Review paper

Current views on the etiopathogenesis of keloids

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Abstract

Keloids are benign mesenchymal tumours, which form post injury as a result of an abnormal wound healing process in genetically susceptible individuals. Clinically they are often confused with hypertrophic scars but in contrast to them, keloids grow beyond the margin of the original wound, invade the surrounding tissue, show no tendency to resolve spontaneously and recur after surgical excision. This kind of disorders occurs only in humans and more often in dark-skinned individuals. The pathogenesis of keloids remains still not completely understood but there are a lot of hypotheses explaining a complicated process of keloids formation: genetic susceptibility, immune function disorder, disturbances in cell proliferation and cell death cycle, altered response to the growth factors and some others. These hypotheses are not alternatives, each one describes another level of the two major processes involved in keloid pathogenesis: hyperproliferation of fibroblasts and excessive production, and deposition of extracellular matrix (ECM) proteins. Further investigations are required for better understanding of the pathogenesis of keloids and should result in more effective treatment of this frustrating clinical problem of wound healing.

Key words: keloids, pathogenesis, fibroblasts, collagen, extracellular matrix proteins, growth factors, receptors, proliferation, apoptosis.

Introduction

Keloids are non-malignant mesenchymal tumours which develop in genetically susceptible individuals as a result of various injuries such as scalds, surgical excisions, bites, injections, stings or as complications in severe forms of acne. They are results of disorders in a wound healing process. Such disorders happen on many levels: genetic, molecular and immunological [1-4].

Many clinicians regard the words "keloids" and "hypertrophic scars" as synonyms. However, despite apparent similarity, these two are different diseases and considering them synonymous is a serious factual error [5].

Keloids, unlike hypertrophic scars, are more extensive than the area of post-traumatic wound; they infiltrate the surrounding tissue and form characteristic processes in the peripheral area. They do not tend to regress spontaneously and they recur after being surgically removed [1-6].

The earliest information about keloids comes from the ancient Egypt. The term "keloid" was mentioned for the first time by Alibert at the beginning of the 19th century. The word "chele" comes from the Greek language and means "crab's pincers" [7].

Epidemiology, clinical and histopathological image

Keloids can be observed only in humans. No other mammal has ever developed similar lesions. Most often they occur in dark-skinned individuals, in 4-16% of the population [1-4]. Keloids appear most frequently between the second and third decade of life. Infants do not develop them at all and keloids hardly ever appear in old people. No differences were observed in the prevalence of keloids in people of both sexes [1-8]. The clinical image presents vivid red or even brown tumour-like lesions. Their density is much higher than that of the surrounding skin since they contain mostly collagen fibres. The lesions are clearly visible and in the peripheral area there are characteristic processes [9, 10]. The patient with keloids suffers from slight burning or itching sensations.

The lesions are mostly located on the chest in the sternum area, back, arms, earlobes and the neck. Hardly ever can they be found on eyelids, reproductive organs, palms or feet. Keloids which occur in the chest area can often have the shape of a butterfly and those which affect earlobes resemble tuberous growths [11, 12]. Extensive keloids located close to big joints might cause various contractures and deformities. In extreme cases they can even

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impair regular motor activity (especially hypertrophic scars which develop after scalds) [13].

Keloids range in size from a few millimetres to dozens of centimetres and there is no relationship between the size of the keloid and the force which caused the injury. It means that a slight injury might lead to an extensive keloid.

Keloids can often be a serious mental problem for the patient. Those who have developed this disease frequently suffer from depression, have low self-esteem or even personality disorders [8-13]. Histopathology of keloids demonstrates proliferation of fibroblasts and among them increased accumulation of extracellular matrix (ECM) proteins. In them, the dominant protein is collagen which has thick, compact fibres and is characterized with the irregular course. Out of many types of collagen which is found in extracellular matrix, type I collagen is dominant. Type III collagen is the second most frequent collagen. The difference between the amount of collagen of type I and type III is most visible in keloids when we consider reactive scars and hypertrophic scars [1, 2, 7].

Table 1 presents the most important distinguishing features of keloids and hypertrophic scars.

Physiology of wound healing

Everybody in his life is exposed to many various injuries. If the epidermis has been injured only, the wound heals without leaving any scars. If the injury is deeper and the integrity of the basement membrane has been broken, the dermis, or more precisely, fibroblasts, which are most numerous in that layer, take part in the healing process [1-4, 7-13].

The healing process consists of a few phases: haemostatic, inflammatory, proliferative and remodelling of the primary scar. In the haemostatic phase, platelets and coagulation factors play the main role. They are involved in bleeding arrest. At that stage, a lot of mediators are secreted, e.g. platelet derived growth factor (PDGF) or transforming growth factor β (TGF-β). A local inflammatory reaction takes place. In this phase extravasated blood and tissue debris are removed and the wound fills with inflammatory cells: macrophages, lymphocytes as well as fibroblasts. A lot of pro-inflammatory cytokines are released, too. Between the third day and the third week following the injury, connective tissue, so called granulation, starts growing. It is composed of rapidly proliferating fibroblasts and myofibroblasts, cells which synthesize and secrete ECM proteins, especially collagen, but also proteoglycans, hyaluronic acid and fibronectin. Initially, the proliferation of fibroblasts is more rapid than their apoptosis and the production of extracellular matrix proteins outdoes the degradation process of these proteins. Between the 4th and the 6th week, a dynamic balance between these two opposing processes starts to be established; in the course of time, cell apoptosis and collagen resorption, so called remodelling, occurs. It is aimed at reducing scar mass to the required minimal value [1-4, 7-14].

Healing wounds is a multi-stage process. A lot of cells and cell mediators are involved in the process. In each of its phases there is some balance between the opposing processes. Certain processes prevail in one phase; later on – they may regress.

The final scar, being a physiological response of the body to an injury, appears only when the mechanism of the healing process remains undisturbed. Should the balance be disrupted in any of these phases, some pathological disorders occur. Keloids are examples of such abnormalities [1-4, 7-14].

No theory explaining the pathogenesis of keloids has been presented to date. However, researchers analysed findings of genetic, molecular and cell studies and established a few hypotheses. Each hypothesis refers to a different stage of keloidogenesis. Thanks to that fact, these theories complement each other.

Table 1. Main differences between keloids and hypertrophic scars

Keloids	Hypertrophic scars
Infiltrate the surrounding tissue	Do not infiltrate the surrounding area
Recur following excision	Do not recur
Grow beyond the area of wound	Do not grow beyond the area of wound
Type I collagen dominates	Type III collagen dominates
The course of collagen fibres – chaotic	The course of collagen fibres – regular

Genetic conditions of keloid prevalence

Significantly frequent occurrence in black people (1/30 of the Afro-American population is affected by keloids; for the whole American population – the number is 1/625) and positive anamnesis in people in whom the disease was observed prove the presence of genetic predispositions [10-19].

It was also proved that occurrence of keloids is related to A blood type and the following tissue compatibility antigens: HLA B14, Bw16, B21, Bw35, DR5, DRB1*, DQA1, DQB1 [20-23]. Keloids are often concomitant with a lot of common genetic diseases of connective tissue. They include scleroderma, progeria, Ehlers-Danlos syndrome or Rubinstein-Tabi syndrome [24].

So far no particular gene responsible for pathogenesis of keloids has been identified. Also the way in which the disease is inherited is unknown. Researchers claim that a polygenic model is the most probable. Moreover, genes condition only susceptibility to keloid development;

the requisite factor is always an injury, even the slightest and unnoticeable.

Thanks to analyses of genetic polymorphisms, researchers managed to identify chromosome *loci* exhibiting a distinctive positive correlation with occurrence of keloids. The *loci* are: 2q23, 7p11 and 14q22-q23 [25-27]. Difficulty in identification of genes responsible for susceptibility to keloid development stems from the lack of the animal model of the disease.

Thanks to comparative analysis of gene expression in fibroblasts of keloids and reactive scars, it was possible to show significant differences in 500 genes responsible for various metabolic and cellular pathways. The study is not aimed at presenting all differences; below there is a presentation of most important groups of candidate genes which probably contribute to keloid development.

Out of genes which are characterized by excessive expression in fibroblasts of keloids, genes for insulin-like growth factor binding protein (IGFBP): 2, 5, 7 and connective tissue growth factor (CTGF) show the most distinct differences [15-18].

It is worth mentioning that the analysis of gene expression was performed in the culture imitating the natural human body as well as in the culture containing appropriate high concentration level of hydrocortisone, which is a known factor inhibiting a wound healing process and counteracting a process of keloid formation.

Adding hydrocortisone led to an increase in gene expression for IGFBP 3 and its intracellular mediator – STAT-1 (in neutral medium, gene expression did not demonstrate any differences in comparison with the expression of fibroblasts of physiological scar) whereas the expression of the remaining IGFBP genes was decreased, which implies the role of IGFBP 3 and STAT-1 genes in keloid resistance to steroids. Apart from the increased expression of proteins, binding insulin-like growth factors in keloid fibroblasts, also genes for nerve growth factor β (NGF-β) and for cyclin D2 exhibited excessive activation [15-19]. Excessive expression was also observed in encoding receptor genes for thrombin and thrombin-like growth factor (F2R and F2RL2 respectively) as well as for TGF-β receptor I [15-19, 28].

A number of genes encoding proteins which are second messengers in intracellular process of signal transmission regulating cell growth and cell differentiation also exhibit excessive expression in keloids. They include dishevelled-associated activator of morphogenesis 1 – activator from Wnt protein family (DAAM1), JAG-1, encoding second messenger of Notch signalling pathway as well as nucleus factor of transcription (NF1B) [15-19, 28].

Another group of genes which exhibit excessive expression in keloids are genes which encode components of extracellular matrix COL1A1 gene encoding type I collagen, ELN gene encoding elastin (only in the medium containing hydrocortisone) as well as genes whose profile clearly resembles osteochondrogenesis. They include genes encoding collagen types: V, VI, X, XV and XVI and also periostin, thrombospondin 4, lumican, mimecan, versican, syndecan-2 and decorin [29, 30].

There is one more gene which demonstrates excessive expression in keloid fibroblasts. It is gene for 311 protein. Its function has not yet been fully discovered. However, researchers suggest it might be an oncogene. Neoplasms of the central nervous system – astrocytomas also show an excessive expression of this protein. The oncogene is probably a factor determining the invasive character of the neoplasm [15-17].

A number of genes show a significantly decreased expression in keloid fibroblasts rather than in fibroblasts of physiological reactive keloid. The most important genes demonstrating a decreased expression in keloids are genes for inhibitors in intracellular signalling pathway from the abovementioned Wnt protein family (DKK1, DKK3 and SFRP1 and 2), as well as for inhibitors in signalling pathway of cyclins (CDKN1C, CDKN3).

Another group of genes which exhibit a decreased expression in keloids includes: interleukin 1 receptor gene, (IL1R) and genes controlled by interleukin 1, encoding chemokines and their ligands (CXCL1, CXCL12 and CXCL14). A decreased expression was also observed for genes encoding IL-7 and IL-8 [15-19].

An extremely decreased expression was also noted for genes encoding metalloproteinases, i.e. enzymes which take part in degradation of extracellular matrix proteins – matrix metalloproteinase 3 (MMP3) and membrane metalloendopeptidase (MME).

Unlike the gene encoding receptor for TGF-β type I, receptor II encoding gene for the growth factor exhibits a significantly decreased expression in keloids in comparison with ordinary fibroblasts [15-19, 28, 31].

The last group of genes which demonstrate a significant decrease in expression in keloids is a semi-conservative family of HOX genes. Genes of this family regulate proper differentiation of cells creating germ layers at the early stage of embryogenesis. A number of these genes participate in the regulation of fibroblast proliferation in adult life. It is known that specific "silencing" of some of these genes contributes to development of lung cancer, adenocarcinoma type. Keloids demonstrate cancerous character probably due to insufficient expression of these genes [15-19, 28].

Tables 2 and 3 list the most important genes whose expression in keloids is excessive and decreased as well as present their general characteristics and chromosome loci.

Abnormal response to inflammatory mediators

It has already been mentioned that in the physiological course of wound healing many inflammatory mediators are secreted (growth factors, cytokines, interleukins). As for keloids, many of these substances are character-

Table 2. Genes showing an increased expression in fibroblasts derived from keloid, their function and chromosomal location

Gene	Function	*Loci*
F2R	Thrombine receptor, blood clotting	5q13
F2RL2	Thrombine-like factor receptor	5q13
CD2	Cycline D2, proliferation regulator	12p13
DAAM1	Second messenger of WNT family	14q22-q23
FHL1	Differentiation regulator	Xq27
IGFBP2	Cellular growth regulator	2q33-q34
IGFBP5	Cellular growth regulator	2q33-q34
IGFBP7	Cellular growth regulator	4q12
NGF-β	Growth factor, intercellular signaling	1p13
MEST	Regulator of mesoderm development	7q32
STAT1	Regulator of transcription	2q32
SIX1	Regulator of transcription	14q23
JAG1	Second messenger of Notch family	20p12

Table 3. Genes showing a decreased expression in fibroblasts derived from keloid, their function and chromosomal location

Gene	Function	*Loci*
ILR1	Interleukin 1 receptor	2q12
IL-7	Interleukin 7, immune reactions	8q12-q13
IL-8	Interleukin 8, proliferation, chemotaxis	4q13-q21
MMP3	Collagen degradation	11q22.3
MME	Proteolysis and peptidolysis	3q25.1-q25.2
SFPR1	Second messenger of WNT family, inhibitor	8p12-p11.1
TGF-βRII	Receptor for TGF-β	3p22
TNC	Tenascin, cell adhesion	9q33
SDC1	Syndecan, cytoskelatal protein	2p24.1
DKK1	Second messenger of WNT family, inhibitor	10q11.2
DKK3	Second messenger of WNT family, inhibitor	11q15.2
CXCL1	Chemokine, inflammation and proliferation regulation	4q21
CXCL12	Chemokine, inflammation and proliferation regulation	10q11.1
CXCL14	Chemokine, inflammation and proliferation regulation	
CDKN1C	Cell proliferation inhibitor	11p15.3
CDKN3	Cell proliferation inhibitor	14q22

ized by much higher concentrations, greater activity as well as they remain active for a longer period of time than if they remained in regular conditions.

Numerous studies proved that keloid fibroblast differently responds to physiological concentration of a particular mediator than a regular cell, which results from molecular changes, starting from a receptor on the surface of cell membrane of keloid fibroblast, second signal messengers to nucleus receptor and transcriptional factors. All these changes result in excessive growth and proliferation of fibroblasts as well as greater production and accumulation of ECM proteins [1-4, 7-13].

Keloid tissues exhibited a significantly higher concentration of the following growth factors: platelet derived growth factor (PDGF), connective tissue growth factor (CTGF) or, mentioned in the previous chapter, insulin growth factors [7-13].

It is currently believed that the family of TGF-β proteins hold the supreme position in pathogenesis of keloids. The family consists of TGF-β isoforms and related mediators. So far three isoforms of TGF-β factors, named TGF-β1, TGF-β2 and TGF-β3 have been described. These three types are distinguished from each other with single amino acids in the peptide chain. However, these changes lead to different activity of particular isoforms [32]. Some independent histochemical studies proved that in keloid tissues, mRNA level for TGF-β1 and TGF-β2 is much higher than in a physiological scar. Completely unlike to TGF-β3, where the level is significantly lower in keloids than in hypertrophic or reactive scars. Interpretation of these results appears to be obvious, after presenting the function of TGF-β3 isoform, demonstrating a decreased expression in keloids. Contrary to the other isoforms, TGF-β3 turns out to possess antifibrotic properties, inhibit fibrosis processes, which are determined by accumulation of extracellular matrix, especially collagen [31-34].

In a physiological course of wound healing, TGF-β3 is a natural inhibitor of collagen accumulation and stands in opposition to TGF-β1 and TGF-β2. It prevents fibrous tissue of a scar from excessive proliferation and provides proper remodelling. In keloid formation process, the balance is clearly disturbed as fibrosis prevails. The TGF-β1 and TGF-β2 which act profibrotically, show an increased concentration and TGF-β3-decreased concentration.

So far this fact has not been explained on a genetic level. Some attempts to identify particular polymorphisms in TGF-β gene were made but no significant differences were observed [31-34].

Dimeric proteins called activins are also related mediators of TGF-β. They mainly stimulate the growth of differentiation of various cell types [28]. There are activin A (composed of two sub-units βa), activin B (composed of two sub-units βb) and activin AB (composed of subunit βa and βb).

Hubner *et al.* proved that in the process of wound healing within the first 24 h the concentration level of activin A increases. In transgenic experiments conducted on mice in which overexpression of gene for activin A was initiated, a significant proliferation of fibroblasts and increased production of collagen were observed. As a result, the mice's skin got thicker.

Other studies showed that activin A is involved in pathogenesis of diseases in which excessive fibrosis is a key process: hepatic cirrhosis, interstitial pulmonary fibrosis and also renal fibrosis [35].

As for keloids, it was noticed that the concentration of activin A is more than 20 times higher than in a physiological reactive scar. Activin A has its own natural inhibitor – follistatin. Its high concentration level was observed in keratinocytes of the basal layer of the epidermis in the keloid area (4 times higher than in a physiological reactive scar). It is suggested that activin A and follistatin should be considered jointly as feedback loop, in which, in the case of keloids, activin A is prevalent [35]. Also on TGF-β receptor level, there are clear differences in keloids and hypertrophic and reactive scars. Similarly to TGF-β, which has a few isoforms, also a receptor for this mediator has its own types. So far, two of them have been described and they are known as TGF-βIR and TGF-βIIR. Both of the receptors are transmembrane proteins which exhibit the activity of serine – threonine kinase. Type II receptor takes part in direct binding of ligand. Such ligand-receptor complex binds type I receptor and activates it in the process of phosphorylation and the latter receptor activates second messengers [36]. When comparing keloids with hypertrophic and physiological scars, an increased expression for TGF-βIR and decreased expression for TGF-βIIR were observed in keloids. It was proved that the ratio of TGF-βIR to TGF-βIIR conditions the final effect induced by TGF-β. A higher level of TGF-βIR than TGF-βIIR promotes intensive fibrosis, which happens in keloids [36, 37].

Second messengers of signalling pathway TGF-β are among other SMAD proteins. SMAD proteins 2, 3 and 4 activate each other in a cascade way, amplifying in this way the activity of TGF-β. SMAD proteins 6 and 7 are inhibitors of this signalling pathway. They are aimed at inhibiting the process initiated by TGF-β. Histochemical studies showed that also on that level of TGF-β signalling pathway there might be disorders. The amount of SMAD proteins 6 and 7 is considerably decreased in keloids, therefore proteins activating SMADs 2, 3 and 4 are prevalent. The TGF-β signalling path in keloids is the evidence that the balance in cell signalling is broken on all its levels.

In keloidogenesis, on the one hand, there is an excessive expression of TGF-β1, TGF-β2, TGF-βIR receptor and SMAD proteins 2, 3 and 4, and on the other hand, a decreased expression of TGF-β3, TGF-βIIR receptor and SMAD proteins 6 and 7, which highly promotes proliferation of fibroblasts and fibrosis [36-41].

Vascular endothelial growth factor (VEGF) is another factor whose concentration in keloids is increased. The factor is mainly supposed to stimulate neoangiogenesis, i.e. the process of forming new capillary vessels. An excessive expression of this mediator was observed in neoplastic processes in the large intestine as well as in the exudative form of age-related macular degeneration (AMD) [42].

Keloid keratinocytes synthesize and secrete much more of this factor than in the regular course of tissue repair. This fact is confirmed by an experiment in which keloid keratinocytes were cultured with fibroblasts of healthy skin. A few days later, excessive proliferation of these fibroblasts and primordium of capillary vessels were noticed. Simultaneously, ordinary keratinocytes were cultured with the same population of fibroblasts. Then, similar results were not observed.

Staining keloid tissue with immunohistochemical methods resulted in a considerable increase in VEGF expression in the basal layer of epidermis [42, 43].

One of the most basic inflammatory cytokines, IL-6, participates in pathogenesis of keloids. Apart from promoting inflammatory processes it also actively stimulates proliferation of fibroblasts. In regular conditions, IL-6 favours secretion of metalloproteinases, whereas in keloid fibroblasts, this process does not occur (the role of metalloproteinases has been described in the chapter on genetic conditions of keloids). Interleukin 6 increases the expression of type I collagen and fibronectin. This was proved in an experiment in which adding exogenous IL-6 to the culture of fibroblasts resulted in an increase in mRNA expression for COL1A and FN1 genes. The opposite result was obtained after adding inhibitor for IL-6 [44].

Immunohistochemical studies on keloids showed an increase in expression of a receptor protein for IL-6 (IL-6αR) and proteins which are second messengers in the signalling pathway of this cytokine: JAK1, STAT3, RAF, ELK and gp130 [44].

Macrophage ihibiting factor (MIF) is a factor produced by tissue macrophages, activating lymphocytes. In keloids its concentration level is significantly higher. It is one of very few cytokines, whose expression is enhanced by glucocorticosteroids; therefore, it influences keloid resistance to anti-inflammatory and immunosuppressive drugs [45]. Macrophage ihibiting factor is a cytokine which demonstrates pleiotropic activity, stimulates angiogenesis, is a tumour proliferative factor and is involved in metastasis of malignant neoplasms. Macrophage ihibiting factor activates two enzymes: cPLA2 phospholipase and COX-2 cyclooxygenase. The first enzyme is involved in the release of arachidonic acid from the surface of membrane; the acid, in turn, is a key substrate for the other enzyme which catalyzes its transformation into metabolites of leukotriene (LTC4) or prostanoid (the most representative is prostaglandin 2 – PGE_2) pathways [45].

As for keloid fibroblasts, it was proved that MIF concentration activates cPLA2 more than ordinary fibroblasts. It results in an increased supply of arachidonic acid. To the contrary, the other enzyme – COX-2 – is less activated in keloid fibroblasts, so despite a greater amount of arachidonic acid there are less products. One of products of COX-2 enzyme is the above-mentioned prostaglandin PGE_2, which is the main mediator preventing cell proliferation (including fibroblasts) and inhibiting synthesis of collagen. Prostaglandin E_2 is thus a factor inhibiting the process of keloid formation and an example of negative feedback loop in tissue repair. Macrophage ihibiting factor, as a factor promoting fibroblast proliferation and fibrosis, stimulates simultaneously PGE_2, which inhibits this process. In keloids, release of PGE_2 is considerably decreased because of the decreased activity of COX-2. Histochemical studies also showed that an expression of the membrane receptor through which prostaglandin acts is also significantly decreased in keloid fibroblasts.

It is thus another example of imbalance between processes stimulating keloidogenesis and inhibiting it [45, 46].

Immune disorders in keloid formation

Apart from genetically determined factors and disorders in response to growth factors, immune disorders also contribute to formation of keloids.

Many researchers proved the relation between unusually high concentration levels of IgG and IgM immunoglobulins and proteins of the complementary system. Oluwasami and Cohen described deposits of IgG class located among collagen fibres in keloids and Kirchner confirmed these observations and proved the presence of IgA and IgM immunoglobulins in keloid tissue [7-9, 47-49]. In 1982, Jansen and Limpens used the immunofluorescence method and stated that patients with keloids have antinuclear antibodies directed against their own fibroblasts (so called anti-fibroblast antibody – AFA). These antibodies appear in the proliferative phase. Then, their concentration level rapidly drops. Many researchers imply that the antibodies stimulate fibroblasts against which they are directed to proliferate and produce collagen [47-50].

Another group of researchers expresses the opposite opinion. They claim that the presence of AFA is a result, not a cause, of forming keloids, a body response to excessive production of collagen and fibroblast proliferation. Current knowledge makes it impossible to solve the controversy but the presence of antibodies directed against their own fibroblasts in keloids makes many researchers classify keloid as an autoimmune disease of connective tissue, similar to systemic scleroderma. A mutual relationship between these two diseases was mentioned in the chapter on genetic conditions of keloids [15-17, 47-50].

Another hypothesis is improper reaction of the immune system to sebum, which is a natural, secreted product of sebaceous glands. The hypothesis seems to be justified by the fact that both keloids and sebaceous glands are located in the same areas of human skin.

According to this theory, a mechanical injury results in breaking skin integrity and deeply located tissues as well as destruction of the sebaceous gland. As a consequence, the content of the sebaceous gland gets to local circulation, which in turn, induces delayed type of cell-immunity reaction [51]. The size of the keloid is bigger than the initial place of the wound and thus presses the surrounding tissue. More sebaceous glands are getting involved in the process. They are destroyed due to pressure of keloid tissue. In that way the process is being maintained and the keloid diameter is growing. Hypersensitivity of patients to sebum was confirmed with intradermal injections with the patients' own sebum. This test allows for making an observation that delayed cell response plays a great role in pathogenesis of keloids [51, 52].

Keloids: the effect of imbalance between proliferation and apoptosis of fibroblasts

In the part of the article on the physiological course of healing wounds, it was mentioned that a dynamic balance between opposing biochemical and cell processes, e.g. proliferation of fibroblasts and their programmed death, i.e. apoptosis must be constantly maintained [7-9].

It should be stressed here that keloid fibroblasts do not make up a homogeneous group, but depending on the location (central or peripheral), they differ substantially in terms of activity, rate of nucleus division, response to growth factors and production of ECM proteins.

In the centre of keloid, mesenchymal framework and single fibroblasts dominate. As for the peripheral area of keloid, active and rapidly proliferating fibroblasts dominate. In experiments in which fibroblasts from the peripheral area of keloid were compared to fibroblasts of healthy skin, it was calculated that the time needed to double the initial number of cells in keloid fibroblasts was about 26 h, whereas for physiological fibroblasts, the time was 43 h. Besides, the number of apoptotic cells in fibroblasts of healthy skin is almost twice as big as the number of apoptotic cells in fibroblasts from the peripheral area of keloid. Immunohistochemical studies showed increased concentration levels of inhibitors for caspases (proteolytic enzymes in the apoptotic pathway) in the peripheral area of keloid in comparison to its centre. It means that keloid fibroblasts located in the peripheral area of keloid are metabolically more active. They divide more rapidly and do not undergo apoptosis to such a great extent. Due to this fact, it can be said that this population of keloid fibroblasts is similar to neoplastic cells, which proliferate in an uncontrolled manner and are immortal.

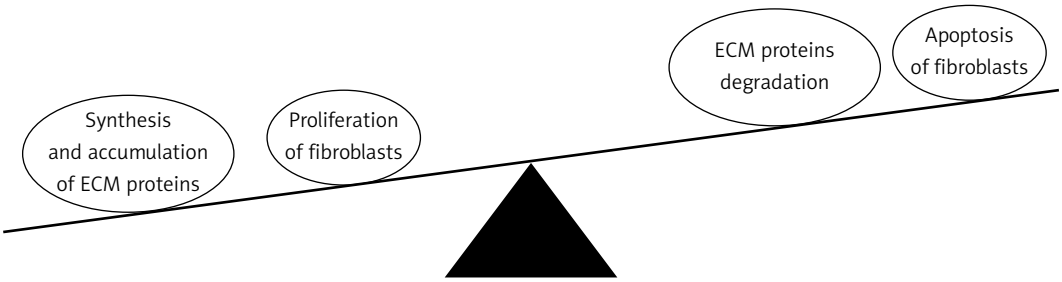

Fig. 1. The pathogenesis of keloids: an imbalance between synthesis and accumulation of extracellular matrix proteins and their degradation, and between proliferation and apoptosis of fibroblasts

Moreover, fibroblasts located in the centre of keloid are characterized by increased expression of ADAMs gene which is responsible for apoptosis [53-55].

This fact proves the existence of imbalance between proliferation of fibroblasts and their apoptosis in the peripheral area of keloid but proliferation is the prevailing process. In the process of keloid formation, various subpopulations of fibroblasts are involved. These are more metabolically active, more proliferative and more "resistant" to apoptosis and they migrate to the peripheral area. The centre of keloid is occupied by the population which is more prone to undergo apoptosis and divide much more slowly. After some time, the centre of keloid becomes hypocellular as most of the fibroblasts atrophy leaving behind mesenchymal framework. The population of fibroblasts from the peripheral area constantly divide and migrate to the periphery, making the area of keloid bigger in size.

The above facts let us understand why mature keloids are atrophic in the centre and elevated and pigmented in the periphery [53-56]. Figure 1 presents the imbalance between proliferation and apoptosis as well as the synthesis of degradation of extracellular matrix in pathogenesis of keloids.

"Tension" hypothesis of forming keloids

According to this hypothesis, keloids are formed significantly more often when the skin is damaged in the place where it is most tense, i.e. above big joints, in the chest area. The hypothesis is an excellent explanation why keloids do not develop in old people (it is a result of lack of skin tension). However, there are places which are often affected by keloid formations but the skin tension is in such places minimal, e.g. on earlobes [57].

A great skin tension considerably hampers the process of wound healing since the wound edges do not fit tight enough. Collagen fibres have a chaotic, irregular course – characteristic of keloids. An increased skin tension is only an additional factor and favours development of both keloids and hypertrophic scars whose formation mechanisms are completely different [57, 58].

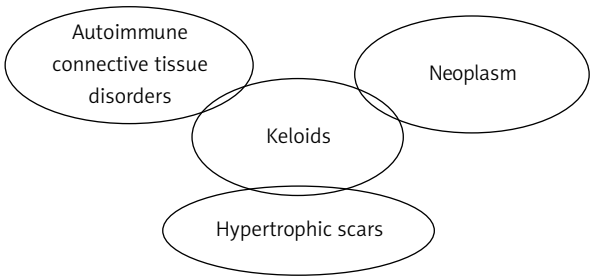

Fig. 2. Classification difficulties with keloids

Summary

Keloids occur as a result of disorders in a wound healing process. Such disorders are observed on genetic, cell and molecular levels. The fact that keloids are constantly growing and are locally invasive makes them different from hypertrophic scars and let us classify them as non-malignant tumours. A number of genes demonstrate a different expression in keloid fibroblasts in comparison with physiological fibroblasts. It is known that these genes determine susceptibility to keloid development. When such people suffer from an injury in which skin integrity is broken, tissue repair is disordered. Proliferation of fibroblasts and production of ECM proteins dominate [1-5, 7-9, 59].

Presence of antibodies directed against fibroblasts (AFA) and the relationship with certain diseases, where also antibodies are produced and excessive fibrosis occurs, help us treat this disease as an autoimmune disorder of connective tissue [60]. Figure 2 presents a complex and still unknown classification of keloids.

Keloids are a serious and frustrating clinical problem, especially in dermatology and cosmetic surgery. Effective methods of treatment and prevention of keloids have not

been identified yet. Thus, further studies on their pathogenesis are entirely justified.

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