CXCL10 and its related key genes as potential biomarkers for psoriasis
Evidence from bioinformatics and real-time quantitative polymerase chain reaction

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Abstract
Although several studies have attempted to investigate the etiology of and mechanism underlying psoriasis, the precise molecular mechanism remains unclear. Our study aimed to explore the molecular mechanism underlying psoriasis based on bioinformatics. GSE30999, GSE34248, GSE41662, and GSE50790 datasets were obtained from the Gene Expression Omnibus database. The Gene Expression Omnibus profiles were integrated to obtain differentially expressed genes in R software. Then a series of analyses was performed, such as Gene Ontology annotation, Kyoto Encyclopedia of Genes and Genomes pathway analysis, protein-protein interaction network analysis, among others. The key genes were obtained by CytoHubba, and validated by real-time quantitative polymerase chain reaction.

A total of 359 differentially expressed genes were identified between 270 paired lesional and non-lesional skin groups. The common enriched pathways were nucleotide-binding and oligomerization domain-like receptor signaling pathway, and cytokine-cytokine receptor interaction. Seven key genes were identified, including CXCL1, ISG15, CXCL10, STAT1, OASL, IFIT1, and IFIT3. These key genes were validated as upregulated in the 4 datasets and M5-induced HaCaT cells.

Our study identified 7 key genes, namely CXCL1, ISG15, CXCL10, STAT1, OASL, IFIT1, and IFIT3, and 2 mostly enriched pathways (nucleotide-binding and oligomerization domain-like receptor signaling pathway, and cytokine-cytokine receptor interaction) involved in psoriatic pathogenesis. More importantly, CXCL1, ISG15, STAT1, OASL, IFIT1, IFIT3, and especially CXCL10 may be potential biomarkers. Therefore, our findings may bring a new perspective to the molecular mechanism underlying psoriasis and suggest potential biomarkers.

Abbreviations: CXCL = chemokine (C-X-C motif) ligand, DEGs = differentially expressed genes, GEO = Gene Expression Omnibus, GO = Gene Ontology, KEGG = Kyoto Encyclopedia of Genes and Genomes, NOD = nucleotide-binding and oligomerization domain, OASL = oligoadenylate synthetases-like, PPI = protein-protein interaction, RT-qPCR = real-time quantitative polymerase chain reaction.

Keywords: bioinformatics, biomarkers, CXCL10, key genes, psoriasis

1. Introduction
Psoriasis is a chronic and systemic inflammatory cutaneous disorder with a global prevalence of 2% to 3%.\textsuperscript{[1,2]} The occurrence of psoriasis is due to complex interactions among genetics, immunology, and the environment.\textsuperscript{[1,3]} Although many studies have investigated the etiologies and mechanisms, the precise molecular mechanism of psoriasis remains unclear.\textsuperscript{[3]} A thorough exploration of psoriasis pathogenesis would be helpful for discovery of potential biomarkers and could provide novel clues for diagnosis and treatment.
Bioinformatics has been extensively applied to many diseases, including psoriasis.\(^4,5\) Gene Expression Omnibus (GEO) is an online and free database, that includes various disease gene expression datasets.\(^4\) Thus, we can utilize bioinformatics analysis to conveniently explore the molecular mechanism underlying psoriasis from the GEO database.

In recent years, many scholars have mined key genes related to the pathogenesis of psoriasis by using bioinformatics methods.\(^5-7\) For example, Gao et al\(^7\) reported 7 key genes in psoriasis (HERC6, MX1, OA52, OASL, OAS3, ISG15, and RSAD2). Meanwhile, Delic et al\(^5\) considered AURKA, CSK2, CDC45, CENPE, DLGP5, HMMR, IFIT1, IFIT1, IFI27, ISG20, NDC80, NUF2, MCM10, RRM2, SPC25, RSAD2, and TTK as key genes. These conclusions demonstrate that numerous genes are related to the pathogenesis of psoriasis. Thus, we can still explore other key genes from different viewpoints.

As we know, the diagnosis of psoriasis is not difficult, however its effective treatment does not exist till now.\(^8\) It is helpful to explore key genes for discovering potential biomarkers and provide therapeutic targets for psoriasis. Thus, our primary objective is to search key genes for psoriasis that could be potential biomarkers.

In our study, we attempted to identify key genes and associated pathways in psoriasis using bioinformatics analysis, and compare the expression levels of key genes between lesional and non-lesional psoriatic skin based on 4 datasets. Finally, we carried out real-time quantitative polymerase chain reaction (RT-qPCR) experiments in M5 induced HaCaT cells for validation. Since some studies confirmed that M5 (IL-22, TNF-a, IL-17A, IL-1a, and Oncostatin M) can induce a better psoriatic cell model,\(^9-14\) we use it to treat HaCaT cells in our experiments. Therefore, our findings may bring a new perspective to the molecular mechanism underlying psoriasis and suggest potential biomarkers.

2. Materials and Methods

2.1. Microarray datasets collection and identification of differentially expressed genes

Figure 1 demonstrates the workflow of this study. Four microarray datasets (GSE30999\(^{15}\), GSE34248\(^{16}\), GSE41662,\(^{16}\) and GSE50790\(^{17}\)) were downloaded from GEO (https://www.ncbi.nlm.nih.gov/geo/). A total of 127 paired lesional and non-lesional skin samples were selected as subjects from plaque psoriasis patients in the 4 datasets. All subjects were from homo sapiens and GPL570 platform ([HG-U133_Plus_2] Affymetrix Human Genome U133 Plus 2.0 Array). The detailed sample information was summarized in Table 1 (The data were by the year of March 2021).

The raw data of 4 datasets were collated and analyzed using R software (version 4.0.2.). Since the raw data were from 4 different microarray datasets, the collated data were processed by background correction and normalized using the “affy” package,\(^18\) and the batch effect was eliminated using the ComBat function of sva package.\(^19\) The limma package was used to identify differentially expressed genes (DEGs) between 127 paired lesional and non-lesional psoriatic skin tissues. The cutoff value was set as \(\log FC > 1.5\) and adjusted \(P < .05\), which was demonstrated as a volcano plot. Then the clustering of samples was shown as a heatmap.

2.2. Gene Ontology and Kyoto Encyclopedia of Genes and Genomes enrichment analyses of DEGs

Gene Ontology (GO) annotation and Kyoto Encyclopedia of Genes and Genomes (KEGG) enrichment analysis of DEGs were performed using Metascape (http://metascape.org), which is a free and online analysis tool.\(^20\) GO annotation comprises cellular component, molecular function, and biological process.
The cutoff criteria were a P value < .05, minimum overlap of 3, and minimum enrichment of 1.5.

### 2.3. Protein-protein interaction network construction and screening for key genes

The STRING database (version 11.0, http://www.string-db.org/) was utilized to obtain protein-protein interaction (PPI) information for DEGs (high confidence of 0.7 was chosen) [21]. Then, Cytoscape 3.8.0 was applied to visualize the PPI network [22]. The modules of the PPI network were explored using MCODE, and key genes were screened with CytoHubba. This could provide 12 topological analysis methods to identify the top 10 genes. A key gene was identified, if the gene was predicted to be one of the top 10 genes in all 12 methods. Finally, the interactions between the hub genes were returned to the STRING database for analysis.

### 2.4. Functional enrichment analysis of 7 key genes

To confirm the validity of the 7 key genes, functional enrichment analysis of these genes was further analyzed with http://www.bioinformatics.com.cn.

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### Table 1

| GEO accession | GSE30999 | GSE34248 | GSE41662 | GSE50790 |
|---------------|----------|----------|----------|----------|
| Organism      | Homo sapiens | Homo sapiens | Homo sapiens | Homo sapiens |
| Tissue        | Skin     | Skin     | Skin     | Skin     |
| Platform      | GPL570   | GPL570   | GPL570   | GPL570   |
| Sample        | Paired LS/NL | Paired LS/NL | Paired LS/NL | Paired LS/NL |
| Pair no.      | 85       | 14       | 24       | 4        |
| Type          | Plaque psoriasis | Plaque psoriasis | Plaque psoriasis | Plaque psoriasis |
| Citation      | Suárez-Fariñas et al [15] | Bigler et al [16] | Bigler et al [16] | Swindell et al [17] |

**GEO** = Gene Expression Omnibus, **LS** = lesional skin, **NL** = non-lesional skin.

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### 2.5. Validation of 7 key genes in the 4 datasets

The 7 key genes were validated in the 4 aforementioned datasets. Moreover, the relationships between CXCL10 and the other hub genes were also confirmed based on the 4 datasets.

### 2.6. Validation of 7 key genes via RT-qPCR

First, HaCaT cells (Shanghai iCell Bioscience Inc.) were cultured in Dulbecco Modified Eagle’s Medium at 37°C in a 5% CO2 environment with 10% fetal bovine serum and 1% penicillin and streptomycin. Then, the cells were treated with 10ng/mL M5 (IL-22, TNF-a, IL-17A, IL-1a, and Oncostatin M) (PeproTech) for 48 hours to induce the psoriasis cell model. After that, total RNA was extracted from M5-treated HaCaT cells using the TRIpure Total RNA Extraction Reagent (#EP013, ELK Biotechnology, China). Total RNA was reverse transcribed using the EntiLink 1st Strand Cdna Synthesis Kit (#EQ003, ELK Biotechnology, China) according to the manufacturer’s protocols. The expression levels of the 7 key genes in HaCaT cells and M5-treated HaCaT cells were detected by RT-qPCR using EnTurbo SYBR Green PCR SuperMix (#EQ001, ELK Biotech-
2.7. Statistical analysis

The data extracted from GEO datasets were examined by the normality test and homogeneity of variance test. The testing and analysis of variance testing were used for comparisons between 2 groups and among 3 or more groups respectively. Spearman correlation analysis was utilized to investigate the relationships between CXCL10 and other hub genes. GraphPad Prism 8.0.1 was used to perform these tests.

3. Results

3.1. Identification of DEGs

Finally, 359 DEGs were identified between 270 paired lesional and non-lesional skin groups with $|\log FC| > 1.5$ and adjusted $P < .05$, of which 284 were up-regulated and 65 were down-regulated. The volcano plot and heatmap of DEGs were shown in Figures 2 and 3, respectively.

3.2. GO and KEGG pathway analyses of DEGs

The GO annotation of DEGs was mostly enriched in 6 clustering groups, including response to bacterium, defense response to other organism, anti-microbial humoral response, flavonoid glucuronidation, skin development, and monocarboxylic acid metabolic process (Fig. 4A, B). The top 20 GO items were of the biological process group (15), and molecular function group (5) (Table 2). KEGG pathway analysis of DEGs indicated that genes were mostly enriched in 3 clustering groups, including steroid hormone biosynthesis, nucleotide-binding and oligomerization domain (NOD)-like receptor signaling pathway, and cytokine-cytokine receptor interaction (Fig. 4C, D). The top 20 KEGG pathway enriched items were shown in Table 3.

3.3. PPI network construction and screening for key genes

The PPI network of 359 DEGs was acquired with the STRING database, which was visualized using Cytoscape software (Fig. 5A). Then MCODE was used to identify functional modules. There were 6 modules, of which cluster 1 consisted of 28 nodes and 196 edges (Fig. 5B), including GAL, CXCL13, GBP1, IFIT3, RTP4, IFIT1, MXI1, OASL, IFI44, ISG15, CCL27, OAS1, K5AD2, CXCR2, CXCL8, IFI44L, IFI7, STAT1, CXCL1, PTGER3, OAS2, HERC6, IFI6, IFI27, CCL20, CXCL9, CXCL10, and CXCL2 (Table 4). Seven genes, namely CXCL1, ISG15, CXCL10, STAT1, OASL, IFIT1, and IFIT3, were considered the key genes by CytoHubba (Table 5). Subsequently, the relationships among the 7 key genes were explored with the STRING database and CXCL10 was central for the connection (Fig. 5C).

3.4. Functional enrichment analysis of 7 key genes

Seven key genes were further analyzed using an online tool (http://www.bioinformatics.com.cn). The enrichment results of 7 hub genes were shown in chord plots based on the adjusted $P$ values (Fig. 6). For GO analysis, the top 5 terms were response to bacterium, defense response to other organism, anti-microbial humoral response, flavonoid glucuronidation, and chemokine receptor binding (Fig. 6A). For KEGG pathway analysis, the top 3 terms were NOD-like receptor signaling pathway, cytokine-cytokine receptor interaction, and amoebiasis (Fig. 6B). These results were consistent with the Metascape analysis, which strengthened the reliability of the results.
3.5. Validation of 7 key genes in 4 datasets

The expression levels of 7 key genes were confirmed in GSE30999, GSE34248, GSE41662, and GSE50790 datasets. Except for CXCL1, STAT1, and OASL in GSE50790, the other key genes were obviously up-regulated in psoriatic lesional skin tissues (Fig. 7).

Table 2
The top 20 GO items of DEGs.

| GO       | Category | Description                           | Count | %    | Log10(P)  | Log10(q)  |
|----------|----------|---------------------------------------|-------|------|-----------|-----------|
| GO:0008008 | BP       | Response to bacterium                 | 53    | 14.25| -22.54    | -18.21    |
| GO:0008542 | BP       | Defense response to other organism    | 48    | 12.9 | -22.25    | -18.21    |
| GO:0019730 | BP       | Anti-microbial humoral response       | 23    | 6.18 | -18.1     | -14.22    |
| GO:0032696 | BP       | Flavonoid glucuronidation             | 9     | 2.42 | -16.93    | -13.18    |
| GO:0035888 | BP       | Skin development                      | 31    | 8.33 | -13.69    | -10.48    |
| GO:0032787 | BP       | Monocarboxylic acid metabolic process | 33    | 8.87 | -9.98     | -7.24     |
| GO:0033066 | BP       | Cell killing                          | 17    | 4.57 | -9.62     | -6.91     |
| GO:0035103 | BP       | Positive regulation of response to external stimulus | 20 | 5.38 | -7.45 | -5.01 |
| GO:0044282 | BP       | Small molecule catabolic process      | 23    | 6.18 | -7.24     | -4.82     |
| GO:0009811 | BP       | Response to wounding                  | 29    | 7.8  | -6.99     | -4.58     |
| GO:0008002 | BP       | Steroid metabolic process             | 19    | 5.11 | -6.9      | -4.51     |
| GO:0016042 | BP       | Lipid catabolic process               | 19    | 5.11 | -6.69     | -4.3      |
| GO:0006766 | BP       | Vitamin metabolic process             | 12    | 3.23 | -6.53     | -4.16     |
| GO:0036293 | BP       | Granulocyte activation                | 23    | 6.18 | -6.38     | -4.02     |
| GO:0002831 | BP       | Regulation of response to biotic stimulus | 12 | 3.23 | -6.17 | -3.82 |
| GO:0042379 | MF       | Chemokine receptor binding            | 16    | 4.3  | -15.05    | -11.66    |
| GO:0016491 | MF       | Oxidoreductase activity               | 31    | 8.33 | -7.43     | -5        |
| GO:0045236 | MF       | CXCR chemokine receptor binding       | 6     | 1.61 | -7.07     | -4.65     |
| GO:0042522 | MF       | Serine-type endopeptidase activity    | 13    | 3.49 | -6.27     | -3.92     |
| GO:0004867 | MF       | Serine-type endopeptidase inhibitor activity | 10 | 2.09 | -6.12 | -3.79 |

BP = biological process, DEGs = differentially expressed genes, GO = Gene Ontology, MF = molecular function.
### Table 3
The top 20 KEGG pathways of DEGs.

| GO        | Category              | Description                              | Count | %     | Log10(P)  | Log10(q)  |
|-----------|-----------------------|------------------------------------------|-------|-------|-----------|-----------|
| ko00140   | KEGG pathway          | Steroid hormone biosynthesis             | 13    | 3.49  | −12.22    | −9.75     |
| hsa04621  | KEGG pathway          | NOD-like receptor signaling pathway      | 19    | 5.11  | −11.11    | −8.9      |
| hsa04060  | KEGG pathway          | Cytokine-cytokine receptor interaction   | 24    | 6.45  | −10.74    | −8.68     |
| ko05146   | KEGG pathway          | Amoebiasis                               | 9     | 2.42  | −5.26     | −3.91     |
| ko03320   | KEGG pathway          | PPAR signaling pathway                   | 7     | 1.88  | −4.31     | −3.01     |
| hsa05219  | KEGG pathway          | Bladder cancer                           | 5     | 1.34  | −3.54     | −2.29     |
| ko04110   | KEGG pathway          | Cell cycle                               | 7     | 1.88  | −2.86     | −1.65     |
| hsa00630  | KEGG pathway          | Glyoxylate and dicarboxylate metabolism  | 4     | 1.08  | −2.84     | −1.65     |
| ko04623   | KEGG pathway          | Cytosolic DNA-sensing pathway            | 5     | 1.34  | −2.82     | −1.64     |
| ko05321   | KEGG pathway          | Inflammatory bowel disease (IBD)         | 5     | 1.34  | −2.76     | −1.59     |
| ko00100   | KEGG pathway          | Steroid biosynthesis                     | 3     | 0.81  | −2.65     | −1.51     |
| hsa00240  | KEGG pathway          | Pyrimidine metabolism                    | 6     | 1.61  | −2.47     | −1.37     |
| hsa00380  | KEGG pathway          | Tryptophan metabolism                    | 4     | 1.08  | −2.36     | −1.28     |
| hsa04978  | KEGG pathway          | Mineral absorption                       | 4     | 1.08  | −2.27     | −1.2      |
| hsa05150  | KEGG pathway          | Staphylococcus aureus infection          | 4     | 1.08  | −1.66     | −0.68     |
| hsa05205  | KEGG pathway          | Proteoglycans in cancer                  | 7     | 1.88  | −1.57     | −0.61     |
| hsa05685  | KEGG pathway          | Ether lipid metabolism                   | 3     | 0.81  | −1.56     | −0.61     |
| ko04972   | KEGG pathway          | Pancreatic secretion                     | 4     | 1.08  | −1.41     | −0.48     |
| ko04310   | KEGG pathway          | Wnt signaling pathway                    | 5     | 1.34  | −1.37     | −0.45     |
| ko00561   | KEGG pathway          | Glycerolipid metabolism                  | 3     | 0.81  | −1.36     | −0.44     |

DEGs = differentially expressed genes, GO = Gene Ontology, KEGG = Kyoto Encyclopedia of Genes and Genomes, NOD = nucleotide-binding and oligomerization domain.

### Figure 5
PPI network and hub genes. (A) The PPI network was constructed in Cytoscape, with upregulated genes revealed in red ellipses, downregulated genes in blue ellipses, and cluster 1 genes in yellow ellipses. (B) Cluster 1 network was constructed with upregulated genes revealed in red ellipses and downregulated genes in blue ellipses. (C) The connection function between 7 hub genes in STRING database. PPI = protein-protein interaction.
3.6. Relationship between CXCL10 and the other key genes

For the relationships among the 7 key genes, CXCL10 was central to the network (Fig. 5C), had a higher rank (Table 5), and a demonstrated positive correlation with the other 6 key genes in the 4 aforementioned datasets (Fig. 8), suggesting the close relationship between CXCL10 and the other 6 key genes.

3.7. Validation of key genes via qRT-PCR

The expression levels of 7 key genes were validated by RT-qPCR (n = 3). The results of qRT-PCR showed that the transcription levels of CXCL10, CXCL1, ISG15, STAT1, OASL, IFIT1, and IFIT3 were significantly up-regulated in 10 ng/mL M5 induced HaCaT cells (Fig. 9). The expression levels of these genes were consistent with the microarray results.

4. Discussion

Psoriasis is a chronic, relapsing-remitting, and inflammatory skin disease that affects 2% to 3% of the population worldwide. Recurrence has been common after treatment in psoriasis. That is to say, there are currently no known radical treatments for

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Table 4

| Cluster | Score (density *#nodes) | Nodes | Edges | Node IDs |
|---------|-------------------------|-------|-------|----------|
| 1       | 14.519                  | 28    | 196   | GAL, CXCL13, GBP1, IFT1, RTP4, IFIT1, MX1, OASL, IF44, ISG15, CCL27, OAS1, RSAD2, CXCR2, CXCL8, IF44L, IFF7, STAT1, CXCL1, PTGER3, OAS2, HERC6, IFI6, IFI27, CCL20, CCL9, CXCL10, CXCL2 |
| 2       | 12.167                  | 13    | 73    | DLGAP5, KIAA0101, KIF20A, CCNA2, CDC6, BUB1, CENPE, RRM2, CDK1, CCNB1, MCM10, TKK, SPC25 |
| 3       | 9.8                     | 11    | 49    | LCE3D, SPRR2B, IFL, SPRR3, SPRR1A, PKP1, DSG3, DCSN, DSC2, TGM1, P3 |
| 4       | 6                       | 9     | 24    | TNC1, IL1B, IL17A, RAB27A, LRG1, HPSE, ARG1, LCN2, LTF |
| 5       | 4                       | 4     | 6     | KRT16, KRT6A, KRT77, KRT6B |
| 6       | 3                       | 3     | 3     | S100A12, S100A9, S100A8 |

Table 5

| Rank | Gene symbol | Change | LogFC | Adj. P value | Occurrences in 12 statistical methods by CytoHubba | Full name (human) |
|------|-------------|--------|-------|--------------|--------------------------------------------------|------------------|
| 1    | CXCL1       | Up     | 3.4145| 9.27E-42     | 8                                                 | C-X-C motif chemokine ligand 1 |
| 1    | ISG15       | Up     | 1.9624| 2.62E-38     | 8                                                 | ISG15 ubiquitin like modifier |
| 2    | CXCL10      | Up     | 2.3289| 2.19E-27     | 7                                                 | C-X-C motif chemokine ligand 10 |
| 3    | STAT1       | Up     | 1.7999| 1.36E-47     | 6                                                 | Signal transducer and activator of transcription 1 |
| 4    | OASL        | Up     | 3.7520| 1.47E-66     | 5                                                 | 2′-5′-oligoadenylate synthetase like |
| 4    | IFIT1       | Up     | 1.7033| 3.18E-33     | 5                                                 | Interferon induced protein with tetratricopeptide repeats 1 |
| 4    | IFIT3       | Up     | 1.6149| 4.02E-34     | 5                                                 | Interferon induced protein with tetratricopeptide repeats 3 |

Figure 6. The chord plots of enrichment analysis of 7 hub genes. (A) GO enrichment analysis. (B) KEGG enrichment analysis. GO = Gene Ontology, KEGG = Kyoto Encyclopedia of Genes and Genomes.
psoriasis. Thus, it is important and urgent to investigate the molecular mechanisms involved in the pathogenesis of psoriasis to provide new clues for treatment. An important discovery of our study was the confirmation of key genes in psoriasis by bioinformatics and via RT-qPCR. The 7 key genes comprised CXCL10, CXCL1, ISG15, STAT1, OASL, IFIT1, and IFIT3, and the 2 common enriched pathways were NOD-like receptor signaling pathway and cytokine-cytokine receptor interaction.

Among the 7 key genes, CXCL10 was a higher ranked gene and had positive correlations with other 6 hub genes, suggesting that it might be the most significant gene. Furthermore, the RT-qPCR results of CXCL10 confirmed this prediction. Some studies have also reported several hub genes in psoriasis, some of which are consistent with ours, especially the study of Luo et al. They reported that CXCR2, CXCL10, IVL, OASL, and ISG15 were hub genes, and CXCL10 was the hub gene with the highest degree. However, they did not perform experiments to validate the hub genes. CXCL10 is a member of the CXC family of chemokines, and plays a significant role in inflammation through its T-cell chemotactic and adhesion properties. Researches have also indicated CXCL10 is up-regulated in psoriatic skin lesions and serum. It has been hypothesized that CXCL10 could be a good marker for psoriasis. Although CXCL10 and CXCL1 belong to the chemokine “CXC” family, they play different roles in psoriasis. CXCL10 attracts T helper (Th) 1 cells, whereas CXCL1 attracts neutrophils. It was reported that CXCL10 production by keratinocytes depends on STAT1. STAT1 is known as a member of the STAT family, involved in type I and type II interferon signaling. The expression of STAT1 is increased in psoriatic skin, and it also has a vital role in the pathogenesis of psoriasis.

The remaining 4 genes (ISG15, OASL, IFIT1, and IFIT3) are anti-viral genes, which may explain the relatively fewer viral skin infections found in psoriasis patients. Ubiquitin-like protein ISG15 is an interferon-stimulated protein that has a critical role in the control of microbial infections. Some studies reported that ISG15 is elevated in psoriatic skin compared with levels in atopic dermatitis skin and healthy skin. The interferon-inducible oligoadenylate synthetases-like (OASL) protein belongs to the atypical oligoadenylate synthetase family, possesses antiviral activity, and boosts innate immunity. Gao et al. also identified OASL as a hub gene, but it has been rarely studied in psoriasis patients. Although the expression of OASL was up-regulated in M5-induced HaCaT cells in our study, this needs to be further confirmed.

Figure 7. The expression of 7 hub genes in 4 datasets. The green bar indicates non-lesional skin groups, and the red bar indicates lesional skin groups. Paired t-testing was performed to compare the means of 2 groups. *P < .05; **P < .01; ***P < .001; ****P < .0001. LS = lesional skin, NL = non-lesional skin, ns = no significance.
Figure 8. Spearman correlation analysis confirming the significant correlation between CXCL10 and the other hub genes. (A) GSE30999: CXCL1 (R = 0.72), ISG15 (R = 0.67), STAT1 (R = 0.77), OASL (R = 0.71), IFIT1 (R = 0.79). (B) GSE34248: CXCL1 (R = 0.70), ISG15 (R = 0.75), STAT1 (R = 0.85), OASL (R = 0.78), IFIT1 (R = 0.67), IFIT3 (R = 0.79). (C) GSE41662: CXCL1 (R = 0.82), ISG15 (R = 0.58), STAT1 (R = 0.79), OASL (R = 0.74), IFIT1 (R = 0.53), IFIT3 (R = 0.68). (D) GSE50790: CXCL1 (R = 0.76), ISG15 (R = 0.81), STAT1 (R = 0.90), OASL (R = 0.92), IFIT1 (R = 0.81), IFIT3 (R = 0.76).

Figure 9. The results of RT-qPCR. The transcription levels of CXCL10, CXCL1, ISG15, STAT1, OASL, IFIT1, and IFIT3 were significantly up-regulated in M5 group. ∗∗∗∗P < .0001. M5, HaCat cells were treated with 10 ng/mL M5 (IL-1α, IL-17, IL-22, TNF-α, and Oncostatin M); Control, HaCat cells without M5 stimulation. RT-qPCR = real-time quantitative polymerase chain reaction.
The IFITs include *IFIT1*, *IFIT2*, *IFIT3*, and *IFIT5*,[37] which regulate immune responses and function as essential anti-viral proteins.[38] One study indicated that *IFIT3* binding to *ITI* is vital for stabilizing *ITI* expression, and is indispensable for inhibiting infection by viruses lacking 2′-O methylation.[38] In this study, *ITI* was enriched in the NOD-like receptor signaling pathway, but *ITI* was not. Currently, there are few studies on IFITs in psoriasis; however, *ITI* and *ITI* are overexpressed in oral squamous cell carcinoma, and promote tumor growth and regional and distant metastasis.[37] Therefore, this new finding warrants further study.

The NOD-like receptor is a type of pattern-recognition receptor. It is also associated with various diseases related to infection and immunity.[39] Some studies showed that the NOD-like receptor signaling pathway was enriched in psoriatic epidermis.[40] Meanwhile, some researchers have found that cytokine-cytokine receptor interaction is related to the pathogenesis of psoriasis via combined transcriptomic analysis.[41] These results are consistent with ours.

In summary, we identified 7 key genes and 2 mostly enriched pathways for psoriasis. Our findings may bring a profound understanding for the molecular mechanism underlying psoriasis. However, this study has some limitations. First, the sample size is limited to a portion of the publicly available datasets and cannot be representative of the entire population. Second, there is potential bias of our data, due to different datasets. Third, we only performed 1 cell experiment. More experiments, such as skin tissues, are needed to support our results.

5. Conclusion
In our study, we tried to identify DEGs between psoriatic and non-psoriatic lesions by bioinformatics, discovered 7 hub genes and 2 mostly enriched pathways that might participate in the pathogenesis of psoriasis, and validated the hub genes upregulated in a psoriatic cell model through RT-qPCR. More interestingly, the 7 hub genes, namely *CXCL1*, *IGF1*, *STAT1*, OASL, *IFIT1*, *IFIT3*, and especially *CXCL10* may be used as potential biomarkers. Therefore, our findings may bring a new perspective to the molecular mechanism underlying psoriasis and suggest potential biomarkers.

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