Web crawling and mRNA sequencing analyze mechanisms of photobiomodulation

Hao Wu

Harbin Medical University

Caiyun Meng

Harbin Medical University

Qing Xia

Harbin Medical University

Hailiang Liu

Harbin Medical University

Fengmin Zhang

Harbin Medical University

Wuqi Song (✉ songwuqi@hrbmu.edu.cn)

Harbin Medical University <https://orcid.org/0000-0001-5900-5154>

Research

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Abstract

Background Photobiomodulation (PBM) is praised as a promising physical therapy, which has many advantages, such as noninvasive, painless. However, the mechanisms are not fully elucidated.

Methods Using web crawling, mRNA sequence and bioinformatics analysis to select genes, functional annotation, and mechanisms. The expressions of inflammatory cytokines were measured using RT-qPCR.

Results A total of 146 human genes and 57 pathways were identified about PBM. The 630nm LED-stimulated-MH7A cells were sequenced for further analyzing the mechanism of PBM. 2950 differentially expressed genes were identified, and the gene ontology term enrichment analysis and Kyoto encyclopedia of genes and genomes pathway analysis were performed to better understand functions and pathways. The 12 pathways were matched with the KEGG results of PBM and MH7A cells. A protein-protein interaction network was performed among genes in 12 pathways, and 10 outstanding proteins were identified. Importantly, the 9 genes were predicted with potential research value. And we also demonstrated that expression of inflammatory factors (IL-6, IL-1 β , IL-8, and MMP-3) was reduced; meanwhile, the expression of anti-inflammatory factor IL-10 was promoted after 630nm LED. In conclusion, using web crawling, bioinformatics analysis, and mRNA sequence, we obtained 9 key genes and 10 important pathways about PBM. Importantly, we demonstrated the anti-inflammatory effect of 630nm LED red light by RT-qPCR.

Introduction

PBM is a new physical therapy, and it utilizes endogenous chromophores with low dose illumination delivered at the target site, which is the opposite of photodynamic therapy by stimulating exogenous chromophore^[1]. PBM is also called low-level laser therapy (LLLT), which is a non-invasive form of phototherapy that utilizes wavelengths of light between 650 and 1000 nm to deliver low irradiance and doses to the target tissue^[2]. So far, low-level laser therapy has been recognized as a light therapy that may be used for anti-inflammation, promoting wound healing and bone regeneration^[3-5].

Although many articles have put forward many opinions, the mechanisms of PBM are not fully elucidated. Studies have demonstrated that NF- κ B was significantly activated on 0.003J /cm² by irradiating rat embryonic fibroblasts with 810nm laser^[6]. Some studies have found that LLLT can inhibit apoptosis through the PI3K-AKT signaling pathway. LPLI inhibits staurosporine (STS)-induced cell apoptosis by inactivating the GSK-3 β /Bax pathway^[7]. However, it has also been demonstrated that LLLT enhances the invasiveness of tumor cells by regulating the Akt/ mTOR signaling pathway^[8].

Photodynamic therapy (PDT) is a highly effective and safe treatment method of phototherapy^[9]. Photodynamic therapy is firstly reported in the 1970s and is honored as the most promising treatment model of the 20th century in the 1980s^[10]. It used chemical reactions between light and photosensitizers

in specific wavelengths to produce reactive oxygen species that kill target cells for therapeutic effect^[11]. Both light and photosensitizers were harmless and can be activated when exposed to a specific wavelength of light in a specific area. Therefore, this treatment has many advantages, including strong controllability, low toxicity, and non-invasive, etc^[12]. To demonstrate whether the mechanism of PBM and PDT is different, this study compares the similarities in the genes and pathways affected by PBM and PDT.

Light sources are laser and light-emitting diode(LED) in phototherapy. LED, as the fourth generation light source, has the advantages of energy-saving, environmental protection, and high electro-optical conversion efficiency. Therefore, LED is becoming more application in LLLT. However, the two light sources have different properties^[13]. Due to the laser is coherent light, but LED is incoherent light, thus one of our aims is to compare the effects of laser and LED.

In conclusion, we crawled 3352 literature, identified 146 human genes from the abstracts of the literature, and performed bioinformatic analyses to get 9 key genes and important 12 pathways about PBM.

Methods

1.Data collection

1.1 Crawling literature on photobiomodulation and photodynamic therapy

The research design was created in the form of a flowchart(Figure 1). In the NCBI PubMed database, with 'photobiomodulation' or 'low level laser therapy' as keywords in [Title and Abstract] to search PBM-related literature. The same way that keywords were 'photodynamic therapy' to search PDT-related literature. And crawled through R language to get the abstracts of the relevant literature.

1.2 Crawling LED, laser-related literature

In the NCBI PubMed database, due to the research on LED started late, there were few articles related to it. To expand the retrieval range, we searched relevant literature directly with the 'light-emitting diode' as a keyword. Similarly, in [title and abstract], 'laser' was used as the keyword to search laser-related literature. And crawled literature through R language to get the abstracts of the relevant literature.

2.Data analysis

2.1 Identification of genes in the relevant literature

Adopting the gene_atomization function in the pubmed. mineR package of the R language, the genes in the abstracts in the relevant literature can be directly extracted. The gene_atomization can be used to identify genes in relevant literature abstracts with PBM, PDT, LED, and laser.

2.2 Confirming the related genes of LED and laser phototherapy

The genes of the PBM group, PDT group, LED group, and laser group was identified, and their respective intersections were obtained through the Venn diagram. The intersection between LED genes and (PBM+PDT) group genes was LED -related phototherapy genes. Similarly, laser-related phototherapy genes were confirmed.

2.3 Enrichment of functions and pathways

To study the genes that have been identified from the relevant literature, we carried out a comprehensive set of functional annotations for them and used the "Clusterprofiler" package for gene ontology (GO) term enrichment analysis and Kyoto genes and Encyclopedia of Genomics (KEGG) pathway analysis^[14]. All KEGG pathway and GO enrichment analysis was based on a threshold of *p-value* < 0.05.

2.4 PPI network

Search Tool for the Retrieval of Interacting Genes/Proteins (STRING) database (<http://www.string-db.org/>) is a systems biology tool to assess protein-protein interaction (PPI) information^[15]. To evaluate the interrelationships behind the genes, we used the STRING database for analysis. Meanwhile, the MCODE^[16] plug-in of Cytoscape software was used to visualize and select hub modules of the PPI network. The degree = 5, node score = 0.2, k-core = 2, and maximum depth = 100 were used as cut-off criteria.

3. Cell culture

Human fibroblast-like synoviocyte line, MH7A was purchased from GuanDao Biological Research Center of Shanghai. Cells were cultured in DMEM supplemented with fetal bovine serum (10%) and penicillin/streptomycin solution (1%). The cultured cells were maintained at 37°C in a 5% CO₂ atmosphere.

4. Light-emitting diode bio-stimulation

Light-emitting diodes (CP-YL3213-630, Truwin Optoelectronic Medical, Beijing, China) were used to deliver 630-nm radiation. Cells were exposed through the bottom of plastic culture plates to the 630-nm radiation. The time it takes to radiation once was 15 min to achieve radiation energies of 39 J/cm². TNF-α stimulation groups in which TNF-α (10 ng/mL, PeproTech) was added before the radiation. Cells of experimental groups were radiated at room temperature. The control groups (non-irradiated cells) were wrapped with aluminum foil and kept at room temperature while the experimental samples were radiated.

5. Gene sequencing

mRNA-seq was performed as reported in Genesky Biotechnologies Inc., Shanghai, China. We used the "limma" R package to screen the differentially expressed genes between LED+TNF-α samples and normal samples^[17]. *P*-value < 0.05 and |log₂fold change (FC)| > 1, were chosen as the cut-off threshold.

6. RT-qPCR

Reverse transcriptase polymerase chain reaction (RT-PCR) was performed on total RNA harvested from synovioblasts with RNeasy mini kit (HaiGene, Harbin, China). Equal amounts of total RNA (2 µg) was reverse transcribed by Thermo script RT-PCR system (Invitrogen, Carlsbad, CA). The resultant cDNA was PCR amplified for pro-inflammatory cytokines by using human gene-specific sense and antisense primers based on sequences published in GenBank: IL-6 sense 5'-CCTGAACCTTCCAAAGATGGC-3', IL-6 antisense 5'-TTCACCAGGCAAGTCTCCTCA-3'; IL-8 sense 5'-ATGACTTCCAAGCTGGCCGTGGCT-3', IL-8 antisense 5'-TCTCAGCCCTCTTCAAAACTTCTC-3'; β-actin sense 5'-AAACTGGAACGGTGAAGGTG-3', β-actin antisense 5'-GTGGACTTGGGAGAGGACTG-3'; IL-1β sense 5'-TGGCAATGAGGATGACTTGT-3', IL-1β antisense 5'-TGGTGGTCGGAGATTCGTA-3'; IL-10 sense 5'-GACTTTAAGGGTTACCTGGGTG-3'; IL-10 antisense 5'-TCACATGCGCCTTGATGTCTG-3'. PCR was carried out on a LightCycler® 96 thermal cycler Instrument (Roche Applied Science, Penzberg, Germany). The program used for quantitative PCR amplification included a 5-min predenaturation step at 95 °C, a 3-step amplification at 95 °C for 10 s, 60 °C for 20 s (for 45 cycles), and a dissociation step (10 s at 98 °C, 60 s at 65 °C, and 1 s at 97 °C).

Results

1. Number of crawled literature

A total of 3,352 literature related to photobiomodulation (PBM group), 20,442 literature related to photodynamic therapy (PDT group), 5,874 LED-related literature (LED group), and 326,426 articles on laser-related were crawled. (laser group).

2. Gene identification

A total of 146 human genes related to PBM, 268 human genes related to PDT were identified, and the frequency of top10 genes was shown in **Figure 2a, 2b**. The 142 human genes related to LED and 3188 human genes related to laser were identified.

3. Confirm the genes in LED/laser-related phototherapy

In this experiment, the intersected genes of the LED group genes and (PBM + PDT) group genes were LED-related phototherapy. In the same way, the genes in laser-related phototherapy were confirmed. By using the Venn diagram, as shown in **Figure 3**, it was clear that there were 161 human genes related to LED, of which 50 genes were intersected with PBM, 54 genes were intersected with PDT, and 29 genes were repeated intersecting. The 3188 human genes related to laser light therapy, of which 138 genes were intersected with PBM, 141 genes were intersected with PDT, and 59 genes were repeated intersecting. In summary, by crawling the literature and identifying genes, a total of 75 phototherapy-related genes with LED and 220 phototherapy-related genes with laser were found. Moreover, the genes were deserved to be attention in the public intersection of LED, laser, PBM, and PDT. We proposed that the genes in the public intersection may be the key genes that play a role in phototherapy. For this reason, enrichment analysis was performed on these genes.

4. Comparison of PBM and PDT

In the R language, the clusterprofiler package was used to enrich functions and pathways. Among them, a total of 57 pathways in the PBM group were enriched, and the results of enrichment were shown in **Figure 4a, b**; a total of 109 pathways in the PDT group were enriched. The results of pathway and function enrichment were shown in **Figure 5a, b**; Compare the two results, a total of 45 pathways were in the intersection, which intersection is 78.9% of the KEGG result of PBM.

5. Comparison of LED and laser

By crawling the literature and identifying genes, a total of 75 phototherapy-related genes with LED and 220 phototherapy-related genes with laser were found. No KEGG result in the LED-related phototherapy, which was caused by the number of identified genes was not enough. A total of 104 pathways were enriched in the laser group, as shown in **Figure 6**. However, it can be known that 74 genes were intersected with laser in 75 LED-related phototherapy genes from the Venn diagram. The intersected genes account for up to 98.7% of the 91 LED genes. Therefore, it can be concluded that the genes affected by the LED in phototherapy are the same as the laser light source.

6. The 29 genes that were located in the common intersection of PBM, PDT, LED, and laser

Known from the Venn diagram that a total of 29 genes were a common intersection in these four groups, and so we speculated that these genes may be the keys in phototherapy. Therefore, we also performed KEGG and GO analysis for these 29 genes. A total of 7 pathways were enriched. The results were shown in **Figure 7a**. At the same time, we also performed a GO enrichment analysis for these genes, and a total of 365 results were enriched, as shown in **Figure 7b**. It was obvious that these genes were related to tyrosine, ultraviolet, light, and oxygen stress, etc.

7. Sequencing results of LED stimulate-MH7A

To explore the mechanisms of PBM for RA disease, we sequenced LED-MH7A cells. First, MH7A cells were stimulated with TNF- α . The experimental groups were irradiated with 630nm LED for 15min at room temperature, while control groups were wrapped with aluminum foil at room temperature. A total of 2950 differentially expressed genes were identified. 1885 and 1065 differentially expressed genes were up-regulated, down-regulated, respectively. The volcano is shown in **Figure 8**. KEGG enriched to 29 signal pathways, as shown in **Figure 9**. Among the 29 signaling pathways, 12 pathways were matched with those obtained by PBM analysis in this paper, which are Cytokine-cytokine receptor interaction, Legionellosis, Prion diseases, Viral protein interaction with cytokine and cytokine receptor, Complement and coagulation cascades, Bladder cancer, Fluid shear stress, and atherosclerosis, Prostate cancer, MAPK signaling pathway, TNF signaling pathway, IL-17 signaling pathway, Hepatitis B. All data, including sequencing reads and single-nucleus expression matrices have been deposited in NCBI's Gene Expression Omnibus (<http://www.ncbi.nlm.nih.gov/geo/>). The accession number for the mRNA-seq data reported in this study is GSE172223.

8. PPI network and cluster analysis

For further analysis, we got 12 pathways that were matched in the KEGG results of MH7A and the KEGG results of PBM. The pathways were Cytokine-cytokine receptor interaction, Legionellosis, Prion diseases, Viral protein interaction with cytokine and cytokine receptor, Complement and coagulation cascades, Bladder cancer, Fluid shear stress and atherosclerosis, Prostate cancer, MAPK signaling pathway, TNF signaling pathway, IL-17 signaling pathway, Hepatitis B. The genes were constructed a PPI network in 12 pathways. The outcomes of the string profile obtained from STRING showed that contained 129 nodes and 1235 edges. The top 10 outstanding proteins were identified as hub genes, which might play critical roles in the PBM through PPI analysis of STRING software including *IL6*, *TNF*, *VEGFA*, *IL10*, *EGFR*, *EGF*, *CCL2*, *MMP9*, *IL1B*, *FOS*. Among these significant proteins, IL6 was the most important protein and connected with 84 nodes in the network. A total of 6 clusters were generated in MCODE, and the top one cluster was selected as hub modules by the scores evaluated in MCODE. MCODE 1, which had 32 nodes and 369 edges, had the highest score in these clusters, such as **Figure 10**. In this cluster, the 9 nodes, including *IL6*, *VEGFA*, *CXCL2*, *CSF1P*, *ITGAM*, *TNF*, *FOS*, *MMP9*, *PGF* were in both MH7A cells and the PBM group. Maybe, these 9 nodes are key genes for PBM. On the one hand, in this cluster, we can know that some protein (red circles) of PBM results obtained by the method proposed in this paper already coincide with the parts of bio-MH7A cell sequencing. It proves the feasibility of our method. On the other hand, in LED stimulate-MH7A cells, the yellow circles, including *CSF3*, *CXCL3*, *SOCS3*, *HMOX1*, *HBEGF*, *IL1B*, *IL1A*, *PLAU*, and *CCL20*, represent the differentially expressed genes and have not been studied. Therefore, these genes have more research value and potential.

9. Effects of 630-nm LED radiation on mRNA gene expressions of inflammatory cytokines

For the accuracy of the PPI network and the effect of 640nm LED red light on rheumatoid disease, we performed RT-qPCR to analysis mRNA levels of inflammatory. The result was shown in Figure 11; there was a significant decrease in the gene expressions of inflammatory cytokines IL-6, IL-8, and IL-1B and MMP-3 expression; meanwhile, the expression of antiinflammatory factor IL-10 was promoted with the radiation doses increased. Collectively, these results demonstrate that the gene expression and production of inflammatory cytokines by MH7A cells can be inhibited by 630-nm LED radiation.

Discussion

Although PBM has great potential applications, the mechanisms are not fully elucidated. By crawling the abstracts from the literature, the genes that have been studied were identified. And the enrichment analysis of KEGG and GO were adopted to know which pathways and functions were involved. Currently, most of the research on PBM is focuses on the PI3K-Akt signaling pathway and rheumatoid arthritis disease. At the same time, in the KEGG results of the laser group, the PI3K-Akt signaling pathway and rheumatoid arthritis signaling pathway appear again, which suggests that PBM regulates the PI3K-Akt and rheumatoid arthritis signaling pathway to play a role. Possibly, we put forward some hypotheses based on these results. Through PI3K-Akt and MAPK signaling pathways, rheumatoid arthritis could be

improved by PBM. Coincidentally, one study has confirmed that 630nm LED can inhibit the proliferation of synovial fibroblasts in rheumatoid arthritis through the PI3K-Akt signaling pathway^[18].

The PI3K-Akt signaling pathway is one of the most investigated pathways in PBM. The p53 and PI3-Akt signaling pathways mediate the up-regulation of *GADD45* to induce human RPE cell apoptosis^[19]; low-level laser therapy induces the proliferation, migration, and tube formation of human umbilical vascular endothelial cells by activating the PI3K/Akt signaling pathway^[20]; LLLT reduces cytokines by inhibiting PI3K activation of protein kinase A16^[21]; LLLT (660 nm or 780 nm) can modify oral dysplastic cells (DOK) and oral cancer cells (SCC9 and SCC25) growth by modulating the Akt/mTOR/CyclinD1 signaling pathway^[8].

A large number of studies have demonstrated that the MAPK signaling pathway plays a key role in PBM. 450nm PBM controls the bacterial infection of periodontitis through ROS/MAPK/mTOR pathway^[22]. Studies have also shown that PBM can improve acute inflammation in mice through the cannabinoid receptor/ATP-sensitive K(+) pathway/P38-MAPK signaling pathway^[23]. Melatonin-treated cells were laser-irradiated, and the differentiation and mineralization of cells were found to involve p38 MAPK and PRKD1 signaling mechanisms^[24].

Crawling information through the Internet is a promising strategy. Georgia crawled obituaries online in 2015 to analyze the link between parity and cancer risk^[25]. In our study, we used web crawling to obtain genes for comparing two different phototherapy methods: PBM and PDT. The result was that 146 human genes related to PBM and 268 genes related to PDT. The proportion of gene intersection is 39.9%, and the proportion of KEGG of PBM and PDT is 78.9%. Therefore, it can be concluded that the two different phototherapy methods, PBM and PDT, may regulate the same pathway to play an important biological role.

Moreover, the light sources of LED and laser were compared to verify if there are significant differences. As a result, 75 phototherapy-related genes with LED and 220 phototherapy-related genes with laser were identified. The ratio of the two intersecting genes in LED genes is 98.9%, which means that most of the phototherapy related genes in LED are covered by the laser-related phototherapy genes. Therefore, it can also be proved that their effects have no significant differences in phototherapy. For the 75 LED genes related to phototherapy, there was no KEGG result caused by few identified genes.

We speculated that the 29 genes in the PBM, PDT, LED, and laser are the key genes for the role of phototherapy. Therefore, the enrichment analysis was performed on the 29 genes to explore the mechanism of phototherapy. The result of KEGG was shown to be related to Longevity regulation, Amphetamine addiction, Melanoma, etc. GO results show that these genes were related to tyrosine, ultraviolet, light, and oxygen stress, etc. On the one hand, the results of KEGG of these 29 genes were only 7 pathways caused by few genes; on the other hand, from GO results, the function of these genes can be understood to further clarify the role of phototherapy.

Due to the repeated occurrence of RA signaling pathways in our results, we also performed gene sequencing in MH7A cells under the conditions of whether there was light or not in TNF- α stimulation to explore the differentially expressed gene. Sequencing results showed that there were 2950 differential genes, and KEGG enriched 29 pathways. The top ten enriched pathways were Cytokine-cytokine receptor interaction, Neuroactive ligand-receptor interaction, Protein processing in the endoplasmic reticulum, Cell cycle, Viral protein interaction with cytokine and cytokine receptor, Complement and coagulation cascades, Legionellosis, DNA replication, Prion diseases, Bladder cancer. Among the 29 signaling pathways, 12 of them coincide with those obtained by PBM analysis, which are Cytokine-cytokine receptor interaction, Legionellosis, Prion diseases, Viral protein interaction with cytokine and cytokine receptor, Complement and coagulation cascades, Bladder cancer, Fluid shear stress, and atherosclerosis, Prostate cancer, MAPK signaling pathway, TNF signaling pathway, IL-17 signaling pathway, Hepatitis B. These 12 signaling pathways may be important in PBM.

A PPI network was constructed in the genes of 12 pathways to explore outstanding proteins. The network contained 129 nodes and 1235 edges. And we identified ten outstanding proteins in this network, which were *IL6*, *TNF*, *VEGFA*, *IL10*, *EGFR*, *EGF*, *CCL2*, *MMP9*, *IL1B*, *FOS*. And *IL6* was the most important that connected with 84 nodes in the network. In fact, in our previous study, we have demonstrated the effect of 630nm LED on MH7A cells, and the IL6 decreased after 630nm LED by ELISA experiment and q-PCR. In this network, there were six clusters generated and the cluster with the highest score was chosen. In this cluster, a total of nine proteins appeared in MH7A and PBM results, and the eight proteins appeared in only LED stimulate-MH7A cells. The nine proteins (red circles) had a very high degree, which proves the feasibility of our method. The eight proteins (yellow circles), including *CSF3*, *CXCL3*, *SOC3*, *HMOX1*, *HBEGF*, *IL1B*, *IL1A*, *PLAU*, *CCL20*, represent the part of differentially expressed genes in LED stimulate-MH7A. It means that they not have been studied and have research value and potential in PBM. For the accuracy of the PPI network and the effect of 640nm LED red light, the RT-qPCR was used to analysis mRNA levels of inflammatory. There was a significant decrease in the gene expressions of inflammatory cytokines IL-6, IL-8, and IL-1B and MMP-3 expression; meanwhile, the expression of antiinflammatory factor IL-10 was promoted with the radiation doses increased. These results demonstrate that the gene expression and production of inflammatory cytokines by MH7A cells can be inhibited by 630-nm LED radiation.

In this experiment, due to the limitations of the PubMed search engine, we tried to use different keywords to search for articles related to light therapy. Although a simple search will expand the number of literature, the accuracy will also decrease. However, according to the late start of LED research, the number of related literature was not much. To expand the number of literature, the simplest search formula was chosen: light-emitting diode. Accuracy was sacrificed, but in post-processing, the intersections of LED genes with those identified in PBM and PDT genes were studied. Therefore, the method can guarantee both quantity and accuracy.

In gene identification, we used the `gene_atomization` (a function) in the `pubmed.mineR` package(R language). But the function has one thing worth noting. In some cases, it recognizes the carbon element

3 as C3 but enters it as the gene: complement 3. Therefore, after we used the function to identify the human gene ID, the confounding genes need to be removed artificially. During manual processing, it was found that most of the confusing genes were 2-bit, such as those of PC and TG, which could be used to focus attention on this type of gene.

Conclusion

In summary, web crawling is a promising strategy to generate study hypotheses for guiding and prioritizing future studies. Used web crawling, bioinformatics analysis, and gene sequence, we identified 12 pathways and 10 genes that play important roles in PBM and predicted nine genes with research value. Anti-inflammatory effect of 630nm LED red light was demonstrated by RT-qPCR. Moreover, it was also verified that PBM and PDT, as well as LED and laser have no significant difference in effect on phototherapy.

List Of Abbreviations

PBM: photobiomodulation;

LED: ight-emitting diode;

LLLT: low-level laser therapy;

PDT: photodynamic therapy;

RT-qPCR: Real Time Quantitative PCR;

KEGG:Kyoto encyclopedia of genes and genomes;

GO: the gene ontology term

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding authors on reasonable request.

Competing interests

The authors declare that they have no competing interests

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Authors' contributions

Hao Wu and Caiyun Meng crawled literature contributed to data analysis and RT-qPCR. Hao Wu and Qing Xia wrote the manuscript. Wuqi Song designed the project. Wuqi Song and Fengmin Zhang reviewed the manuscript. Wuqi Song and Hailiang Liu revised the manuscript.

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Authors' information (optional)

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Figures

Figure 1

Data collection and analysis.

Figure 2

Frequency of the top 15 genes with the most occurrences in the PBM group(a) and the PDT group(b).

Figure 3

Venn diagram of each gene in every group. There are 161 human genes related to LED, among which 50 intersect with PBM, 54 intersect with PDT, and 29 intersect repeatedly. There are 3,188 human genes related to laser therapy, among which 138 intersect with PBM, 141 intersect with PDT, and 59 intersect repeatedly.

Figure 4

The top 20 KEGG results of the PBM group(a), the top 10 GO results for the PBM group(b). All KEGG and GO enrichment analysis was based on a threshold of p-value <0.05.

Figure 5

The top 20 KEGG results of PDT(a), top 10 GO results in PDT group(b). All KEGG pathway and GO enrichment analysis was based on a threshold of p-value <0.05.

Figure 6

The top 20 KEGG results of laser group. All KEGG pathway and GO enrichment analysis was based on a threshold of p-value <0.05.

Figure 7

KEGG results of 29 common intersection genes(a), the top ten GO results of 29 common intersection genes(b). All KEGG pathway and GO enrichment analysis was based on a threshold of p-value <0.05.

Figure 8

the volcano of differentially expressed genes in MH7A cells. A total of 2950 differentially expressed genes were identified. 1885 and 1065 differentially expressed genes are up-regulated, down-regulated, respectively. $P\text{-value} < 0.05$ and $|\log_2(\text{fold change (FC)})| > 1$, were chosen as the cut-off threshold.

Figure 9

The top 10 KEGG results of differentially expressed genes in MH7A cells. All KEGG analysis was based on a threshold of p-value <0.05.

Figure 10

The top one hub modules of the PPI network evaluated in MOCDE. MCODE1 contained 32 nodes and 359 edges. The blue circles represent PBM genes, the blue circles represent MH7A genes, and red circles represent the genes repeated PBM and MH7A. The circle size represents the degree.

Figure 11

The effects of 630-nm LED radiation on inflammatory cytokines in synoviocyte MH7A cells at varying light doses. RT-qPCR showed changes in the relative expression levels of IL-8(a), IL-6(b), MMP3(c), IL-1B(d), and IL10 (e) in MH7A cells.