observed in our study between tumor grade and Her2/neu receptor expression ($p>0.05$). Concurrently, Arafah, 2010 [12] stated that the vast majority of their studied invasive duct carcinoma cases of all grades, were Her2/neu receptor negative. Furthermore, she concluded that the correlation between Her2/neu receptor immunoreactivity and the tumor grade was also insignificant. Likewise, Ivković-Kapicel et al. [6], recorded that none of the invasive duct carcinoma grade I cases showed Her2/neu receptor positivity.

In contrast to our findings, Ivković-Kapicel et al., 2007 [6], conducted a study on the expression of Her2/neu receptor in different grades of invasive duct carcinoma and concluded that there was a statistically significant correlation between Her2/neu receptor expression and tumor grade. They found Her2/neu receptor overexpression in less than half of cases of invasive duct carcinoma grade III (41%) and 21% of invasive duct carcinoma grade II cases.

In the present work, positive stromal CD10 immunostaining was observed in most of the cases diagnosed as extensive duct carcinoma in situ with focal invasion (66.7%) and invasive duct carcinoma grades II (94.7%) and III (63.6%) as opposed to only half of cases (50%) diagnosed as invasive duct carcinoma grade I and duct carcinoma in situ.

Our results were insignificant concerning the correlation between stromal CD 10 positive immunostaining and tumor grade ($p>0.05$). This may be explained by the small sample size in our study and the uneven distribution of the different grades of mammary duct carcinoma cases studied (2 cases of duct carcinoma in situ versus 12 cases of extensive duct carcinoma in situ versus 36 cases of invasive duct carcinoma).

Somewhat similarly, Vandana et al. [13], observed that most of their cases, of all grades, showed CD 10 receptor positivity with an insignificant correlation between CD 10 immunostaining positivity and tumor grade.

Dissimilarly, Iwaya et al. [14], observed positive CD10 immunostaining in only 18% of invasive ductal carcinomas cases included in their study but in none of their non-invasive ductal carcinoma cases studied. Likewise, no statistically significant correlation between CD 10 immunostaining positivity and tumor grade was achieved in their study.

Dissimilar to our results, Makretsov et al. [4] found a significant correlation between CD 10 immunostaining positivity and tumor grade, as they noticed that most of invasive duct carcinoma cases were positively stained for CD 10 compared to less than half of duct carcinoma in situ cases.

Our current work achieved a highly significant correlation between the intensity of CD 10 immunostaining and tumor grade ($p<0.01$), as all cases diagnosed as duct carcinoma in situ with or without focal invasion, as well as invasive duct carcinoma grade II and most of invasive duct carcinoma grade I cases (66.7%) showed weak immunostaining compared to invasive duct carcinoma grade III in
which 71.4% of its cases showed strong immunostaining.

Somewhat similarly, Makretsov et al. [4] concluded that 94.3% of duct carcinoma in situ cases were weakly stained for CD 10. While strong CD 10 immunostaining was found in 59% of invasive duct carcinoma grade III cases. As for invasive duct carcinoma grades I and II, most of their cases showed weak staining. Their results were also statistically significant.

This statistically significant correlation between CD 10 immunostaining intensity and tumor aggressiveness may be attributed to the fact that CD 10, being one of the matrix metalloproteinases, cleaves the protein components of extracellular matrix and thereby plays a central role in tissue remodeling. Therefore, in mammary duct carcinoma, CD 10 by the degradation of the extracellular matrix, helps the tumor cells to invade the surrounding tissue stroma [15].

Our study did not achieve any significant correlation between stromal CD 10 immunostaining intensity and tumor aggressiveness in relation to patients' age or tumor size. In accordance with our study, the results obtained by Makretsov et al. [4], revealed the same insignificant correlation, as they concluded that neither patients' age nor tumor size correlated with stromal CD10 immunostaining intensity. Another study done by Iwaya et al. [14], similarly showed insignificant correlation between stromal CD 10 expression and patient's age, tumor size, moreover, clinical stage of the tumor.

Most of the invasive duct carcinoma cases in our study whether associated or not with lymph node metastasis showed weak stromal CD10 immunostaining. A significant correlation between stromal CD 10 immunostaining intensity and lymph node metastasis was not attained. Concurrently, Makretsov et al., 2007 [4], also found that CD10 immunostaining intensity was not affected by lymph node status.

Contradictory results were concluded by Iwaya et al. [14], which showed a significant correlation between lymph node metastasis and CD 10 immunostaining expression, stating that the positivity as well as the intensity of CD 10 stromal immunostaining were significantly higher in the cases associated with axillary lymph node metastasis. Weak stromal CD 10 immunostaining was observed in most of the cases showing positive staining for estrogen, progesterone and Her2/neu receptors (p>0.05). An insignificant correlation between stromal CD 10 immunostaining intensity and estrogen, progesterone or Her2/neu receptors positivity, including their staining intensity was observed in our study.

Similar conclusions were drawn by Makretsov et al. [4] concerning progesterone and Her2/neu receptors only. As regards estrogen receptors, unlike our study, they were found to be negative in most of the CD 10 positive cases (whether weak or strong intensity). A significant correlation between CD 10 immunostaining intensity and estrogen receptors expression negativity was confirmed in their study.

Although Vandana et al. [13], observed that the majority of estrogen and progesterone receptor negative cases showed stromal CD10 receptor overexpression, although they concluded that CD 10 receptor immunostaining intensity did not significantly correlate with either estrogen or progesterone receptor expression.

On the contrary to our results, Vandana et al., 2011 [13] stated that there was a strongly positive statistically significant correlation between CD 10 expression and HER2-neu expression.

CD 10 constitutes a clinically important prognostic marker and a potential target for development of novel therapies. We concluded in our study that there was a highly significant correlation between tumor grade and lymph node metastasis. The higher the tumor grade the greater incidence of lymph node metastasis. A highly significant correlation was also attained between the intensity of CD 10 immunostaining and tumor grade. The CD 10 immunostaining intensity is directly proportional to tumor grade. Most of the studied cases, regardless of the tumor grade, were that of patients above the age of 40 years. However, no significant correlation was noticed between tumor grade and patients' age. Most of the tumor sizes, regardless of the tumor grade, were less than 5cm in maximal diameters. However, no significant correlation was noticed between tumor grade and tumor size. Furthermore, no significant correlation was found between tumor grade and estrogen, progesterone or Her2/neu receptor expression. No significant correlation was noticed between CD10 immunostaining intensity and patients' age, tumor size, lymph node status and expression of estrogen, progesterone or Her2/neu receptors was attained. Lack of statistical significance attainment is most likely to the limited number of studied cases. Further studies with larger number of cases is recommended to further validate these findings.
References


