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Ferroptosis-related genes are considered as potential targets for CPAP treatment of obstructive sleep apnea

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Obstructive sleep apnea (OSA) is a common syndrome characterized by upper airway dysfunction during sleep. Continuous positive airway pressure (CPAP) is the most frequently utilized non-surgical treatment for OSA. Ferroptosis play a crucial role in the physiological diseases caused by chronic intermittent hypoxia, but its involvement in the development of OSA and the exact mechanisms have incompletely elucidated. GSE75097 microarray dataset was used to identify differentially expressed genes between OSA patients and CPAP-treated OSA patients. Subsequently, Gene Ontology (GO) annotation, Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway, STRING database, and FerrDb database were conducted to analyze the biological functions of differentially expressed genes and screen ferroptosis-related genes. Finally, GSE135917 dataset employed for validation. There were 1,540 differentially expressed genes between OSA patients and CPAP-treated OSA patients. These differentially expressed genes were significantly enriched in the regulation of interleukin-1-mediated signaling pathway and ferroptosis-related signaling pathway. Subsequently, 13 ferroptosis-related genes (DRD5, TSC22D3, TFAP2A, STMN1, DDIT3, MYCN, ELAVL1, JUN, DUSP1, MIB1, PSAT1, LCE2C, and MIR27A) were identified from the interaction between differentially expressed genes and FerrDb database, which are regarded as the potential targets of CPAP-treated OSA. These ferroptosis-related genes were mainly involved in cell proliferation and apoptosis and MAPK signaling pathway. Furthermore, DRD5 and TFAP2A were downregulated in OSA patients, which showed good diagnostic properties for OSA, but these abnormal signatures are not reversed with short-term effective CPAP therapy. In summary, the identification of 13 ferroptosis-related genes as potential targets for the CPAP treatment of OSA provides valuable insights into the development of novel, reliable, and accurate therapeutic options.

KEYWORDS

obstructive sleep apnea, chronic intermittent hypoxia, continuous positive airway pressure, ferroptosis, bioinformatics analysis

1 Introduction

Obstructive sleep apnea (OSA) is a common sleep-disordered breathing disease, occurring in all age groups from early infancy through adolescence. The primary pathological features of OSA mainly involve sleep apnea, characterized by chronic intermittent hypoxia and sleep fragmentation, which can result in oxidative damage to brain, neuronal cell injury, and chronic inflammation, contributing to the development of complications in multi-organ and systems such as coronary heart disease, hypertension, and diabetes (1–3). The main treatment for OSA is continuous positive airway pressure (CPAP), which effectively alleviates drowsiness symptoms and improves the quality of life in patients with OSA (4, 5). However, the underlying mechanism of CPAP therapy for OSA remains elusive, as the pathogenesis of OSA involves multiple biological functions, including pyroptosis, autophagy, and ferroptosis (6–8). Therefore, it is crucial to explore the potential molecular targets of CPAP therapy for OSA to further examine the pathological mechanism of OSA and improve the clinical management of patients.

Ferroptosis is a newly regulated form of cell death characterized by iron-dependent accumulation of lipid peroxidation, resulting in intracellular reactive oxygen species (ROS) and abnormal lipid metabolism, ultimately leading to cell death (9, 10). Previous studies have demonstrated the pivotal role of ferroptosis in pathological processes such as cancer, respiratory disease, cerebral hemorrhage, myocardial infarction, and ischemic stroke (11). Activation or inhibition of ferroptosis can interfere with the development of diseases, so it is of great practical significance for clinical treatment of human diseases to explore the role of ferroptosis in various diseases by sorting out ferroptosis-related genes (12). Recent studies have found that serum ferritin levels tend to rise with increasing OSA severity (13). Chronic intermittent hypoxia is the signature manifestation of OSA, and intermittent hypoxia in the brain will lead to excessive accumulation of reactive oxygen species (ROS) in neurons, which may cause permanent death of neurons (14). Recent studies have shown that hypoxia can increase the iron content in the brain of animals leading to neurodegeneration, and disorder the metabolism of nerve cells by affecting the ferroptosis-related to genes, resulting in excess oxygen free radicals (15, 16). Simultaneously, Liu et al. (17) reported that two ferroptosis-related genes can serve as biomarkers for diagnosing OSA. Abnormal iron metabolism is associated with disease severity and poor oxygenation in patients with OSA (18). However, the specific regulatory mechanism of ferroptosis in the pathophysiological process of OSA and its clinical application still need to be further studied and explored.

Here, we innovatively identified the molecular targets of CPAP-treated OSA and explored the relationship between ferroptosis and CPAP therapy in order to enhance the understanding of the transcriptomic characteristics underlying CPAP treatment of OSA.

2 Methods

2.1 Data collection and preprocessing

The flow chart illustrating bioinformatics analysis is shown in Figure 1. The microarray datasets related to OSA (GSE75097 and GSE135917) were obtained from the Gene Expression Omnibus (GEO) database[1] (19, 20). As indicated in Table 1, the sequencing platform of GSE75097 is GPL10904, and GSE135917 is GPL6244. There are 28 OSA patients and 14 OSA patients undergoing CPAP treatment in GSE75097. Additionally, GSE135917 included 8 health subjects and 10 OSA patients, as well as 24 patients with OSA at baseline and 24 after exposure to CPAP. Peripheral blood mononuclear cells (PBMC) samples from GSE75097 were included for screening differentially expressed mRNAs (DEmRNAs) and ferroptosis-related genes. Meanwhile, GSE135917 was employed to validate the diagnostic value of ferroptosis-related genes. The raw data in cell format files were read using the "affy" package, followed by normalization of the raw data using the Affymetrix platform in the R package and data processing with RMA functions.

2.2 Screening of differentially expressed genes

The limma package was used to identify DEmRNAs, considering genes with a p value < 0.05 and $|\log_2\text{-fold change (FC)}| > 1.5$ as significant. Volcano plots generated using the "ggplot2" packages (version 3.3.3) of R software (version 3.6.3) were employed for visualizing the identified DEmRNAs.

2.3 Functional enrichment analysis

The Gene Ontology (GO) annotation and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment analyses of DEmRNAs were conducted using DAVID 6.8 (21) and Metascape[2] (22) $p < 0.05$ was applied for statistical analysis.

2.4 Protein–protein interaction network creation

Based on the online STRING database,[3] the interactions of DEmRNAs were predicted by constructing and analyzing PPI networks. In the PPI network, target genes are represented as nodes, while the lines between nodes depict related interactions. The color of these lines reflects the intensity of the interactions.

2.5 Identification of ferroptosis-related genes

We obtained ferroptosis-related genes from the FerrDb database,[4] encompassing "Driver," "Suppressor," and "Marker." Subsequently, Venn diagram was used to demonstrate the overlap between DEmRNAs and ferroptosis-related genes in the FerrDb database. GO and KEGG pathway analyses were conducted using the Metascape

1 https://www.ncbi.nlm.nih.gov/geo/
2 http://metascape.org
3 https://string-db.org/
4 http://www.zhounan.org/ferrdb/

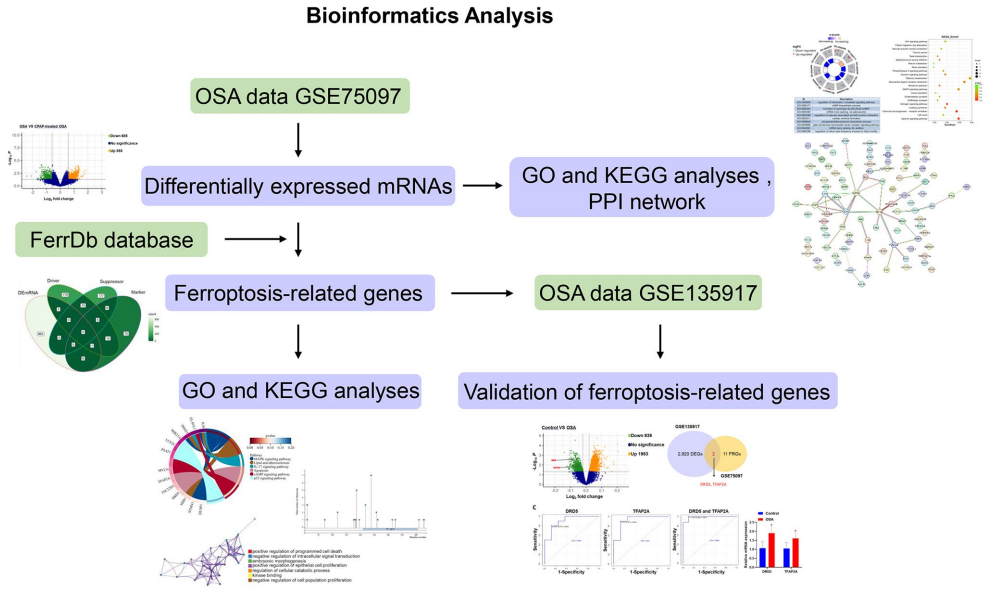


FIGURE 1
Flowchart of bioinformatics analysis.

TABLE 1 Detailed information of GSE75097 and GSE135917 datasets.

Accession	Platform	Sample	Normal	OSA	CPAP-OSA	Gene	Application
GSE75097	GPL10904	PBMC	0	28	14 (1 year)	mRNA	Training
GSE135917	GPL6244	Fat Tissue	8	10/24 (OSA at baseline)	24 (2 weeks)	mRNA	Verification

OSA, Obstructive sleep apnea; CPAP, Continuous positive airway pressure; the design of GSE135917: (1) 10 subjects with OSA vs. 8 controls; (2) 24 subjects with OSA at baseline vs. after effective CPAP therapy.

website and DAVID 6.8. PhosphoSitePlus[5] was performed to examine protein translational modifications (PTMs) of ferroptosis-related genes.

2.6 Validation of the identified ferroptosis-related genes

HiPlot software was applied to perform the receiver operating characteristic (ROC) curve analysis. SPSS 22.0 version was used for binary logistic regression to calculate the prediction probability of multiple genes in each sample, enabling multigene ROC analysis. The area under the ROC curve (AUC) was adopted as a quantitative measure to evaluate the results, with genes exhibiting an AUC > 0.8 considered to have exceptional diagnostic performance.

2.7 Quantitative real–time PCR

Three healthy subjects and three OSA patients were recruited from October 2023 to November 2023 at Shenzhen Children's Hospital Affiliated to Shantou University Medical College. The study was approved by the Ethics Committee of Shenzhen Children's Hospital Affiliated to Shantou University Medical College (202309102). Peripheral blood samples were collected for q-PCR detection. Total RNA was extracted from blood using TRNzol Universal Total RNA extraction reagent (TIANGEN, Beijing, China) according to manufacturer's instructions. The cDNA was synthesized using Hifair® III 1st Strand cDNA Synthesis SuperMix (YESEN, Shanghai, China). On an ABI Stepone plus PCR equipment, q-PCR was performed using Hieff® qPCR SYBR Green Master Mix (YESEN, Shanghai, China). The following were the primers utilized in this study: DRD5-Forward: GGGCAGTTCGCTCTATACCAG; DRD5-Reverse: GGTCCAGAT GATGAGTAGGGTC; TFAP2A-Forward: AGGTCAATCTCCCTA CACGAG; TFAP2A-Reverse: GGAGTAAGGATCTTGCGACTGG; GAPDH-Forward: TGGAAATCCCATCACCATCT; and GAPDH-Reverse: TGGACTCCACGACGTACTCA. Relative quantification was determined using the $2^{-\Delta\Delta Ct}$ method.

2.8 Statistical analysis

Data were expressed as mean ± standard deviation (SD) and the Graph Pad 8.0 were performed for analysis in this study. The Student's *t*-test was utilized to determine the difference between the two groups. $p < 0.05$ was considered significant.

5 https://www.phosphosite.org/homeAction

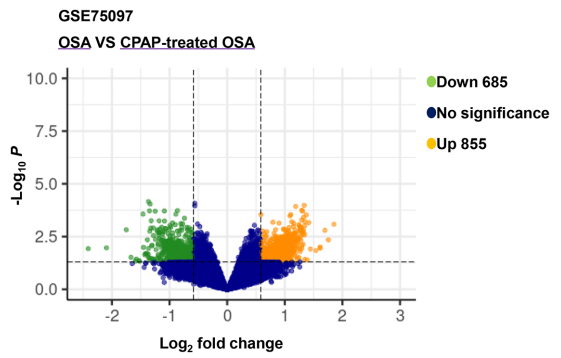


FIGURE 2
Volcano plot of differentially expressed genes between OSA and CPAP-treated OSA in GSE75097.

TABLE 2 The top five of upregulated genes and downregulated genes.

Gene	logFC	*p* value	Up/Down
LOC401131	1.336715442	0.000105821	Up
LOC643324	1.189998627	0.0001183	Up
TNR	1.297333582	0.00017904	Up
LOC645726	1.307407514	0.000198165	Up
HIRIP3	0.587443645	0.000285797	Up
RNU4ATAC	−1.366824666	0.000069506	Down
OXGR1	−1.336719153	0.000090044	Down
JUNB	−0.862500468	0.000185445	Down
LOC646513	−1.092160889	0.000192185	Down
RNU11	−1.348415132	0.000192794	Down

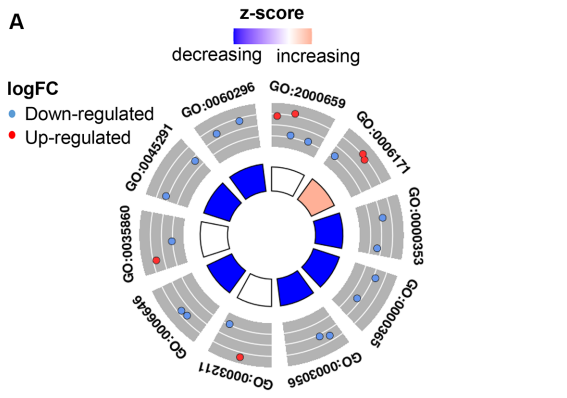
3 Results

3.1 Identification of DEmRNAs between OSA and CPAP-treated OSA

After analyzing the expression profiling data from GSE75097, a total of 1,540 DEmRNAs, including 685 downregulated and 855 upregulated genes, were selected from OSA and CPAP-treated OSA by applying screening criteria of $|\log_2(FC)| > 1.5$ and value of $p < 0.05$ (Figure 2). Additionally, Table 2 presented the top five upregulated genes and the top five downregulated genes, which included LOC401131, LOC643324, TNR, LOC645726, HIRIP3, RNU4ATAC, OXGR1, JUNB, LOC646513, and RNU11.

3.2 GO and KEGG enrichment analysis for DEmRNAs

To further understand the biological function of DEmRNAs, we conducted functional enrichment analysis. The results of GO terms enrichment analysis indicated that the genes significantly enriched were involved in "regulation of interleukin-1-mediated signaling pathway," "cAMP biosynthetic process," "regulation of vascular associated smooth muscle contraction," and "phosphatidylethanolamine biosynthetic process" (Figure 3A). KEGG pathways revealed that DEmRNAs mainly involved the "Wnt signaling pathway," "MAPK signaling pathway," "Calcium signaling pathway," and "Vascular smooth muscle contraction" (Figure 3B). Therefore, it can be reasonably speculated that biological functions mediated by DEmRNAs are involved in the physiological processes of OSA treated with CPAP.



ID	Description
GO:2000659	regulation of interleukin-1-mediated signaling pathway
GO:0006171	cAMP biosynthetic process
GO:0000353	formation of quadruple SL/U4/U5/U6 snRNP
GO:0000365	mRNA trans splicing, via spliceosome
GO:0003056	regulation of vascular associated smooth muscle contraction
GO:0003211	cardiac ventricle formation
GO:0006646	phosphatidylethanolamine biosynthetic process
GO:0035860	glial cell-derived neurotrophic factor receptor signaling pathway
GO:0045291	mRNA trans splicing, SL addition
GO:0060296	regulation of cilium beat frequency involved in ciliary motility

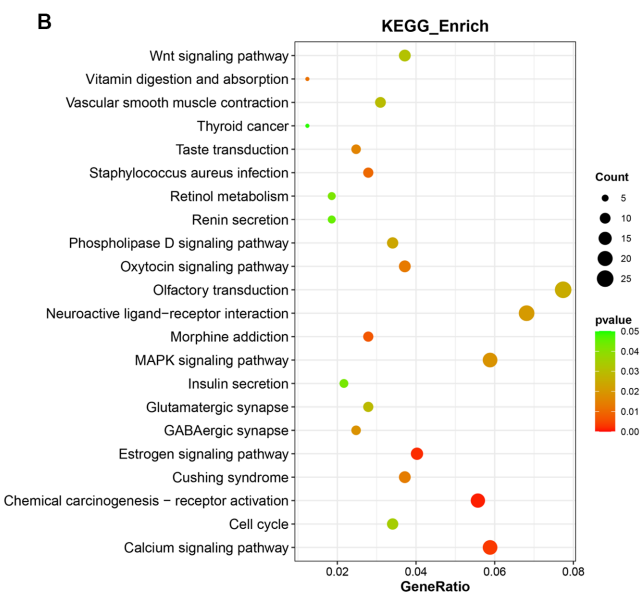


FIGURE 3
Functional analysis of differentially expressed genes. **(A)** Differentially expressed genes-enriched GO categories. **(B)** KEGG pathways analysis of differentially expressed genes.

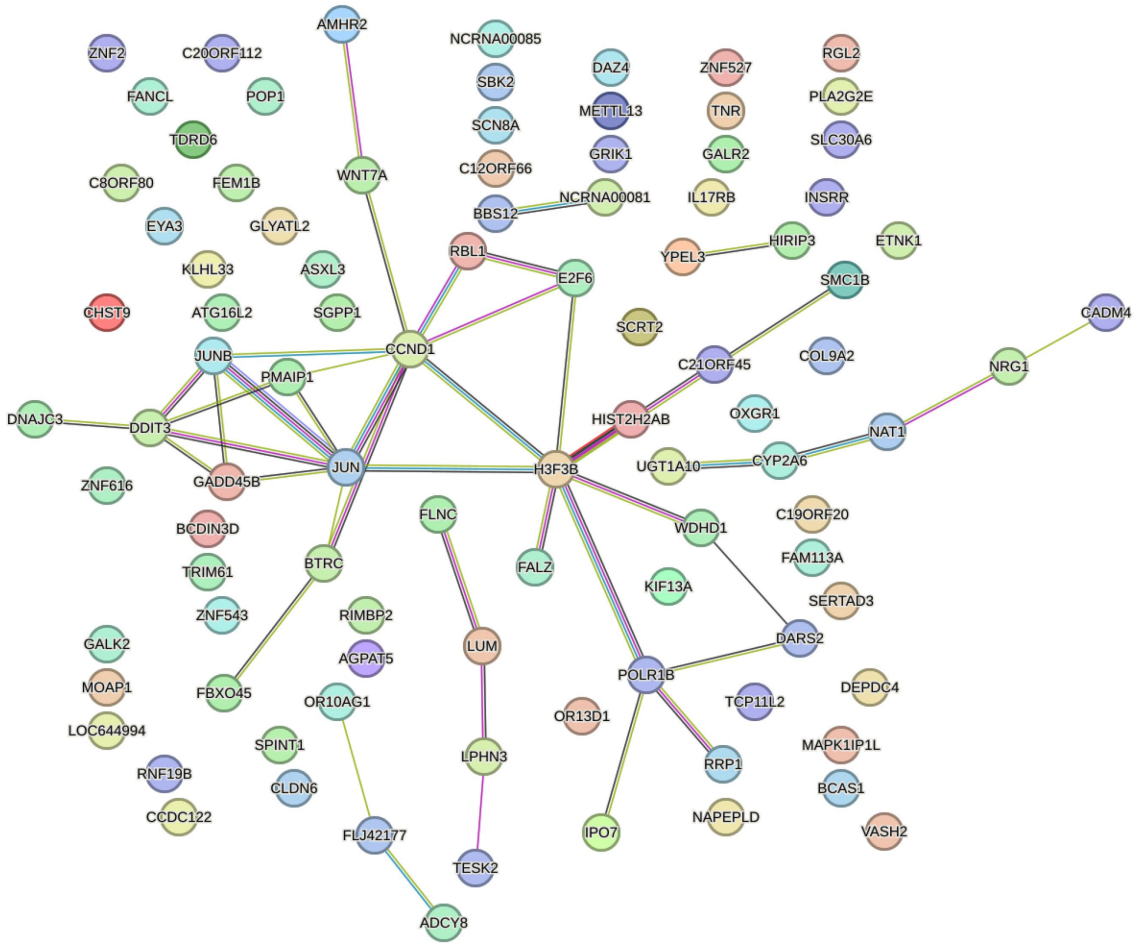


FIGURE 4
STRING database was used to construct PPI network.

3.3 PPI network analysis of DEmRNAs

The PPI network demonstrated the interaction between DEmRNAs. We constructed the PPI network to distinguish the connections among the top 200 DEmRNAs above. As depicted in Figure 4, the network consisted of 96 nodes and 44 edges, with an average local clustering coefficient is 0.244. The PPI network diagram showed JUN, CCDN1, and H3-3B were the most interacting proteins.

3.4 Identification of ferroptosis-related genes, biological function analysis, and PTMs sites prediction

Our study found that DEmRNAs are significantly enriched in the WNT signaling pathway. Some studies have reported activation of Wnt/beta-catenin signaling attenuated intracellular lipid ROS production, thereby inhibiting ferroptosis in gastric cancer cells (23). To investigate the association between DEmRNAs and ferroptosis, we further screened differentially ferroptosis-related genes according to the FerrDb database and DEmRNAs. Venn diagram analysis of the FerrDb database (included "Drivers," "Suppressors," and "Makers") and DEmRNAs identified the following genes which were differentially expressed in ferroptosis: DRD5, TSC22D3, TFAP2A, STMN1, DDIT3, MYCN, ELAVL1, JUN, DUSP1, MIB1, PSAT1, LCE2C, and MIR27A (Figure 5A; Table 3). Subsequently, these ferroptosis-related genes were uploaded to Metascape website and DAVID database, and it was shown that the biological pathways were remarkably enriched involvement of ferroptosis-associated pathways, such as MAPK signaling pathway, Lipid and atherosclerosis pathway, and IL-17 signaling pathway (Figure 5B). Additionally, GO analysis indicated that these ferroptosis-related genes regulate cell proliferation and death (Figure 5C). Besides, we predicted the PTMs sites of genes associated with ferroptosis. Except for DRD5, LEC2C, and MIR27A, PTMs were found in all genes (Supplementary Figures S1, S2), which is crucial for us to understand cellular signaling pathways. Taken together, we screened 13 ferroptosis-related genes that are considered as therapeutic targets for CPAP-treated OSA.

3.5 DRD5 and TFAP2A were key genes in the pathogenesis of OSA

Obesity is a significant risk factor for developing OSA patients, and the interaction between the respiratory system and adipose tissue plays a crucial role in the preventing and treatment of OSA (24, 25). Compared to control patients, OSA patients exhibited 939 downregulated genes and 1,983 upregulated genes (Figure 6A).

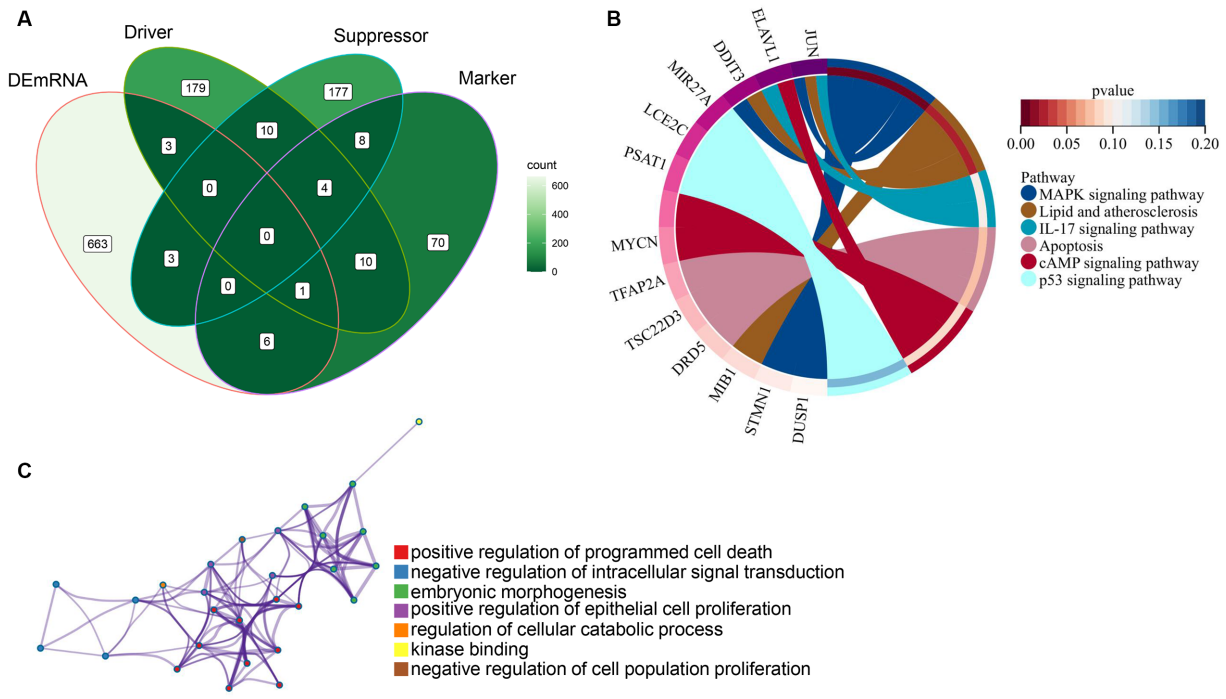


FIGURE 5
Identification and functional enrichment analysis of ferroptosis-related genes. **(A)** Ferroptosis-related genes were obtained from the intersection of the FerrDb database with differentially expressed genes. **(B)** KEGG pathways analysis of ferroptosis-related genes using the Metascape website and DAVID database. **(C)** GO categories of ferroptosis-related genes using the Metascape website.

TABLE 3 Details of 13 ferroptosis-related genes.

Gene	logFC	*p* value	Up/Down
DDIT3	−0.7886347	0.001624304	Down
DRD5	0.7604257	0.034925645	Up
DUSP1	−0.6341032	0.013720922	Down
ELAVL1	0.9008274	0.021881973	Up
JUN	−1.2806116	0.002822219	Down
LCE2C	0.8773638	0.016244034	Up
MIB1	0.6993263	0.031003364	Up
MIR27A	0.8125076	0.010206382	Up
MYCN	−0.6484463	0.044747900	Down
PSAT1	1.0379086	0.011651924	Up
STMN1	0.6377915	0.015467237	Up
TFAP2A	1.1186158	0.046086407	Up
TSC22D3	−0.6130752	0.015658925	Down

Interestingly, two of these genes overlapped with 13 ferroptosis-related genes obtained as described above, namely, DRD5 and TFAP2A (Figure 6B). Therefore, we speculated that DRD5 and TFAP2A are not only potential targets for CPAP treatment of OSA, but also key genes in OSA pathogenesis. Subsequently, ROC curves were generated to assess the diagnostic value of DRD5 and TFAP2A. The results demonstrated that both DRD5 (AUC = 0.900) and TFAP2A (AUC = 0.925), as well as their combination (AUC = 0.988), held significant potential for diagnosing OSA patients (Figure 6C). GSE75097 revealed an increase in the expressions of DRD5 and TFAP2A in PBMC samples from CPAP-treated OSA patients compared to those with OSA (Table 3). However, CPAP therapy did not improve the abnormal expression of DRD5 and TFAP2A in adipose tissue among OSA patients in the GSE135917 dataset (Supplementary Table S1). We thought that this may be due to the long duration of CPAP treatment (1 year) in GSE75097 (19), as opposed to 2 weeks in GSE135917 (20), which could account for these abnormal expression of DRD5 and TFAP2A was not improved with effective but short-term CPAP therapy.

In addition, in order to further explore the clinical expression of DRD5 and TFAP2A, we collected peripheral blood samples from healthy subjects and OSA patients for examination (due to limited clinical resources, we do not obtain the OSA subject before CPAP and OSA patient with long-term CPAP treatment). Compared with healthy subjects, the expression of DRD5 and TFAP2A in OSA patients was significantly upregulated (Supplementary Figure S3, $^*p < 0.05$), which was contrary to the results in Figure 6A. We analyzed the reasons for this: (1) there were too few clinical samples, so more clinical samples needed to be included to verify the reliability of the results; (2) Since the risk factors of OSA pathogenesis is related to age, gender, and obesity (26), it is necessary to consider these clinical factors in the included samples to ensure the correctness of the results. Therefore, we will include more clinical trials for verification in the future, and explore the potential molecular mechanism *in vivo* and *in vitro* experiments.

Collectively, these results indicated that DRD5 and TFAP2A, as potential targets of CPAP for OSA treatment, were also key genes in the pathogenesis of OSA, and had good diagnostic value. Nevertheless, due to the complex relationship between the pathogenesis of OSA and age, gender, and obesity, and the relationship between OSA treatment and these risk factors is still unclear, we still need to further explore the deeper molecular mechanism through *in vitro* and *in vivo* experiments, and include more detailed clinical samples to verify from different perspectives.

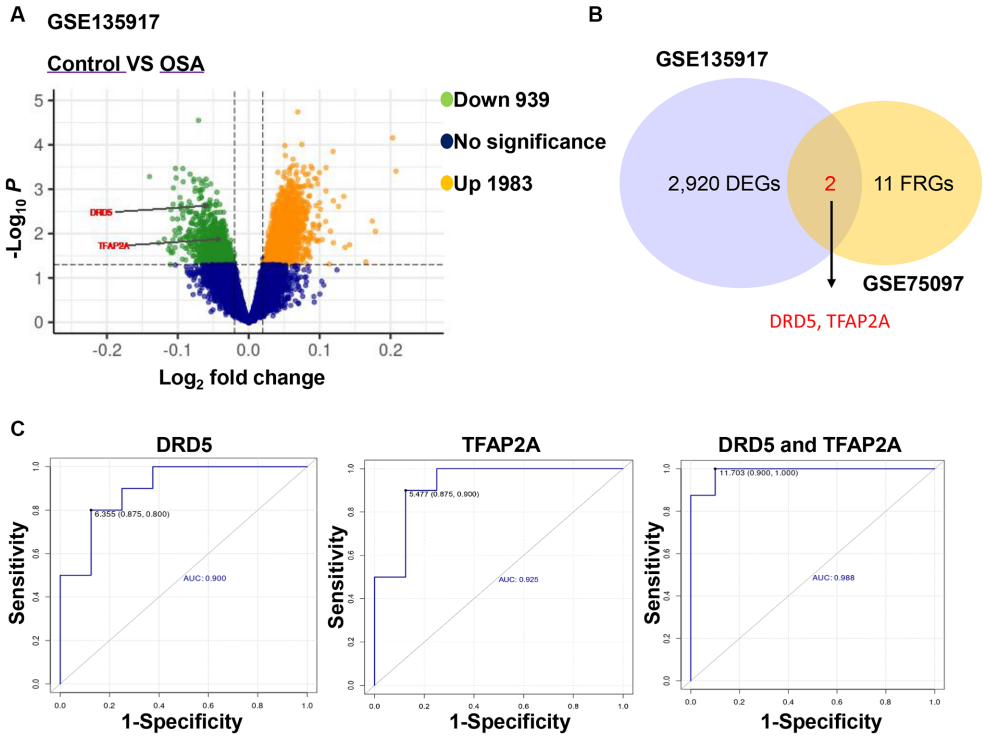


FIGURE 6
DRD5 and TFAP2A had the diagnostic value for OSA. **(A)** Volcano plot of differentially expressed genes between healthy samples and OSA in GSE135917. **(B)** Venn diagram showed the overlapping genes of 13 ferroptosis-related genes and differentially expressed genes of GSE135917. DEGs indicates differentially expressed genes and FRGs ferroptosis-related genes. **(C)** The diagnostic ROC curves of DRD5 and TFAP2A in OSA and healthy samples.

4 Discussion

Continuous positive airway pressure is the primary treatment for OSA, but due to limited data on molecular and pathological mechanisms, the interaction of target genes induced by CPAP therapy in OSA patients is unknown. The latest research confirms that serum ferritin levels tend to rise with increasing OSA severity (13). Besides, chronic intermittent hypoxia is the main pathophysiological mechanism of OSA. Since chronic intermittent hypoxia can trigger an increase in intracellular ROS, it may cause cellular ferroptosis (27). Previous studies have shown that ferroptosis plays an important role in chronic intermittent hypoxia-induced liver and heart damage (8, 28). Here, we screened 13 ferroptosis-related genes as treatment targets for CPAP-treated OSA, and the ROC of combined DRD5 and TFAP2A was 0.988, which had the diagnostic value for OSA.

Currently, 2–4% of adults suffer from OSA (29), and its prevalence has increased with the obesity epidemic (30). Clinically, CPAP is a common means of effective treatment of OSA, improving clinical symptoms in patients with OSA, such as excessive sleepiness and improved sleep therapy (31, 32), CPAP can also reduce cardiovascular risk in moderate-to-severe OSA patients by reducing circulating markers of cardiovascular risk (33, 34) and blood pressure (35). However, how CPAP improves OSA at the molecular level remains unknown. In our study, CPAP-treated OSA patients had 1,540 differentially expressed genes in their blood compared to OSA patients, with 685 downregulated and 855 upregulated genes, indicating CPAP treated OSA patients by regulating transcriptional signatures. Moreover, these DEmRNAs were mainly enriched in "Wnt signaling pathway," "MAPK signaling pathway," and "Calcium signaling pathway." Abnormal Wnt/β-catenin signaling pathway caused by chronic intermittent hypoxia, which is the most typical pathophysiological component of OSA (36). Meanwhile, Wang et al. (37) demonstrated that chronic intermittent hypoxia-mediated MAPK signaling pathway disturbed insulin secretion, which may be of important meaning for the clinical treatment of OSA. Calcium (Ca^{2+}) and ROS are multifunctional signaling molecules that coordinate physiological and pathophysiological processes (38), thus maintaining Ca^{2+} and ROS homeostasis may be critical for the clinical treatment of OSA. Therefore, these pathways may contain potential markers and future drug intervention targets of OSA.

Chronic intermittent hypoxia is the main clinical feature of OSA patients. Chen et al. (39) reported chronic intermittent hypoxia induced intracellular ROS accumulation, which leads to ferroptosis. Simultaneously, DEmRNAs-enriched signaling pathways (such as Wnt signaling pathway, MAPK signaling pathway, and Calcium signaling pathway) are also involved in the regulation of ferroptosis (23, 40, 41), so we speculated that ferroptosis-related genes are the molecular targets of CPAP treatment of OSA. Finally, we screened 13 ferroptosis-related genes (DRD5, TSC22D3, TFAP2A, STMN1, DDIT3, MYCN, ELAVL1, JUN, DUSP1, MIB1, PSAT1, LCE2C, and MIR27A) as potential targets for improving OSA with CPAP. DRD5 is a biomarker associated with ferroptosis that can be used for disease diagnosis and treatment monitoring in breast cancer (42). In addition, the production of mitochondrial ROS was increased in the renal cortex of DRD5 knockout mice (43). Furthermore, TFAP2A was upregulated in gallbladder carcinoma, promoting the increase of Fe^{2+} and malondialdehyde levels, and TFAP2A silencing attenuated the expression of key genes associated

with oxidative stress (44). Our study found that DRD5 and TFAP2A expressions were upregulated in OSA patients after CPAP treatment, suggesting that CPAP may improve abnormal ROS levels and inhibited chronic intermittent hypoxia-caused ferroptosis caused by targeting elevating the expressions of DRD5 and TFAP2A. In addition, DUSP1 is a hub-gene shared of OSA and Alzheimer Disease (AD) patients, and DUSP1-mediated oxidative stress involved in the pathogenesis of OSA affecting AD (45). DUSP1 gene expression was inhibited after CPAP treatment for OSA (46), which is similar to our findings. Besides, DDIT3 has been shown to be a diagnostic target for patients with AD (47). Considering that OSA-mediated chronic intermittent hypoxia affected the pathogenesis of AD, DDIT3 may also be a key gene in ameliorating OSA-associated complications. These results were helpful for the diagnosis and treatment of OSA, and may lay the foundation for further investigation of the molecular mechanism of CPAP treatment of OSA.

5 Conclusion

In summary, we identified 13 ferroptosis-related genes (DRD5, TSC22D3, TFAP2A, STMN1, DDIT3, MYCN, ELAVL1, JUN, DUSP1, MIB1, PSAT1, LCE2C, and MIR27A) as the target genes induced by CPAP therapy (1 year) in the patient with OSA. Moreover, DRD5 and TFAP2A were key genes in the pathogenesis of OSA, which showed good diagnostic properties for OSA, but these abnormal expression are not reversed with short-term effective CPAP therapy (2 weeks). These results provided a reference for further research on the therapeutic mechanism and develop on drug intervention targets of OSA.

6 Limitations

There are some limitations to our study. The sample size of our screened dataset is relatively small, and our discoveries need to be validated in a larger sample. Second, we demonstrated that ferroptosis-related genes (DRD5 and TFAP2A) exhibited good diagnostic properties in OSA patients. However, considering that the CPAP intervention group in the GSE135917 dataset may not be sufficient to completely eliminate OSA-related abnormal transcriptional signatures due to the short duration of CPAP treatment. Besides, OSA pathogenesis-related risk factors such as age, gender, and obesity should be considered in the included clinical samples to better explain the representative genes of OSA pathogenesis and explore effective targets for CPAP therapy. Therefore, based on these limitations, we will continue to explore the molecular mechanism of CPAP therapy for OSA *in vivo* and *in vitro* experiments.

Data availability statement

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and accession number(s) can be found in the article/Supplementary material.

Ethics statement

The studies involving humans were approved by the Ethics Committee of Shenzhen Children's Hospital Affiliated to Shantou University Medical College (202309102). The studies were conducted in accordance with the local legislation and institutional requirements. Written informed consent for participation in this study was provided by the participants' legal guardians/next of kin.

Author contributions

JH: Conceptualization, Writing – original draft. HZ: Conceptualization, Writing – original draft. LC: Investigation, Software, Writing – review & editing. FC: Methodology, Writing – review & editing. WL: Data curation, Writing – review & editing. QL: Data curation, Writing – review & editing. XH: Data curation, Writing – review & editing. QW: Methodology, Writing – review & editing. QY: Conceptualization, Funding acquisition, Project administration, Writing – review & editing.

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Conflict of interest

HZ, LC, FC, and QW were employed by Shenzhen Nuclear Gene Technology Co., Ltd.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's note

Supplementary material

The Supplementary material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fneur.2023.1320954/full#supplementary-material

SUPPLEMENTARY FIGURE S1
PhosphoSitePlus lollipop plots of 6 ferroptosis-related genes. Circles 362 indicate PTM sites with a height reflecting the number of references describing the 363 site.

SUPPLEMENTARY FIGURE S2
PhosphoSitePlus lollipop plots of 4 ferroptosis-related genes. Circles indicate PTM sites with a height reflecting the number of references describing the site.

SUPPLEMENTARY FIGURE S3
Clinical validation of DRD5 and TFAP2A expression (*$P < 0.05$, $n = 3$).

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