Excessive angiogenesis associated with psoriasis as a cause for cardiovascular ischaemia

<table>
<thead>
<tr>
<th>Journal</th>
<th>Experimental Dermatology</th>
</tr>
</thead>
<tbody>
<tr>
<td>Manuscript ID</td>
<td>EXD-14-0219.R3</td>
</tr>
<tr>
<td>Manuscript Type</td>
<td>Viewpoint</td>
</tr>
<tr>
<td>Date Submitted by the Author</td>
<td>n/a</td>
</tr>
<tr>
<td>Complete List of Authors</td>
<td>Malecic, Nina; The University of Manchester, Manchester Academic Health Science Centre, Department of Dermatology Young, Helen; The University of Manchester, Manchester Academic Health Science Centre, Department of Dermatology</td>
</tr>
<tr>
<td>Keywords</td>
<td>Psoriasis, Vascular endothelial growth factor, Angiogenesis, Cardiovascular disease, Atherosclerosis</td>
</tr>
</tbody>
</table>
Excessive angiogenesis associated with psoriasis as a cause for cardiovascular ischaemia

Nina Malecic and Helen S Young (orcid.org/0000-0003-1538-445X)

The University of Manchester, Manchester Academic Health Science Centre, Department of Dermatology, Manchester, UK

Corresponding Author:
Dr. Helen Young,
Department of Dermatology,
Salford Royal Hospital,
Manchester,
M6 8HD, UK.
Email: Helen.s.young@manchester.ac.uk

Key words: psoriasis, vascular endothelial growth factor, angiogenesis, cardiovascular disease, atherosclerosis

Funding source
None

Conflict of interest
No conflict of interest to declare
Abstract

Psoriasis, a common disease affecting 2-3% of the UK population, produces significant impairment of quality of life and is an immense burden on sufferers and their families. Psoriasis is associated with significant cardiovascular co-morbidity and the metabolic syndrome. Angiogenesis, a relatively under-researched component of psoriasis, is a key factor in pathogenesis of psoriasis and also contributes to the development of atherosclerosis. Vascular endothelial growth factor (VEGF) is a well established mediator of pathological angiogenesis which is upregulated in psoriasis. It is possible that, in patients with psoriasis, cutaneous angiogenesis may be both a marker for systemic vascular pathology and a novel therapeutic target. In this viewpoint paper the role of VEGF mediated angiogenesis as a cause for cardiovascular events in patients with psoriasis is explored.

Introduction

Psoriasis is a common, immune-mediated inflammatory disease that occurs in 2-3% of the population of the UK\(^1\). In early-onset psoriasis, developing before the age of 40 years (Type 1 psoriasis)\(^2\) and accounting for over 75% of patients, genetic predisposition in conjunction with an environmental trigger, such as infection or stress, is important for disease expression\(^1\).

Psoriasis is associated with an increased risk of cardiovascular disease (CVD) and patients with psoriasis develop major adverse cardiovascular events more frequently than individuals without psoriasis. A prospective population based cohort study in British patients with psoriasis reported that after adjustment for traditional cardiovascular risk factors; patients with severe psoriasis had an increased relative risk of myocardial infarction and that psoriasis was an independent risk factor for CVD\(^3\). In addition, there is strong evidence that patients with psoriasis are more frequently affected by components of the metabolic syndrome (traditional risk factors for CVD) than healthy controls\(^3,4\). Although, the high prevalence of traditional risk factors for CVD in patients with psoriasis raises the possibility that the disease per se may not be an independent risk factor for CVD – the “intrinsic” psoriasis-related CVD risk, deserves
further investigation at the molecular level\textsuperscript{5,6}. Although a number of explanatory models, mostly related to the underlying inflammatory process, have been experimentally documented, angiogenesis as direct disease-related influence on CVD risk in psoriasis is relevant and merits further consideration.

Angiogenesis, the formation of new blood vessels from a pre-existing vascular bed, is a significant component of the pathogenic mechanisms involved in tumour growth and metastasis, arteriosclerosis and psoriasis\textsuperscript{7,8}. Vascular endothelial growth factor (VEGF; also known as VEGFA) produced by epidermal keratinocytes promotes angiogenesis, enhances vascular permeability\textsuperscript{7} and is upregulated in psoriasis\textsuperscript{8,9}.

Atherosclerosis is the primary cause of coronary artery disease and stroke in western populations\textsuperscript{10}. Evidence suggests that angiogenesis of the arterial vasa vasorum is a key factor in the growth and subsequent destabilisation of atherosclerotic plaques\textsuperscript{11}. VEGF, in conjunction with other cytokines, has an important function in co-ordinating and promoting the growth of the atherosclerotic plaque (Figure 1)\textsuperscript{12}. In this paper the role of VEGF mediated angiogenesis in the development of both psoriasis and atherosclerosis is discussed with key evidence summarised in Table 1. A key role for VEGF as a mediator of cardiovascular co-morbidity in patients with psoriasis is postulated.

### Cardiovascular disease (CVD), Angiogenesis and VEGF

Atherosclerosis is an inflammatory process, characterised by a progressive series of events within the arterial wall\textsuperscript{10}. Initially lipid accumulation in the arterial wall produces a fatty streak, subsequent infiltration by monocytes produces the lipid core of the atheromatous plaque\textsuperscript{10}. Advanced atheromatous plaques can cause local obstruction of the arterial lumen or they can destabilise and rupture\textsuperscript{10,12}. Ruptured atherosclerotic plaques cause 75% of the total fatal acute myocardial infarction cases reported in the Western world\textsuperscript{13}.

Although the pathogenesis of atherosclerosis has been extensively investigated the key question of how an asymptomatic stable atherosclerotic plaque is transformed into a high-risk
lesion capable of rupture remains unanswered\textsuperscript{12}. Over the last decade clinical investigation has focused on identifying the morphology and characteristics of stable versus vulnerable plaques\textsuperscript{14-16} whereas animal studies have investigated the mechanisms of plaque destabilisation\textsuperscript{17-20}. VEGF mediated angiogenesis appears to play crucial role in the progression from stable atherosclerosis to rupture-prone lesions\textsuperscript{16} and recently a disease progression model of angiogenic regulation of vulnerable plaque development was postulated\textsuperscript{12}. The key features of this model are detailed below and schematically outlined in Figure 1.

The healthy arterial wall receives oxygen by diffusion from the vessel lumen. In early atherosclerosis the expanded extracellular matrix results in thickening of the vessel intima and oxygen diffusion becomes insufficient to meet metabolic demand. Consequently, expression of hypoxic-inducible factor (HIF) promotes local angiogenesis\textsuperscript{21,22}. HIF is a transcription factor which is made up of two sub-units, a HIF-1β subunit and a hypoxic responsive subunit HIF-1α. Migration of the HIF-1α-β dimer to the nucleus initiates upregulation of multiple angiogenic factors, including VEGF\textsuperscript{21}. In a physiological context, this up-regulation of angiogenesis can be helpful in restoring vessel wall normoxia, removal of intimal fat and the regression of atherosclerosis\textsuperscript{21}.

In progressive atherosclerosis, vascular inflammation causing increased metabolic demand for oxygen and local hypoxia due to increased arterial intima thickness results in a sustained trigger for angiogenesis\textsuperscript{21,23}. An animal model of atherosclerosis - the hypercholesterolaemic apolipoprotein E-deficient (ApoE\textsuperscript{-/-}) mouse - provided the first direct evidence that angiogenesis was involved in the progression and pathogenesis of atherosclerosis by demonstrating that endothelium-specific inhibitors of angiogenesis, endostatin and angiostatin reduce plaque area and atherosclerosis\textsuperscript{24}. The extent of new vessel formation within atheromatous plaques is directly related to rupture susceptibility\textsuperscript{23,25}. Studies have identified that vasa vasorum expansion from the adventitia into the arterial intima is increased 2-fold in advanced atherosclerotic lesions and 4-fold in ruptured lesions, as compared to stable plaques\textsuperscript{16}. 
Levels of VEGF in lesions of atherosclerosis have been observed to rise during disease progression\textsuperscript{12,26-27}. Notch and fibroblast growth factor (FGF) signalling are important pathways which regulate and synergise VEGF-induced angiogenesis in the early phase of vulnerable plaque development\textsuperscript{28,29}. Subsequently, survival and expansion of the intra-plaque vascular network is facilitated by angiopoietin-(Ang)\textsubscript{1}\textsuperscript{30-32}. VEGF and other cytokines recruit inflammatory cells into the plaque via the luminal endothelium and the newly formed microvasculature. Chemotaxis of CD16+ monocytes can further augment plaque progression and vulnerability\textsuperscript{33}. As the inflammatory state of the vulnerable plaque increases key molecular triggers such as TNF-\(\alpha\) in conjunction with persistently high levels of VEGF effects a switch in Ang stimulation toward Ang-2-dominated signalling\textsuperscript{12,34}. This promotes further inflammation by activation of adhesion molecules on the endothelial cell membrane and through repression of eNOS-mediated atheroprotection\textsuperscript{35-37}. Loss of cell-cell junctional integrity permits extravasation of white blood cells and leakage of erythrocytes into the plaque of atheroma\textsuperscript{12}. Finally, impaired Platelet-Derived Growth Factor (PDGF) B /PDGF Receptor (PDGFR) signalling results in diminished pericyte coverage of the intraplaque microvessels which can haemorrhage and cause rupture of the plaque\textsuperscript{38}.

These observations support a role for angiogenesis causing atheromatous plaque growth beyond a critical thickness - intimal thickening may have an initial angiogenesis-independent phase, followed by an angiogenesis-dependent phase\textsuperscript{39}. VEGF concentration is critical in determining biological outcomes in vivo\textsuperscript{40}. VEGF in low concentration appears to be cardio-protective whereas, high concentrations of VEGF are pro-atherogenic\textsuperscript{11,40}.

The VEGF gene is polymorphic and the two most commonly occurring single nucleotide polymorphisms (SNPs) in the promoter and 5’ untranslated region have been associated with regulation of VEGF production\textsuperscript{41,42}. Several studies have identified polymorphisms from this region of the VEGF gene may modulate clinical outcome in a variety of angiogenesis-dependent diseases\textsuperscript{43-47}. An association between polymorphisms / haplotypes from this key area of the VEGF gene and the development of atherosclerosis was observed in a large UK
This study hypothesised that genetic regulation of VEGF expression could be a pivotal risk / protective factor in the pathogenesis of atherosclerosis. Similarly, a sub-study of the Metoprolol CR/XL Randomised Intervention Trial in Heart Failure (MERIT/HF), identified an association between the VEGF +405 CC polymorphism and poor prognosis in patients with chronic heart failure (CHF). The authors speculated that insufficient production of VEGF / angiogenesis consequent upon VEGF gene variation could influence the pathophysiology of CHF.

**Psoriasis and Angiogenesis**

Microvascular changes in plaques of psoriasis include pronounced dilation, increased permeability and endothelial cell proliferation within the venous limb of capillaries in the superficial dermis. This increased upper dermal vascularity is evident clinically as the Auspitz sign where successive removal of psoriatic scales reveal numerous small bleeding points where the thinned suprapapillary epithelium has been torn off to expose the elongated, dilated and tortuous papillary capillaries.

Examination of the skin has demonstrated that structural change in the cutaneous capillary bed is the first (visible) step in the pathogenesis of psoriasis. Excessive capillary-venular dilatation precedes development of inflammation in patients with psoriasis and resolution of these vascular changes heralds clearance of plaques of psoriasis. Vascular expansion in plaques of psoriasis is limited to vascular enlargement, increased tortuosity and elongation rather than new growth per se from the pre-existing vascular bed – the term inflammatory angiogenesis has been coined to describe this phenomenon. Histological study of early lesions of psoriasis has established that these changes are due to variation in amounts of VEGF isoforms within the skin. In addition, the cytokine responsiveness of microvascular endothelial cells is altered in psoriasis in a pattern, which mimics the plaque type configuration and epidermal involvement of individual lesions.
VEGF is produced predominantly by keratinocytes in both the clinically involved and uninvolved skin of patients with stable, chronic plaque psoriasis\(^8\) and TGF-\(\alpha\) potently upregulates VEGF levels in a paracrine fashion\(^8,9,58-60\). VEGF and endothelial cell stimulating angiogenesis factor, are significantly elevated in plaques of psoriasis and that these levels appear to correlate with clinical severity\(^9\). Overexpression of VEGFR-1 and VEGFR-2 receptors in dermal microvascular endothelium has been reported in psoriasis\(^8\), healing skin wounds\(^61\) and delayed hypersensitivity reactions\(^62\). Furthermore, elevated levels of VEGF have been reported in the plasma of patients with erythrodermic psoriasis\(^63\) and it has been reported that patients with moderate-severe or severe psoriasis have plasma VEGF levels that are significantly increased during relapse of psoriasis as compared with remission\(^64\). Plasma levels of VEGF are also significantly increased in patients with stable chronic plaque psoriasis\(^65\).

The VEGF gene is located on chromosome 6 at 6p21, close to PSORS 1 a known chromosomal locus for psoriasis susceptibility\(^66\). VEGF genotype distinguishes two groups of patients with psoriasis - “high” and “low” VEGF producers\(^66,68\). The “high VEGF producing” genotypes show significant association with early-onset psoriasis and development of severe disease whereas the “low VEGF producing” genotypes show no association with psoriasis. These findings suggest that the “angiogenetic constitution” of an individual might influence both psoriasis susceptibility and phenotype such that those individuals with “high VEGF producing” genotypes could manifest a “pro-angiogenic” psoriasis phenotype\(^65,69,70\).

VEGF-transgenic mice, which overexpress VEGF in the epidermis have vascular expansion within the superficial dermis\(^71\). The chronic inflammatory response mediated by constitutive VEGF expression, in mice homozygous for the VEGF transgene, bears a resemblance to key features of psoriasis morphologically, histologically and immunologically\(^72\). This psoriasis-like phenotype can be reversed by treatment with a potent VEGF receptor antagonist - VEGF-Trap – suggesting that maintenance of psoriasis-like chronic inflammation is a VEGF dependent process\(^72\).
Molecular mechanisms common to both atherosclerosis and psoriasis.

Psoriasis and atherosclerosis have many similarities in their underlying pathophysiology including key stimuli / initiating events, cytokines and molecular signalling pathways\textsuperscript{73}.

In the early stages of development of both psoriasis and atherosclerosis, stimuli such as local hypoxia trigger the release of proangiogenic factors including hypoxia inducible factor-1 (HIF-1)\textsuperscript{74-75}. Hypoxia may result from increased oxygen demand or decreased oxygen supply as a consequence of active inflammation or increasing diffusion distance in a thickening psoriasis or atheromatous plaque. Thereafter, expression of further pro-angiogenic cytokines, including VEGF, results in the formation of new vessels and facilitates leukocyte transmigration into areas of inflammation via enhanced expression of cell adhesion molecules\textsuperscript{55,71}. Inflammatory cells such as macrophages and T lymphocytes infiltrate the skin or vessel wall through these newly formed capillaries, effecting release of a number of pro-inflammatory cytokines many of which have pro-angiogenic effects, including IL-8, TNF-α, and IL-17\textsuperscript{76-77}. These observations have led to suggestion that in addition to similarities in the angiogenic pathway between atherosclerosis and psoriasis, oxidative stress is also a key area of commonality between both pathologies\textsuperscript{73}. Oxidised phospholipids (OxPL) are key promoters of angiogenesis in atherosclerosis which stimulate transcription of other pro-angiogenic and pro-inflammatory mediators. OxPL also upregulate VEGF expression from keratinocytes, suggesting a potential role in psoriasis angiogenesis\textsuperscript{78}. Ischaemia is a key metabolic determinant for ROS production for both diseases and the main enzymatic sources of ROS are similar for both conditions. \textcolor{blue}{Increased production of ROS and upregulation of HIF-1α results in activation of the JAK-STAT, NF-κB, and MAPK signaling pathways.} which have been implicated in the promotion of both psoriasis and cardiovascular disease\textsuperscript{73}.

There is a growing body of evidence to support Wnt signalling involvement in many key aspects of atherosclerotic lesion development, from the initially dysfunctional endothelium to the vascular remodelling observed following myocardial infarction\textsuperscript{79}. There is altered expression of Wnt signalling proteins in patients with psoriasis including a five-fold
upregulation of Wnt5A transcripts accompanied by increased Wnt-5a protein levels in lesional skin. Expression of Frizzled 2 (FZD2) and FZD5, which encode receptors for Wnt5A have also been reported as increased in lesional psoriatic skin. Studies has evidenced a shift towards noncanonical Wnt signalling pathways in psoriasis accompanied by impairment of the homeostatic inhibition of Wnt signalling by Wnt inhibitory factor (WIF)-1 and dickkopf

The Notch signalling pathway is important in regulating both physiological and pathological angiogenesis. Notch modulation of VEGF signalling has also been described. Notch ligand DLL4-Notch signalling is specifically upregulated in the proliferating micro-vessels of atherosclerotic plaques that are vulnerable to rupture. Notch signalling can modulate the inflammatory response of vulnerable atherosclerotic lesions and DLL4 blockade can diminish further plaque development in a murine model of atherosclerosis. Notch signalling has been identified as a coordinating factor in VEGF mediated angiogenesis in psoriatic arthritis and may influence the fate and differentiation of T cells in patients with psoriasis.

Opportunity for novel treatment strategies.

Controlling pathological angiogenesis by regulating inappropriately activated VEGF / VEGFR-2 is a potential therapeutic strategy for the treatment of vascular diseases. Clinically, plasma levels of VEGF have been shown to predict adverse cardiac events in patients with known atherosclerosis. The amount of VEGF in plaques of psoriasis has been shown to correlate with clinical severity of disease. It is possible that upregulation of VEGF secondary to one disease process may influence or worsen the other. The likely time-course of the development of these parallel pathologies is speculative but worthy of further research. TNF-α has been shown to function as an upstream inducer of several pro-angiogenic pathways. Anti-TNF-α therapy can downregulate levels of many inflammatory cytokines within psoriatic plaques, including the angiogenic cytokines Ang 1 and 2 and their receptor Tie2. There is emerging evidence demonstrating improvement in cardiovascular outcomes in inflammatory disease such as rheumatoid arthritis following treatment with TNF-α inhibitors.
Existing VEGF inhibitors target the VEGF pathway in various ways including: i) direct inhibition of VEGF protein (anti-VEGF monoclonal antibodies - bevacizumab and ranibizumab); ii) prevention of VEGF receptor binding (VEGF receptor antagonists - alflibercept/VEGF-Trap and pegaptanib) and; iii) inhibition of VEGF receptor function through inhibition of tyrosine kinase (tyrosine kinase inhibitors (TKIs) - sunitinib, sorafenib, vandetanib and pazopanib). To date there are reports of five patients with psoriasis who were receiving treatment for malignancy with the VEGF inhibitors bevacizumab, sunitinib, and sorafenib who demonstrated improvement in their psoriasis. These clinical observations have also been replicated in animal models where dual inhibition of VEGFR-1 and VEGFR-2 (using the fusion-protein Afiblercept (Eylea™, Regeneron Pharmaceuticals, also known as V-Trap)) achieved significant amelioration of a psoriasis-like phenotype in transgenic VEGF mice. Furthermore, antiangiogenic (non-viral somatic) gene therapy was highly efficacious both in the prevention and treatment of psoriasis lesions in vivo by inhibiting angiogenesis and reduces the number and size of the microvessels in the skin. Recent work published in Experimental Dermatology identified a novel small-molecule inhibitor of VEGF / VEGFR-2 which demonstrated potent anti-angiogenic activity in both in vitro and in vivo investigations.

It is important to note that the VEGF inhibitors in current clinical use are associated with a number of potentially serious side effects including hypertension, left ventricular dysfunction and gastrointestinal perforation – a risk/benefit analysis which might be unfavourable for patients with psoriasis.

Conclusions

VEGF mediated angiogenesis is central to both the development of psoriasis and arterosclerosis and may contribute to the propensity for certain individuals to develop both conditions. It is possible that, genetically determined “high VEGF production” may drive expression of a severe psoriasis phenotype and contribute to the development of cardiovascular co-morbidity in a sub-group of patients with psoriasis.
Assessment of cutaneous vascularity in psoriasis could be a key determinant for disease prognosis and ultimately health outcomes for patients with psoriasis. Vascular signatures in both the skin and within the arterial vasa vasorum could be key in the design of personalised treatment regimens for patients.

Acknowledgements

The authors acknowledge the work of all who have contributed to this field and apologise to those investigators whose work has not been cited due to space and citation restrictions. Nina Malecic and Helen Young designed, wrote and approved the manuscript.

References


Figure Legend

Figure 1

A diagrammatic illustration of the recently proposed disease progression model of angiogenic regulation of vulnerable plaque development\textsuperscript{12}. \textcolor{red}{VEGF is a key driver of angiogenesis in all phases of the disease.}

Panel 1 (Phase 1) – thickening of the vessel intima results in expression of hypoxic-inducible factor (HIF) and promotion of local Vascular Endothelial Growth Factor (VEGF)-mediated angiogenesis.

Panel 2 (Phase 2) – persistently high levels of VEGF facilitates a switch in angiopoietin (Ang) stimulation toward Ang-2-dominated signalling.

Panel 3 (Final phase) - impaired Platelet-Derived Growth Factor (PDGF) B/PDGF Receptor (PDGFR) signalling results in diminished pericyte coverage of the intraplaque microvessels which can haemorrhage and promote rupture of the plaque.
Table 1: Key similarities between psoriasis and atherosclerosis

<table>
<thead>
<tr>
<th>PSORIASIS</th>
<th>Atherosclerosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Angiogenesis in the dermis of lesional skin</td>
<td>Intra-plaque angiogenesis</td>
</tr>
<tr>
<td>Plasma levels of VEGF are significantly increased in patients with i) psoriasis, ii) erythrodermic psoriasis and iii) during relapse of psoriasis as compared with remission</td>
<td></td>
</tr>
<tr>
<td>The extent of new vessel formation within atheromatous plaques is directly related to rupture susceptibility</td>
<td></td>
</tr>
<tr>
<td>Levels of VEGF, produced by keratinocytes, are significantly elevated in plaques of psoriasis and correlate with clinical severity</td>
<td></td>
</tr>
<tr>
<td>Levels of VEGF in plaques of atherosclerosis rise during disease progression</td>
<td></td>
</tr>
<tr>
<td>VEGF genotype distinguishes two groups of patients with psoriasis - “high” and “low” VEGF producers</td>
<td></td>
</tr>
<tr>
<td>VEGF promoter polymorphisms are associated with the development of atherosclerosis and may regulate progression of disease</td>
<td></td>
</tr>
<tr>
<td>The “high VEGF producing” genotype (+405 CC) is associated with early-onset psoriasis and development of severe disease</td>
<td></td>
</tr>
<tr>
<td>The VEGF +405 CC genotype is associated with poor prognosis in patients with chronic heart failure (CHF)</td>
<td></td>
</tr>
</tbody>
</table>
**Phase 1**

- Growth of tunica intima of blood vessel wall
- Hypoxia in core region of blood vessel wall
- Production of VEGF
- Adventitial microvascular response
- Induction of angiogenic sprouting of tip and stalk cell structures
- FGFR activation
- Proliferation and migration of endothelial cells
- Synergy with VEGF

**Phase 2**

- High levels of Ang1, FGF and VEGF
- Inflammatory cells enter via luminal epithelium and neovasculature
- Amplification of plaque inflammation
- Increased Ang1/Ang2 ratios
- Activation of adhesion molecules and loss of cell to cell junction integrity
- Further amplification of plaque
- Promoted by VEGF

**Phase 3**

- Impaired mural cell recruitment via PDGFB in neovasculature
- Absence of mural cell coverage in plaque microvessels
- Lack of inhibition of VEGF by PDGF causes reduced endothelial cell permeability
- Diminished pericyte-endothelial cell contact
- Hyperproliferation and functional dedifferentiation of endothelial cells
- Further weakening of the advanced lesion and tortuous microvessels with increased susceptibility to intraplaque haemorrhage
- Promoted by VEGF