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VASCULAR ENDOTHELIAL GROWTH FACTOR (VEGF) UP-REGULATION IN SEVER INTERSTITIAL CYSTITIS

Hypothesis / aims of study

Interstitial cystitis (IC) is characterized clinically by urinary frequency, urgency and pain with cystoscopic appearance of glomerulations, fissuring or ulceration. The etiology of interstitial cystitis remains unclarified. Previous study[1)] proposed the etiology of IC was associated with bladder ischemia. A state of vascular insufficiency may cause decreased blood supply to the epithelium, which has important implications such as reduced urothelial barrier function. This insufficiency may explain the symptom including the pain that develops with increasing volume. Additionally, glomerulations have been one of the important cystoscopic appearances for diagnosis of IC. A recent report[2)] has demonstrated that glomerulation during hydrodistention was highly associated with overexpression of angiogenic factors, including vascular endothelial growth factor (VEGF) and platelet derived endothelial cell growth factor/thimidine phosphorylase (PD-ECGF/TP)

Therefore, our hypothesis is that patients with severe symptom may have decreased blood supply in the bladder, which results in the overexpression of angiogenic factors. In this study we determined the correlations between the expression of VEGF and the symptom or clinical features of the IC patients.

Study design, materials and methods

Included in our study were 19 women and one man with a mean age ±standard deviation of 58.2±15.3 years who had interstitial cystitis. For symptom assessment select questions from the National Institute of Diabetes and Digestive and Kidney Disease International Database Symptom Questionnaire were used to assess symptom severity. Each study patients underwent hydrodistention under spinal anesthesia with sorbitol irrigating solution until bladder capacity reached 600 ml or the bladder pressure reached 80 cm. water pressure. After 10 minutes distension of the bladder, the site of maximum glomerulation was obtained with cold cup biopsy. The specimens were fixed in 10% buffered formalin and embedded in paraffin blocks. Four micrometer thick paraffin sections was used for immunohistochemical study using polyclonal anti-VEGF and monoclonal anti-CD-34. Immunohistochemical reactions were developed with diaminobenzidine as the chromogenic peroxidase substrate. Sections were counterstained with hematoxylin after immunohistochemistry. All subjects provided informed consent, and the study was approved by the ethics committees at our hospital.

VEGF staining was considered positive if appropriate red staining was seen in the cell cytoplasm. We considered microvessels as individual or clusters of cells with or without lumens, positively stained by anti-CD34. In each specimen, the three areas with the highest vascularization (hot spot) were selected. Individual microvessel counts were then made on a 200× field (20×objective and 10×occular, corresponding to an area of 0.363 mm^2) by two independent observers. The average count from the two observers was used as the final score. Results were expressed as the mean microvessel density (MVD) per field for the three hot spots.

Results

The expression of VEGF was located in the extracellular space of the lamina propria, epithelium and within inflammatory cells. In all specimens VEGF was expressed within epithelium cells. In the lamina propria, 9(45%) expressed VEGF. VEGF-positive patients were significantly ($p = 0.047$) higher in symptom score than VEGF-negative patients (14.6 vs. 10.5, respectively). Of 8 patients with pain, 7 were VEGF –positive and only one was VEGF-negative ($p=0.0045$). MDV was 46.7 /mm^2 for VEGF-positive and 46.4 /mm^2 for VEGF negative. There was no significant difference between VEGF-positive patients and VEGF-negative patients in problem score, age, duration time and compliance.

Interpretation of results

In bladder biopsy specimens from IC patients, we found that the VEGF expression was up-regulated in the lamina propria in patients with severe symptom. Further, all IC patients with pain but one were VEGF-positive in the lamina propria. A recent study demonstrated that bladder hypoxia/ischemia following partial bladder outlet obstruction could increase the expression of VEGF. Patients with severe symptom may have decreased blood supply to the bladder, since the VEGF expression was increased. In a previous report, when the bladder was filled to capacity, overall blood flow increased a mean of 7.6 laser Doppler flowmetry units in the control group and only 3.4 in the interstitial cystitis group. A state of vascular insufficiency may cause decreased blood supply to the epithelium, which has important implications such as reduced urothelial barrier function. This insufficiency may explain the symptom including the pain that develops with increasing volume. Thus, decreased bladder perfusion in IC patients may increase the expression of VEGF and symptom such as urinary frequency, pain and urgency.

MVD was not significant different between VEGF-positive and VEGF-negative patients. Our data are consistent with previous results[3)]. They have demonstrated a lower blood vessel count in the IC patients in the suburothelium by CD-34 staning than in controls, but no significant difference between the IC patients and severe IC patients. Therefore, MVD may not explain the severity of the symptom in IC patients.

Concluding message

We demonstrate VEGF is highly associated with severity of symptom in IC patients. Thus, it seems likely that ischemic state, which may promote angiogenic growth factors, leads the mucosal permeability disturbance causing pain in IC patients.

Table 1. Characteristics of VEGF-positive or VEGF-negative IC patients
(Figures show mean value or number (%))

	VEGF(+) n=9	VEGF(-) n=11	p value
age(y)	65.2	52.5	0.13
symptom score	14.6	10.0	0.047
problem score	11.9	10.4	0.22
pain	7 78	1 (9)	0.0045
MVD (/mm2)	46.7	46.4	0.98
compliance (ml/cmH20)	5.9	8.9	0.25

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