

## Expression of Vascular endothelial growth factor in psoriasis: An Immunohistochemical Study



### Medical Science

KEYWORDS :

**Nath Dwijendra** Department of Pathology, MLB Medical College, Jhansi

**Singh Mayank** Department of Pathology, MLB Medical College, Jhansi

**Srivastava Neeraj** Department of Dermatology, MLB Medical College, Jhansi

**Sahay Renu** Department of Pathology, MLB Medical College, Jhansi

**Rawat Niharika** Department of Pathology, MLB Medical College, Jhansi

**Singh Arun** Department of Pathology, MLB Medical College, Jhansi

### ABSTRACT

*Background: Psoriasis is a chronic inflammatory disorder which is immune mediated and affects skin and joints of genetically predisposed individuals. It is characterized by hyperproliferative epidermis along with abnormal differentiation of keratinocytes, lymphocytic infiltration in the dermis along with vascular changes such as angiogenesis and dilatation. Vascular Endothelial growth factor (VEGF) works as a bridging link between angiogenesis and inflammation & plays a key role in the pathogenesis of psoriasis. The aim*

*of this study was to investigate the immunohistochemical expression of VEGF in psoriatic skin and to detect their role in the pathogenesis of psoriasis*

*Method: VEGF expression was studied in lesional skin of psoriatics by immunohistochemistry.*

*Results: In psoriatic skin VEGF was strongly expressed by epidermal keratinocytes and moderately by inflammatory infiltrate and blood vessels.*

*Conclusion: VEGF expression was upregulated in psoriatic's skin. Hence there is possible link between VEGF expression and psoriasis pathogenesis.*

### Introduction:

Psoriasis is a chronic, T-cell mediated inflammatory disease of the skin and occasionally involves the joints [1]. The disease is characterized by the occurrence of red scaly patches and, in 5% of cases, is complicated by severe arthritis. The pathogenesis of psoriasis has not been elucidated yet but there is a role of T-lymphocytes and proinflammatory cytokines in the development of skin lesions in psoriasis [2]. Psoriasis is characterized by hyperproliferative epidermis along with abnormal differentiation of epidermal keratinocytes, lymphocytic infiltration in the dermis along with vascular changes such as angiogenesis and dilatation [3].

Vascular endothelial growth factor (VEGF), also known as vascular permeability factor originally described as an endothelial cell-specific mitogen is an essential growth factor for endothelial cells. In addition, it is a potent angiogenic agent and very potent hyperpermeability factor [4].

Several angiogenic factors have been identified in psoriatic epidermis, including VEGF, IL-8, TNF-alpha, transforming growth factor-alpha, Endothelial Cell-stimulating angiogenesis factor [5]. It is suggested that Psoriasis is an angiogenesis-dependent disease, and vasoproliferation is a suitable target for the development of antipsoriatic drugs [6].

All these findings indicate that VEGF plays an important role in the pathogenesis of psoriatic skin lesions. However, the exact mechanisms of VEGF in pathogenesis of psoriasis is unknown.

### Materials and Methods

This study was carried out on 52 patients with Psoriasis. There were 37 male patients and 15 female patients and their ages ranged between 10 and

70 years. The participants included in this study were selected randomly from the outpatient clinic of the Dermatology and Venereology Department, MLB Medical College, Jhansi during

the period from April 2014 to March 2015. The patients were selected on the basis of clinical features favouring psoriasis. Five healthy volunteers were also included in this study as a control group.

A 6-mm punch biopsy was taken from each patient under local anesthesia (2% xylocaine). The punch biopsy specimen was obtained from a fully developed psoriatic plaque, near the margin. Each specimen was fixed in 10% buffered neutral formalin solution and submitted in the Department of Pathology for histopathological evaluation. The biopsies were embedded in paraffin to form paraffin blocks, and serial sections of 4 µm thickness were obtained from each block. The diagnosis of Psoriasis was confirmed by histopathological features (epithelial hyperplasia, parakeratosis, Munro's micro-abscesses, suprapapillary thinning, inflammatory infiltrate in the papillary dermis, widened rete ridges, and capillary dilatation). An immunohistochemical staining for anti-VEGF (Biogenex) was done.

### Immunohistochemical analysis

Serial 4 mm thick sections were made and mounted on poly-L-lysine coated slides. Paraffin sections were immersed in xylene for 5 min and hydrated using a gradient series of alcohol. Antigen retrieval was routinely performed by immersing the sections in citric acid buffer (pH 6.0); in a microwave oven for 15 min. Endogenous peroxidase activity was blocked with 3% hydrogen peroxide for 10 min and then incubated with a primary antibody in a humidified chamber at 4°C overnight. Primary antibody was monoclonal VEGF at a 1:200 dilution. A total of 500 keratinocytes were enumerated in an area that was stained intensely at ×400 under the light microscope. The results of the immunostaining were analyzed semi-quantitatively as a percentage of positive cells. Localization of immunohistochemical staining was grouped and classified as epidermal cells, lymphocyte and endothelial cells staining.

### Immunostaining for VEGF was semiquantitatively scored as follows:

0, Negative.

1, Weak (<10% expression of cells).

2, Moderate (10–20% expression of cells).

3, Strong (expression in >50%).

A score of 2 or higher was considered positive [7].

#### Statistical analysis

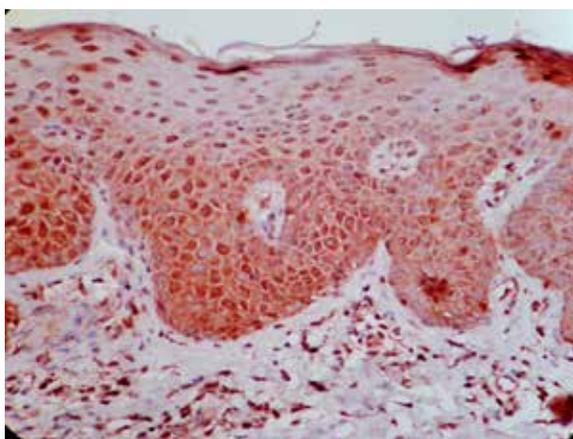
The results of the study were statistically analyzed using SPSS 13.0 statistical package program (Statistical Package for Social Sciences, Lead Technologies Inc., USA). A value of  $P < 0.05$  was considered to indicate statistical significance.

#### Results

52 cases of psoriasis have been evaluated in the present study. The mean age was 36.4 years with a age range of 11-70 years. Out of 52 patients 37 were males and 15 were females.

#### Immunostaining

In Psoriasis, we found a strong cytoplasmic VEGF expression throughout the epidermis and a moderate expression in inflammatory infiltrate and blood vessels in 49 out of 52 (94.3%) of cases. [Fig 1 & 2] However, in the healthy skin, VEGF expression was faint in the epidermis. VEGF was significantly upregulated in psoriatic specimen in comparison with normal healthy skin ( $P < 0.05$ ).



#### Discussion

Psoriasis is a T-cell mediated disease with hyperproliferative epidermis along with abnormal differentiation of epidermal keratinocytes, lymphocytic infiltration in the dermis and vascular changes such as angiogenesis and dilatation. Angiogenesis is an active process and appears to be an important inflammatory response in the pathogenesis of Psoriasis [8].

Vascular Endothelial Growth Factor (VEGF) signalling plays a critical rate limiting step in physiological and pathological angi-

ogenesis. It is a potent angiogenic factor and was first described as an essential growth factor for vascular endothelial cells. The receptors for VEGF (VEGFR-1 or VEGFR-2) are primarily expressed by Vascular Endothelial cells [9]. VEGF receptors are also expressed on numerous non-endothelial cells including the keratinocytes.

In psoriasis, besides its potential role in causing angiogenesis and vascular leakage in the upper dermis, VEGF also contribute to keratinocyte proliferation [10]. In psoriatic skin, the VEGF receptors, VEGFR-1 and VEGFR-2, are detectable and functional in the keratinocytes [11].

In Psoriasis, we detected a strong VEGF expression throughout the epidermis in 94.3% of patient as compared to the healthy control which showed faint & diffuse expression of the VEGF in the epidermis.

These findings are comparable with findings of Simonetti *et al.* [12]

who found that VEGF was strongly expressed in Psoriasis throughout the epidermis.

Avramidis *et al.* [13] found that the mean VEGF expression in psoriatic skin, and they reported reduction in VEGF expression by 81.6% at week 10 under etanercept treatment.

In contrast, in the healthy skin, we noted that VEGF expression was faintly diffused, a finding that is in agreement with the study by Simonetti *et al.* [12]. VEGF was significantly upregulated in psoriatic skin in comparison with normal healthy skin ( $P < 0.05$ ).

VEGF enhances the mitogenic activity of keratinocytes *in vitro* and also upregulates expression of VEGF receptor on both keratinocytes and Endothelial cells. This appears to play a key role in contributing to the process of angiogenesis and the hyperplastic psoriasiform epidermis [14].

Since much literature is not available in this context it can only be said that VEGF probably actively contributes to the pathogenesis of Psoriasis by the induction of permeability in dermal vessels, its chemotactic action on inflammatory cells and Endothelial cells, along with its angiogenic effect [15].

#### Conclusion

VEGF proved to be an important factor in the pathogenesis of Psoriasis. These findings form a basis of developing therapeutic strategies in which drugs inhibiting angiogenesis play a key role.

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