Genetic susceptibility to raised dermal scarring

J.J. Brown* and A. Bayat†

*Plastic and Reconstructive Surgery Research, Dermatological Sciences, Manchester Interdisciplinary Biocentre, University of Manchester, Manchester, U.K.
†Department of Plastic and Reconstructive Surgery, South Manchester University Hospital Foundation Trust, Manchester, U.K.

Summary

Raised skin scars, such as keloid and hypertrophic scars mostly occur post-wounding in the human dermis. There is compelling evidence for a genetic component to these conditions, given the familial predisposition, varied incidence in different ethnic populations and the presence in twins. The aim of this study was to perform a systematic review of the literature regarding genetic susceptibility to raised dermal scarring. We identified relevant articles by a systematic search of relevant search engines. Key search terms included: keloid disease, hypertrophic scarring, fibrosis, linkage analysis, gene expression, human leucocyte antigen system (HLA), twins, families, case–control association study and congenital syndromes. Numerous candidate genes have been identified, along with potential linkage regions on different chromosomes. Recent data also suggest that carriers of specific major histocompatibility complex (MHC) alleles, in particular HLA-DRB1*15, HLA-DQA1*0104, DQB1*0501 and DQB1*0503, are at increased risk of developing keloid scarring. In addition, distinct immunophenotypical profiles can distinguish between keloid and hypertrophic scars. Keloid and hypertrophic scars are multifaceted aberrations of the healing process with as yet incompletely understood aetiologies. Current data suggest a genetic susceptibility with a strong immunogenic component to dermal fibrosis with MHC genes being implicated. It appears unlikely that a single gene is responsible for the development of raised dermal scars. A likely scenario may involve the interaction of several gene pathways in addition to environmental factors. The ability to assess accurately an individual’s potential genetic susceptibility to raised scarring may lead to a more personalized approach to their management in the future.
Identifying genetically susceptible individuals is critical, not only in the context of potential complications following invasive surgery, but also in terms of pre-, peri- and post-operative management and lifestyle choices for the susceptible individual. Most cases of HS and KD are managed under standard clinical regimens. In particular the management of KD has remained a difficult challenge for clinicians, with a reported high incidence of recurrence following any treatment modality including radiation. The recurrence rates on short-term follow-ups are reported to be lower for particular treatment modalities and particular phenotypes of keloids. However, longer-term follow-ups have shown similar recurrence rates for radiation, surgery or steroid injection, which are currently the most used methods of therapy.

Multiple treatments have been advocated, all with varying degrees of success, including established therapies such as surgery, steroids, radiation, lasers and silicone gel sheets, plus novel therapies such as interferon, 5-fluorouracil, etc. As with any therapy, the level of response to the treatment will vary from those who respond well, to those who show little or no improvement. Whether an individual is a ‘responder’ or a ‘nonresponder’ is likely to be influenced by the individual’s genetic characteristics as well as environmental factors.

Currently two patients both presenting with KD or HS, irrespective of their ethnicity, will be offered very similar treatments without the potential ‘range of responses’ being taken into consideration. Therefore, the ability to determine patients’ responses before treatment could enable clinicians to target treatment more effectively.

Following dermal trauma, the best wound healing response is considered to be a fine line scar. However a broad spectrum of scarring phenotypes exists, ranging from fine line (normal) scars to abnormal scars such as flat, atrophic (depressed), stretched (widespread), contracted (contractures) and raised scars (KD and HS). Abnormal scars never resolve and often demonstrate a high incidence of recurrence following treatment.

The aim of this review therefore is to summarize what is currently known regarding the genetics of raised dermal scarring and to offer perspectives on the direction of future research.

**Method**

We identified relevant articles by the systematic search of scientific and medical electronic search engines (PubMed, Scopus, Scirus). Key terms for the searches included: keloid disease, hypertrophic scarring, fibrosis, linkage analysis, gene expression, human leucocyte antigen system (HLA), twins, families, case–control association study and congenital syndromes.

---

Fig 1. (a) A typical hypertrophic scar on the upper outer arm of a white individual, which occurred following an excision of a simple lesion less than 6 months earlier. Note the hypertrophic scar has overgrown but remained within the boundary of the original lesion. (b) A typical keloid scar on the sternum in a white subject, which occurred after scratching a small pea-sized chicken pox lesion several years previously. Note the keloid scar has overgrown beyond the boundary of the original lesion.

**Hypertrophic scars**

HS are characterized by an exaggerated proliferative response to wound healing which, in contrast to keloid scars, stay within the boundaries of the original wound (Fig. 1a). HS are indurated, elevated and are characterized by hypervascularity, giving them an erythematous appearance at least during the first 7 months postinjury. The collagen in HS is generally present in a disorganized, whorl-like arrangement rather than in the normal parallel orientation observed in normal skin. HS can frequently develop within 2 months of a burn, wound closure with excess tension, wound infection or other traumatic skin injury. Their normal course involves a rapid growth phase for up to 6 months that may be followed by regression during the next 12–18 months. The incidence of HS is generally higher than that of KD; however, insufficient data exist to enable a statistically significant comparison to be made.

**Keloid disease**

KD is an abnormal wound healing response to cutaneous injury resulting in symptomatich, disfiguring and dysfunctional scars without any satisfactory treatment (Fig. 1b). KD is often described as a benign dermal fibroproliferative tumour characterized by excessive accumulation of extracellular matrix (ECM) proteins, leading in particular to an overabundance of collagen formation. KD is often a familial condition, occurring more commonly in ethnic groups with a common predisposition to darker skinned individuals where it has been estimated to occur at a frequency of 4–6% and up to 16% in random samples of black Africans. In Caucasians of northern European ancestry, the incidence of KD is reported to be as low as 0.09%. However, few epidemiological studies
have been conducted in individuals of Caucasian origin and consequently this reported level of incidence may be misleading. Strong evidence for a genetic component to KD is perhaps best demonstrated by its varied incidence in different ethnic populations, a familial history of the disease, its prevalence in twins and development of aggressive disease in multiple sites in those individuals with a family history. Jacobson in 1948 observed that twins who received a small pox vaccination both developed keloids at the same time (cited in Ref. 13). Various modes of inheritance from autosomal recessive to autosomal dominant with incomplete clinical penetrance and variable expression have been proposed for KD; however, no single disease-causing gene has so far been identified.\(^{1,11}\) Therefore, KD appears to be a complex oligogenic condition rather than a simple monogenic Mendelian disorder.

Genetically susceptible individuals form keloid scars following disruption to the dermis but not at every wound site. This clearly suggests that factors other than an individual’s ‘genetic make up’ are also involved in KD aetiology. A number of precipitating factors, all involving dermal disruption, have been reported to induce keloid scar formation in the susceptible individual.\(^5\) Spontaneous keloids have been reported in the literature; however, on detailed history taking and examination, the senior author (A.B.) has linked KD with a history of previous breach of the skin in the majority of cases.\(^6\)

**Familial predisposition and raised dermal scarring**

To date, a number of studies have been conducted to characterize further familial predisposition to keloids and modes of inheritance by investigating ‘keloid pedigrees’, i.e. large families with a high incidence of keloids. In most cases the data generated have been conflicting. Omo-Dare (1975) proposed an autosomal recessive inheritance pattern for KD, based on his study of a number of relatively small pedigrees.\(^{12}\) In contrast to this suggestion, Bloom (1956) proposed an autosomal dominant inheritance pattern for KD based on data generated from a single but large Italian family spanning five generations (cited in Ref. 13).

More recently, Marneros et al. (2001)\(^{13}\) studied clinical and inheritance patterns of KD in 14 pedigrees, all of which spanned a minimum of three generations and up to five generations. The observed pattern of inheritance of KD in these families was consistent with an autosomal dominant mode with incomplete clinical penetrance and variable pattern of expression. Their data suggested that a child with only one parent affected by KD had a 50% chance of developing the disease and that single gene mutations may predispose specifically to keloids. To date, no such studies have been performed in relation to HS.

**Congenital syndromes associated with keloid disease**

A number of rare congenital syndromes are reported to be associated with familial keloids. The most commonly associated syndromes are Rubinstein–Taybi syndrome,\(^{15–19}\) Goeminne syndrome\(^{20}\) and Ehlers–Danlos syndrome.\(^{21}\) Rubinstein–Taybi syndrome is the consequence of a mutation in the gene encoding the transcriptional coactivator cAMP-response element-binding protein (CRBP), which is located on chromosome 16p13.3. Goeminne syndrome on the other hand has been described as an X-linked condition with the gene locus assigned to Xq28.\(^{22}\) Extensive linkage analysis by Marneros et al.\(^{23}\) failed to demonstrate genetic linkage to these regions in any of the pedigrees investigated, and highlighted the fact that gene mutations can specifically predispose individuals to KD without causing other clinical conditions.

**Linkage analysis of keloid disease**

Several linkage analysis studies have been performed with the aim of pinpointing the ‘keloid gene locus’. The most comprehensive studies to date are those of Marneros et al., Chen et al. and Yan et al.\(^{23–25}\) Marneros et al.\(^{23}\) performed whole genome-wide scans in both an African-American family and a Japanese family. For the African-American family linkage to chromosome 7p11 with a likelihood ratio (LOD) score of 3.16 was reported. Of particular interest in this region is the gene epidermal growth factor receptor (EGFR) which has been implicated in keloid aetiology.\(^{26}\) The Japanese family pedigree showed evidence for a susceptibility locus on chromosome 2q23 (LOD score 3.01). Interestingly, Chen et al.\(^{24}\) analysed the keloid susceptibility loci on chromosome 7p11 in a Chinese Han pedigree. They concluded that this locus was not associated with keloid susceptibility, further endorsing the heterogeneity of KD and the different aetiology likely to exist in different ethnic populations. Yan et al.\(^{25}\) investigated two loci in the Chinese Han population, 15q22.31–q23 and 18q21.1. Their data suggested that in this population, 18q21.1 might be the susceptibility locus. The significance of this particular study is that the genes SMAD2, SMAD4 and SMAD7 [SMAD proteins are homologues of the Drosophila protein, mothers against decapentaplegic (MAD) and the Caenorhabditis elegans protein SMA] and PIAS2 [protein inhibitor of activated STAT (signal transducer and activator of transcription)] are located in this genomic region. SMAD genes and their interaction with transforming growth factor (TGF)-β have long been lauded as potential candidate genes in fibrosis. PIAS proteins are involved in transcriptional regulation and therefore warrant further investigation in relation to fibrotic disorders.\(^{25}\) In the 15q22.31–q23 region, a potentially important gene is DPP8, the dipeptidyl peptidase IV-related protein DP8-encoding gene, which is known to be involved in ECM interactions and tissue remodelling.\(^{27}\) One candidate gene in the 2q23 locus encodes for the tumour necrosis factor (TNF)-inhibitory protein 6 (TNFAIP6), and is located on chromosome 2q23 at 153 cM (152 Mbp). This gene may have a potential role in wound healing as a hyaluronan-binding domain, which is known to be involved in ECM stability and cell migration. The expression of this gene is shown to be induced by TNF and interleukin (IL)-1. The EGFR gene is located in
the 7p11 region (76 cM; 55 Mbp). Several studies provide evidence for a role for the EGF-signalling pathway in keloid pathogenesis.28 Cell culture studies demonstrated that EGF induces a significantly greater growth response in keloid fibroblasts than in normal fibroblasts. In the 15q22.31–q23 region, DPP8, of which there are four variants, are known to influence ECM interactions and consequently may be important in tissue remodelling. Figure 2 shows a summary of the linkage study results, with the significant chromosome regions highlighted.

**Gene expression studies**

Microarray analysis of gene expression in disease and control samples is a unique yet nonspecific approach to identifying altered regulation of large numbers of genes. However, when used in tandem with more specific techniques such as quantitative reverse transcriptase-polymerase chain reaction (qRT-PCR), it provides a means of rapidly screening tens of thousands of genes to investigate their potential involvement in diseased tissue. Several studies to date have compared gene expression patterns in cultured keloid and normal skin fibroblasts (Table 1).28–41 All these studies have demonstrated differential gene expression in genes involved in fibrosis or fibrotic pathways when comparing keloid tissue and normal tissue-derived fibroblasts. Smith et al.40 used an Affymetrix-based microarray to analyse RNA derived from fibroblasts cultured from ‘fine line’ scars and keloid scars grown in the absence and presence of hydrocortisone. These findings imply that multiple fibrosis-related pathways are involved in the pathogenesis of keloids. Lu et al.42 investigated the expression of cell cycle genes using flow cytometry in fibroblasts originating from central and peripheral regions of the keloid scar. Perhaps not surprisingly, greater levels of activity were observed at the scar margin.

In the study by Seifert et al.,41 an original approach was adopted that not only compared fibroblasts from normal tissue with fibroblasts from keloid tissue, but also investigated fibroblasts originating from different lesional sites within the keloid scar. All samples were screened using Affymetrix technology. In total, 105 genes were differentially regulated, 79 genes upregulated and 26 downregulated in unique gene expression profiles at the different lesion sites.

From an immunological point of view the varied fold upregulation of IGF-binding protein-5 and -7 and the down-regulation of TGF-β1 and TGF-βIII were of interest. However, whether these changes are the consequence of keloid aetiology or a consequence of tissue culture conditions requires further investigation. A direct comparison with gene expression data produced from keloid-derived tissue and normal tissue may provide a different dataset.

Gene expression profiles have also been generated for fibroblasts derived from HS. Dasu et al.30 cultured fibroblasts from HS and investigated their response to IL-6 stimulation by defining their gene expression profiles. Affymetrix gene chip analyses were used to identify up- or downregulation in 12625 genes. An increase of 12 genes in HS fibroblasts was observed when compared with normal skin fibroblasts, while the expression of 14 genes decreased. Thirty-three genes were affected by IL-6 treatment in the HS fibroblasts, with 57 genes affected in normal skin fibroblasts. Ratios of messenger RNA to β-actin for matrix metalloproteinase (MMP)-1 and MMP-3 were increased with IL-6 in normal skin fibroblasts. IL-6 stimulation did not alter MMP expression in HS fibroblasts.
Secreted protein levels of pro-MMP-1 and MMP-3 were elevated in the supernatants from normal skin fibroblasts after treatment with IL-6. No changes were observed in HS fibroblasts treated with IL-6. Therefore the apparent lack of upregulation of MMP-1 and MMP-3 in HS fibroblasts, in response to IL-6, suggests that the suppression of MMPs may play a role in the excessive accumulation of collagen formed in HS. The significance of this study infers that genes encoding cytokines, such as IL-6, suggest that the suppression of MMPs may play a role in keloid disease and hypertrophic scars.

### Table 1

<table>
<thead>
<tr>
<th>Reference (first author)</th>
<th>Year</th>
<th>Study type</th>
<th>Sample type</th>
<th>Passage number</th>
<th>Differentially expressed genes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chen29</td>
<td>2004</td>
<td>cDNA microarray #444 (targets)</td>
<td>Tissue</td>
<td>N/A</td>
<td>402</td>
</tr>
<tr>
<td>Dasu30</td>
<td>2004</td>
<td>Affymetrix GeneChip arrays (HG-U95 A v2, 12.625 genes)</td>
<td>Fibroblast cell culture</td>
<td>4–8</td>
<td>26</td>
</tr>
<tr>
<td>Na31</td>
<td>2004</td>
<td>Human 3k cDNA chip</td>
<td>Tissue</td>
<td>N/A</td>
<td>9</td>
</tr>
<tr>
<td>Naitoh13</td>
<td>2005</td>
<td>HUMAN UniGEM version 2.0 array (9182 cDNA fragments)</td>
<td>Snap-frozen punch biopsies</td>
<td>N/A</td>
<td>32</td>
</tr>
<tr>
<td>Satish28</td>
<td>2006</td>
<td>Affymetrix Human Genome U133a array and RT-PCR</td>
<td>Fibroblast cell culture</td>
<td>&lt; 10</td>
<td>48</td>
</tr>
<tr>
<td>Leppert33a</td>
<td>2006</td>
<td>Affymetrix HG-U133 A&amp;B chips</td>
<td>Tissue</td>
<td>N/A</td>
<td>TGF-β</td>
</tr>
<tr>
<td>Hu34</td>
<td>2006</td>
<td>cDNA microarray (8064 clones of human genes) and RT-PCR</td>
<td>Tissue</td>
<td>N/A</td>
<td>277</td>
</tr>
<tr>
<td>Ghazizadeh35</td>
<td>2007</td>
<td>IL-6 signaling pathway</td>
<td>Fibroblast cell culture</td>
<td>3–4</td>
<td>N/A</td>
</tr>
<tr>
<td>Xia38</td>
<td>2007</td>
<td>Transcriptional activity</td>
<td>Fibroblast cell culture</td>
<td>3–5</td>
<td>Increased transcription of CCN2 after serum stimulation</td>
</tr>
<tr>
<td>Luo37</td>
<td>2007</td>
<td>Human U133A Affymetrix GeneChips</td>
<td>Tissue</td>
<td>N/A</td>
<td>400</td>
</tr>
<tr>
<td>Chu18</td>
<td>2008</td>
<td>Novel truncated TGF-β receptor</td>
<td>Fibroblast cell culture</td>
<td>4–6</td>
<td>N/A</td>
</tr>
<tr>
<td>Sadick39</td>
<td>2008</td>
<td>MMPs</td>
<td>Fibroblast cell culture</td>
<td>N/A</td>
<td>904</td>
</tr>
<tr>
<td>Smith40</td>
<td>2008</td>
<td>Affymetrix Human genome U133 plus 2.0 arrays and RT-PCR</td>
<td>Fibroblast cell culture</td>
<td>N/A</td>
<td>105</td>
</tr>
<tr>
<td>Seifert41</td>
<td>2008</td>
<td>Affymetrix Human genome U133 plus 2.0 arrays and RT-PCR</td>
<td>Fibroblast cell culture</td>
<td>2–4</td>
<td>105</td>
</tr>
</tbody>
</table>

*Comparison of fibroblasts derived from keloid and uterine fibroids. RT-PCR, reverse transcriptase–polymerase chain reaction; N/A, not available; IL, interleukin; TGF, transforming growth factor; MMP, matrix metalloproteinase.

#### Case–control association studies

Several case–control association studies have been conducted with the aim of identifying genes associated with KD; these include genes for TGF-β1, 44, 45 TGF-β2, 46 TGF-β3, 47 and TGF-β8, 48 SMAD3, 6 and 749 (Table 244, 46–51). TGF-β is a multifunctional growth factor involved in the regulation of immune cell function, epithelial cell growth and ECM deposition. Overproduction of TGF-β is associated with excessive deposition of scar tissue and fibrosis. TGF-β signals through a heteromeric receptor complex of type I and II receptor serine/threonine kinases. The signal is propagated downstream through SMADs, a family of evolutionarily conserved intracellular mediators that convey information from the cell membrane into the nucleus. In dermal fibroblasts, TGF-β transduces signals through the p38 mitogen-activated protein kinase (MAPK) pathway to stimulate collagen production. As KD is often characterized by excessive ECM deposition, a great deal of research has focused on TGF-β and TGF-β receptors. However, to date, none of the case–control polymorphism association studies involving TGF-β and TGF-β receptor gene families have demonstrated a statistically significant positive association with KD. TGF-β family members signal via cell surface serine/threonine kinase receptors. Signals from the cell surface ligand are transduced to the cell nucleus by the proteins collectively known as SMADs. 52 A recent case–control single nucleotide polymorphism (SNP) association study...
(183 cases and 121 controls) investigated the potential involvement of 35 SNPs distributed across the genes SMAD3, SMAD6 and SMAD7. Although none of the SMAD gene SNPs investigated were categorically associated with a predisposition to KD, this does not necessarily exclude all possible involvement of these genes or other polymorphisms in these genes from a role in keloid aetiology. Despite the negative findings of the case–control association studies, TGF-β1 isoforms and TGF-β3 receptor genes were less well-defined with regard to the levels of SNPs occurring in these genes.

The potential importance of SMAD3 and its relationship with TGF-β in keloid aetiology has been further demonstrated by Wang et al.51 who used small interfering (si) RNA to examine the function of SMAD3 in keloid fibroblasts. They demonstrated that downregulation of SMAD3 expression can significantly decrease procollagen gene expression and reduce ECM deposition by keloid fibroblasts.

**Immunogenetics of keloid disease**

Currently, the potential importance of immunogenetic factors in the aetiology of abnormal wound healing is receiving increasing attention. Most of the research in this area has focused specifically on the major histocompatibility complex (MHC), also known as the human leucocyte antigen (HLA) system and in particular the class II region of this system. A study investigating the immunophenotypical profiles in both HS and KD confirmed the presence of immune cell infiltrates in keloid scars.54,55 Elevated levels of HLA-DR and CD1a molecules in keloid and hypertrophic tissue were also reported compared with levels detected in normal tissue. Furthermore, investigations into the aetiopathogenesis of HS have implicated a role for MHC molecules, in particular HLA-DR, during wound healing. This may suggest an immunological component of both HS and KD, with particular importance relating to MHC-driven responses.

A positive association between the HLA-DRB1*1501 allele and KD susceptibility has recently been established in Caucasians of northern European ancestry.50 HLA-DRB1*15 phenotype frequencies of Caucasian patients with keloid scars were compared against those observed in a control ethnically matched population (n = 537). The HLA-DRB1*15 phenotype frequency was higher in KD-positive Caucasians (39%) when compared with controls (21%), achieving statistical significance [corrected P (Pc) = 0.017]. These data suggest that in Caucasians of European origin, HLA-DRB1*15 may be associated with the risk of developing KD. Furthermore the positive association of HLA-DQA and DQB alleles with KD susceptibility in the Chinese Han population further supports a potential role for the MHC in KD aetiology.51 In this study, the authors established HLA-DQA1 and DQB1 allele phenotype frequencies among 192 patients with keloids and 273 healthy controls. The frequencies of HLA-DQA1*0104, DQB1*0501 and DQB1*0503 [odds ratio (OR) 2.13, Pc = 0.0063; OR 14.42, Pc < 10⁻⁷ and OR 6.09, Pc < 10⁻⁷, respectively] were significantly higher, while the frequencies of DQA1*0501, DQB1*0201 and DQB1*0402 (OR 0.46, P = 0.0099; OR 0.24, Pc < 0.0001 and OR 0.10, P = 0.0054, respectively) were lower in patients than in controls.

---

**Table 2. Case–control association studies in keloid disease (KD) and hypertrophic scars (HS)**

<table>
<thead>
<tr>
<th>Reference (first author)</th>
<th>Year</th>
<th>Experimental platform(s)</th>
<th>Target gene(s)</th>
<th>Ethnicity</th>
<th>Number of cases</th>
<th>Number of controls</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brown50</td>
<td>2008</td>
<td>PCR-SSOP, Dynal HLA typing kits</td>
<td>HLA-DRB1</td>
<td>Caucasian</td>
<td>67</td>
<td>537</td>
<td>No positive associations</td>
</tr>
<tr>
<td>Lu51</td>
<td>2008</td>
<td>PCR-SSP</td>
<td>HLA-DQA1 and HLA-DQB1</td>
<td>Chinese Han</td>
<td>192</td>
<td>273</td>
<td>No positive associations</td>
</tr>
<tr>
<td>Brown49</td>
<td>2008</td>
<td>Sequenom™ SNP genotyping</td>
<td>SMAD3, 6 and 7</td>
<td>Jamaican Afro-Caribbean</td>
<td>183</td>
<td>121</td>
<td>No positive associations</td>
</tr>
<tr>
<td>Bayat54</td>
<td>2005</td>
<td>PCR and dHPLC mutation detection technology</td>
<td>TGF-β3</td>
<td>Caucasian</td>
<td>95</td>
<td>95</td>
<td>No positive associations</td>
</tr>
<tr>
<td>Bayat46</td>
<td>2004</td>
<td>PCR-RFLP</td>
<td>TGF-β receptors I, II and III SNPs</td>
<td>Caucasian</td>
<td>92</td>
<td>181</td>
<td>No positive associations</td>
</tr>
<tr>
<td>Bayat47</td>
<td>2003</td>
<td>PCR-RFLP</td>
<td>TGF-β1 SNPs</td>
<td>Caucasian</td>
<td>60 (15 HS, 45 KD)</td>
<td>200</td>
<td>No positive associations</td>
</tr>
<tr>
<td>Bayat48</td>
<td>2002</td>
<td>PCR, SSCP, DNA sequencing</td>
<td>TGF-β2</td>
<td>Caucasian</td>
<td>101</td>
<td>187</td>
<td>No positive associations</td>
</tr>
</tbody>
</table>

PCR, polymerase chain reaction; SSOP, sequence-specific oligonucleotide probes; HLA, human leucocyte antigens; SSP, sequence-specific primer; SNP, single nucleotide polymorphism; dHPLC, denaturing high-performance liquid chromatography; SSCP, single-strand conformational polymorphism; RFLP, restriction fragment length polymorphism; TGF, transforming growth factor.
Fig 3. Potential involvement of HLA-DRB1 alleles in keloids. (a) This figure shows a potential role for a keloid-associated major histocompatibility complex (MHC) II bound to an antigen-presenting cell (APC) interacting with a T-cell receptor (TCR) leading to raised dermal scarring as opposed to a normal scar. (b) The potential cascade of events following dermal injury involving keloid-associated MHC II bound to an APC interacting with a TCR in the wound milieu affected by profibrotic cytokines leading to raised dermal scarring. TGF, transforming growth factor; IL, interleukin; ECM, extracellular matrix.

Immunogenetics of hypertrophic scarring

As described above, studies investigating KD and HS demonstrated elevated levels of HLA-DR and CD1a molecules in both conditions. However, unlike KD, where a positive association has been established with the MHC in different ethnic groups,50,51 the potential role of the MHC with HS susceptibility is not well established. It is possible that the same MHC-driven responses potentially involved in KD are also involved to some degree in HS. In view of differences between HS and KD it would be useful to determine the phenotype frequencies of MHC alleles in a cohort of ethnically matched individuals with HS compared with KD cohorts. This would further characterize both conditions and indeed may enable a clearer distinction between the two.

Discussion

KD and HS are both multifaceted aberrations of the normal wound healing process with as yet incompletely understood complex aetiologies. Current data suggest a significant immunogenic component to dermal fibrosis, with MHC genes, cytokines and cytokine antagonists all being implicated in the disease processes. Moreover, there also appears to be a distinct difference in the immunophenotypical profiles of KD, HS and normal tissue, which may help to distinguish more accurately between the two scar types at the histological level. Given the complexities of both KD and HS it is unlikely that a single gene is responsible in either disease. A more likely scenario involves the interaction of one or more genes or gene pathways and/or environmental factors with the culminating effect being the precipitation of dermal fibrosis (Fig. 4). Furthermore, the multifactorial nature of KD and HS is emphasized by the fact that susceptible individuals do not always develop KD or HS following every episode of dermal trauma. Other factors such as skin tension, infection, allergic reactions, excision technique and suture type following surgery can all be contributing elements. Other contributing factors considered to be involved in the development of raised scarring include anatomical location, sex and hormone levels.

KD is believed to be more common in premenopausal women and more severe in susceptible women during pregnancy.50,56,57 Aggressive or morbid keloids, a severe variant of familial KD is more common in darker skinned individuals than in white subjects, further advocating the genetic component of KD.5

KD appears to be a more sustained and aggressive fibrotic disorder than HS. Evidence to date strongly implies a more prolonged inflammatory period with immune cell infiltrate present in the scar tissue of keloids, the consequence of which

Haplotypes DQA1*0104-DQB1*0501 and DQA1*0104-DQB1*0503 proved to be significant susceptibility haplotypes to KD. HLA-DQB1*0501 and DQB1*0503 were positively associated with all subgroups of patients with keloids. In contrast, DQA1*0104 (OR 2.51, \( P = 0.0099 \)) and DQB1*0402 (OR 2.22, \( P = 0.0090 \)) and DQB1*0503 (OR 2.14, \( P = 0.0117 \)) were prevalent only in patients with keloids with single site, moderate severity and negative family history. HLA-DQB1*0201 (OR 0.27, \( P = 0.0018 \)) and DQB1*0402 (OR 0.07, \( P = 0.0270 \)) and DQB1*0401 (OR 0.07, \( P = 0.0306 \)) were negatively associated with moderate severity and negative family history of keloids; moreover, HLA-DQB1*0201 (OR 0.23, \( P = 0.0003 \)) and DQA1*0501 (OR 0.43, \( P = 0.0234 \)) were less prevalent in patients with single-site lesions. This study clearly demonstrated the positive association of HLA-DQA1 and DQB1 alleles and haplotypes with susceptibility to KD. The data generated in both of these studies support a strong immunogenic component to KD although the precise mechanisms involved in MHC-driven abnormal scarring require further investigation (Fig 3a,b). Furthermore, the MHC is an extremely 'gene dense' region located on the short arm of chromosome 6p21.3. Strong linkage disequilibrium exists across the MHC making it difficult to pinpoint the exact causative gene or genes.

Other immunohistological studies in keloid and hypertrophic scars

An interesting observation with a clear distinction between both types of scarring was provided by the findings of Santucci et al.,55 who investigated the immunophenotypic profiles of keloid and hypertrophic scar tissue. The immunohistochemical analysis of paraffin-embedded sections revealed an immune cell infiltrate composed of CD3+, CD45RO+ and CD4+ T lymphocytes associated with CD3+, CD45RO+ and CD1a+ dendritic cells in the dermis of both HS and KD. A significant amount of cellular infiltrate was observed in young HS lesions. The amount of the cellular infiltrate had progressively decreased in fully developed HS and in old HS, where very low quantities of immune cells were found. Conversely, in keloids, a large amount of the cellular infiltrate was observed both in young and in old lesions; the number of immune cells was constantly higher than that observed in all types of HS. This suggests that the continuous presence of an immune cell infiltrate, with the probable effect of prolonged inflammation, cell recruitment to the wound site and ECM deposition, may help explain the 'continued growth' of keloid lesions beyond the margins of the original wound which is not, by definition, observed with HS. The exact immunogenetic mechanisms involved however remain unclear.
Keloid-associated MHC II heterodimer
Antigenic peptide
TCR

APC

Raised scar
Normal scar
e.g. keloid scar

(a)

Keloid-associated MHC II heterodimer
Antigenic peptide
TCR

Prolonged immune response
Prolonged inflammation

Excessive ECM deposition
(type I collagen)

Pro-fibrotic cytokines
e.g. IL-13

Fibroblasts

Fibroblast differentiation (myofibroblasts), proliferation and migration

ECM = Extracellular matrix
GF’s = Growth factors
APC = Antigen presenting cell
TCR = T cell receptor
CD4+ T cell = CD4-positive T cell
CD4+ TH2 cell = CD4-positive T-helper 2 cell

(b)
may contribute to increased fibroblast activity with greater and more sustained ECM deposition (Fig 3a,b). This in turn may help to explain why keloid scars spread beyond the margins of the original wound, while HS, in which the immune cell infiltrate decreases over time, remain within the original wound margins and often regress over time. Moreover, recent data suggest the positive association of MHC II alleles with a predisposition to the development of KD following dermal trauma. MHC alleles may provide clinicians with an accurate indication of an individual’s risk of raised scarring following dermal trauma if the individual is positive for a specific MHC allele or MHC haplotype. Although the MHC allele or haplotype is unlikely to be the sole cause of raised scarring its potential predictive value for the development of raised scarring following dermal trauma cannot be ignored.

Gene expression studies have provided further insights into the potential involvement of candidate genes. However, most of the data generated are based on fibroblast cell cultures with inherent problems of extrapolating data relevant to in vivo biological and genetic events occurring in the wound. The problems with standard cell culture techniques are several fold but undoubtedly the major drawbacks are the dissimilarity to the actual environment of the dermis. Consequently gene expression data derived from normal tissue. Such an animal model were to be developed. Furthermore, the use of organ culture as well as organotypic or three-dimensional tissue cultures may also provide more representative gene expression data when comparing fibroblast and/or keratinocyte cell cultures derived from KD or HS with those derived from normal tissue.

Future research into aberrant wound healing will also include defining the potential role of hormonal effects on dermal scarring, the importance of which is often emphasized by the increased severity of KD and HS during pregnancy and increased incidence post-puberty and in adolescence. In addition, being able to reduce wound tension during wound healing may also be of benefit as it is well established in the literature that increased wound tension has a negative effect on the normal wound healing process.

A better understanding of genetic factors responsible for raised skin scarring may enable improved pre-, peri- and post-operative management of cutaneous scars. Being able to diagnose high-risk individuals, coupled with the ability to determine more accurately the prognosis and the most appropriate or successful treatment strategy based on potential response will have beneficial consequences not only for the clinician and patient, but also in terms of reducing the financial burdens placed upon healthcare systems.

In conclusion, KD and HS are multifaceted aberrations of the healing process with as yet incompletely understood aetiologies. Current data suggest a genetic susceptibility with a strong immunogenic component to dermal fibrosis with MHC genes being implicated. The ability to assess accurately an individual’s potential genetic susceptibility to raised scarring may lead to a more personalized approach to their management in the future.

References

Genetic susceptibility to raised dermal scarring, J.J. Brown and A. Bayat

17

48 Bayat A, Bock O, Mrowietz U et al. Genetic susceptibility to keloid disease: transforming growth factor beta receptor gene

© 2009 The Authors
Journal Compilation © 2009 British Association of Dermatologists • British Journal of Dermatology 2009 161, pp8–18


