Original paper

Analysis of the correlation between the levels of HIF-1α and miR-199a in lesions and the psoriasis severity index

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Abstract

Introduction: The pathogenesis of psoriasis is complex. Previous bioinformatics studies have found differential expression of HIF-1α and miR-199 in psoriasis, but the correlation between miR-199a and HIF-1α and the severity of psoriasis is still unclear. This study found differential expression of HIF-1α and miR-199a levels in the skin lesions of patients with different degrees of psoriasis. HIF-1α mRNA and miR-199a are PASI influencing factors, and the levels of HIF-1 α and miR-199a in the skin lesions can serve as potential indicators for evaluating the severity index of psoriasis.

Aim: To examine the relationships between the levels of HIF-1α and miR-199a and psoriasis severity.

Material and methods: Sixty-five patients with psoriasis vulgaris treated from January 2023 to May 2024 were divided into a mild group (n = 16), a moderate group (n = 23), and a severe group (n = 26) according to the lesion area and severity index (PASI) score, and 40 healthy people were included in the control group (group A). The expression of HIF-1a and miR-199a in psoriatic lesions and normal skin tissues was detected by fluorescence quantitative PCR.

Results: The level of miR-199a in lesions in the observation group (group B) was lower than that in group A, but the level of HIF-1αmRNA in group B was greater than that in group A. The level of miR-199a in moderate and severe psoriasis patients was lower than that in mild psoriasis patients, and that in severe psoriasis patients was lower than that in moderate psoriasis patients. HIF-1α mRNA in lesions was positively associated with the PASI in psoriasis patients, and the miR-199a level in lesions was positively associated with the PASI in psoriasis patients. Furthermore, taking the PASI as the dependent variable and HIF-1αmRNA and miR-199a with a linear relationship with the dependent variable as the independent variables, multiple stepwise regression analysis showed that HIF-1α mRNA and miR-199a influenced the PASI.

Conclusions: HIF-1α and miR-199a are differentially expressed in the lesions of patients with different severities of psoriasis. The HIF-1α and miR-199a levels in lesions can be used as potential indices to evaluate the severity of psoriasis.

Key words: psoriasis, HIF-1α, miR-199a, PASI.

Introduction

One prevalent, recurring, immune system-related, chronic skin condition is psoriasis, which is characterised by abnormal proliferation of epidermal keratinocytes, inflammatory cell infiltration, and microangiogenesis [1]. Psoriasis has a complicated aetiology, mostly attributed to the interaction of genetic factors, immunology, and the environment [2]. Recent research has indicated that there may be a strong correlation between hypoxia and the onset and progression of psoriasis. Under hypoxic conditions, psoriasis may arise from a number of cytokines and signalling pathways that are triggered by hypoxia in animals [3]. As a factor closely related to cell proliferation, miR-199a has been found to be downregulated in breast cancer, liver cancer, and other cancer cells [4], and bioinformatics has revealed that miR-199 is differentially expressed in psoriasis [5]; however, research on its mechanism in psoriasis is very limited. HIF-1α, a DNA binding protein induced by ischaemia and hypoxia, has been shown to activate a range of linked target genes and mediate many pathways involved in a range of biological functions, including angiogenesis, energy metabolism, and cell division [6]. It is evident that HIF-1α and miR-199a contribute to angiogenesis and cell proliferation and may play a role in the onset and progression of psoriasis.

Aim

The aim of this research was to observe the expression of miR-199a and HIF-1α in psoriatic lesions and to

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explore the correlation between their expression and the severity of psoriasis.

Material and methods

General information

The data of patients with psoriasis vulgaris admitted from January 2023 to May 2024 were collected.

The inclusion criteria for patients were as follows: (1) satisfied the diagnostic requirements for psoriasis vulgaris according to the Chinese Clinical Dermatology [7]; (2) aged between 18 and 75 years; and (3) provided informed consent.

The exclusion criteria were as follows: (1) chronic inflammation-related disease; (2) other immune system disease; (3) severe heart, liver, renal, or other vital organ dysfunction; (4) malignant tumour; (5) treatment with psoriasis vulgaris-related drugs; and (6) long-term use of immunosuppressive drugs and/or corticosteroids.

During the same period, 40 healthy people who underwent surgical operations for accidental injuries in our hospital comprised the control group (group A). The inclusion criteria for group A were as follows: (1) no acute or chronic inflammatory disease; (2) normal heart, liver, kidney, and other important organ function; (3) no malignant tumours; and (4) provided informed consent.

There were 65 patients with psoriasis vulgaris, including 34 males and 31 females, with an average age of 46.58 ±18.65 years. Group A consisted of 40 patients, with an average age of 45.58 ±17.56 years, and consisted of 24 males and 16 females. Age and sex differences were not statistically significant across the 3 groups.

Observation indices

The main results were as follows: (1) The skin lesion area and severity index (PASI) were used to quantitatively evaluate the severity of skin lesions in the 2 groups. The 4 sections of the whole-body surface area were the trunk, lower extremities, upper limb, and head and neck. The more significant the lesion, the greater the score.

(2) Fluorescence quantitative PCR was used to detect the expression of HIF-1αmRNA and miR-199a in psoriatic lesions and normal skin tissues. The typical skin lesions of psoriasis patients were removed and at the normal skin tissue was taken from surgically cut skin tissue, which was frozen in liquid nitrogen and stored at –80°C after quick freezing in liquid nitrogen. Total RNA was extracted with TRIzol reagent according to the manufacturer's instructions. After extraction and purification, an appropriate amount of total RNA was obtained, and all steps were carried out according to the standard operation of the PrimeScriptTM 1st Strand cDNA Synthesis Kit to synthesise cDNA. Real-time fluorescence quantitative PCR was used for qPCR. The PCR conditions were 95°C for 5 min, followed by 45 cycles of 95°C for 35 s, 54°C for 45 s, and 72°C for 30 s, followed by 72°C for 5 min. The primers used are shown in Table 1.

Table 1. Primer sequences

Gene	Primer	Sequence
HIF-1α	F	5'-GATGGAAGCACTAGACAAAGTTCA-3'
	R	5'-ATCAGTGGTGGCAGTGGTAGTG-3'
miR-199a	F	5'-TCCAGCTGGGCCCAGTGTTCAGACTAC-3'
	R	5'-GTGTCGTGGAGTCGGCAATT-3'
U6	F	5'-CTCGCTTCGGCAGCACA-3'
	R	5'-AACGCTTCACG AATTTGCGT-3'

Statistical analysis

The SPSS 20.0 software package was used to analyse the study's data, and ($x \pm s$) was used to compare all measurement data that fit a normal distribution. Independent sample *t* tests were used for comparisons between 2 groups, and Pearson's linear correlation was used to analyse the correlation of the changes in each factor. There was a statistically significant difference in the outcomes ($p < 0.05$). Compared to the mild group, $a^P < 0.05$ indicates a moderate group and $b^P < 0.05$ indicates a moderate group.

Results

HIF-1α and miR-199a levels in the skin lesions of subjects in each group

The level of miR-199a in the lesion in group B was lower than that in group A, but the HIF-1α mRNA level in group B was greater than that in group A (Table 2).

Correlation analysis of miR-199a and HIF-1α

In group B, there was a negative correlation between the levels of miR-199a and HIF-1α ($r = 0.427$, $p < 0.01$).

The HIF-1α and miR-199a levels in skin lesions in the PASI groups

The level of HIF-1αmRNA in moderate and severe psoriasis patients was greater than that in mild psoriasis patients, and that in severe psoriasis patients was greater than that in moderate psoriasis patients. The miR-199a level in moderate and severe psoriasis patients was lower than that in mild psoriasis patients, and that in severe psoriasis patients was lower than that in moderate psoriasis patients (Table 3).

Table 2. HIF-1α and miR-199a levels in the skin lesions of subjects in each group ($x \pm s$)

Group	*N*	HIF-1α mRNA	miR-199a
A	40	1.04 ±0.15	1.35 ±0.43
B	65	1.53 ±0.41	0.78 ±0.26
t		7.254	8.474
P-value		< 0.001	< 0.001

Table 3. HIF-1α and miR-199a levels in skin lesions in the PASI groups (x ± s)

Group	*N*	HIF-1α mRNA	miR-199a
Mild	16	1.25 ±0.35	1.28 ±0.25
Moderate	23	1.51 ±0.29	0.87 ±0.19
Severe	26	1.84 ±0.46	0.53 ±0.18
F		12.470	68.492
P-value		< 0.001	< 0.001

Analysis of the relationship between the levels of HIF-1α and miR-199a in lesions and the PASI in psoriasis patients

HIF-1α mRNA was positively correlated with the psoriatic PASI, and there was a positive correlation between the amount of miR-199a and the psoriatic PASI (Table 4).

Multiple linear regression analysis of PASI and HIF-1α and miR-199a levels in lesions

Furthermore, taking the PASI as the dependent variable and HIF-1αmRNA and miR-199a with a linear relationship with the dependent variable as the independent variables, multiple stepwise regression analysis showed that HIF-1αmRNA and miR-199a influenced the PASI (Table 5).

Discussion

Previous research using miRNA biochip analysis revealed variations in the expression of many kinds of miRNAs, such as miR-199a, in psoriasis and healthy skin. In this study, the level of miR-199a in psoriatic lesions was lower than that in healthy lesions. The results are similar to those of others. Similarly, miR-199a is expressed at low levels in breast cancer. High expression of miR-199a can effectively inhibit the target gene HIF-1α, prevent breast cancer cells from proliferating, and promote their death [8]. In rats under hypoxia, miR-199a-5p suppresses the production of hypoxia inducible factor-1α and the proliferation of airway smooth muscle [9]. Some studies have confirmed the enduring connection between miR-199a and HIF-1α and the transfection of human immortalised epidermal cells with miR-199 mimics. It was found that overexpression of miR-199a could downregulate HIF-1α. It can be seen that miR-199a targeting HIF-1α can regulate the proliferation of epidermal cells [10]. In the hypoxia model of cardiomyocytes, the upregulation of

Table 4. Analysis of the relationship between the levels of HIF-1α and miR-199a in lesions and PASI in psoriasis patients

Project	PASI	
	r	*P*-value
HIF-1α mRNA	0.416	0.001
miR-199a	–0.278	0.029

HIF1α expression can eliminate miR-199a-5p-induced cell proliferation [11]. In the adrenal pheochromocytoma cell line PC12, Dex upregulates HIF-1α by reducing the level of miR-199a because miR-199a targets the 3'-UTR of HIF-1α [12]. By reducing HIF-1α and STAT3, small cell lung cancer cells are sensitive to bevacizumab, while the HIF-1α/STAT3 axis inhibits miR-199a-5p, establishes a positive feedback cycle, and promotes the continuous progression of non-small cell lung cancer [13]. According to this study, there was a poor relationship between HIF-1α and miR-199a levels in psoriatic lesions, indicating that miR-199a may also control the development and course of psoriasis by targeting HIF-1α in psoriasis.

In vitro, human keratinocyte cell line growth increases with increasing HIF-1α expression, and there is a considerable increase in HIF-1α expression in HaCaT cells and HKCs [14]. In another study, the reduction in terminal differentiation of keratinocytes was linked to increased expression of HIF-1α in primary human keratinocytes [15]. There is a positive correlation between HIF-1α and VEGF in psoriatic skin. In addition, exogenous administration of IGF-II has been widely reported to be related to increased VEGF and HIF-1α in HaCaT cells and is involved in the regulation of psoriasis [16]. It has been confirmed that the increase in HIF-1α and VEGF induced by IGF-II may be mediated by ERK2 phosphorylation. In addition, an ERK2 inhibitor (PD98059) inhibits the phosphorylation of ERK2 and reduces HIF-1α and VEGF, suggesting that IGF-II increases HIF-1α through the ERK2 signalling pathway, thus promoting the expression of VEGF [17].

An essential intracellular signal route that controls the biological activity of the skin and other organs is PI3K/Akt. According to some studies, psoriasis is largely caused by activation of the PI3K/Akt pathway, and there is a connection between the Akt pathway and HIF-1α in psoriasis [18]. Additionally, there is a connection between the Akt pathway and HIF-1α in psoriasis. Previous studies have shown that in keratinocytes from psoriatic patients, especially in the dermis, the levels of phosphorylated Akt

Table 5. Multiple linear regression analysis of the PASI score and HIF-1α and miR-199a levels in lesions

Independent variable	Regression coefficient	Standard error	Quasi-regression coefficient	*t*	*P*-value
Constant	23.65	2.514	–	10.247	< 0.001
HIF-1α mRNA	3.874	1.309	0.165	3.041	0.002
miR-199a	–2.512	1.254	–0.108	–2.057	0.028

and HIF-1α increase in parallel [19]. Human keratinocytes (Hacat cells) treated with LPS express HIF-1α and phosphorylate Akt at Ser473. This finding suggests that the enhanced production of HIF-1α may be associated with the activation of the PI3K/Ak pathway. In addition, wortmannin treatment eliminated the increase in HIF-1α expression induced by LPS in keratinocytes, thus verifying the critical function of the PI3K/Akt pathway in keratinocyte (20)-induced HIF-1α expression [20]. It is evident that HIF-1α acts on multiple target genes to increase the development and incidence of psoriasis.

Conclusions

HIF-1α and miR-199a are differentially expressed in the lesions of patients with psoriasis of different severities, and the levels of HIF-1α and miR-199a in lesions can be used as potential indices to evaluate the severity of psoriasis.

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Ethical approval

Not applicable.

Conflict of interest

The authors declare no conflict of interest.

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