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RESEARCH ARTICLE

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Bioinformatics analysis to identify action targets in NCI-N87 gastric cancer cells exposed to quercetin

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ABSTRACT

Context: Quercetin exerts antiproliferative effects on gastric cancer. However, its mechanisms of action on gastric cancer have not been comprehensively revealed.

Objective: We investigated the mechanisms of action of quercetin against gastric cancer cells.

Materials and methods: Human NCI-N87 gastric cancer cells were treated with 15 μM quercetin or dimethyl sulfoxide (as a control) for 48 h. DNA isolated from cells was sequenced on a HiSeq 2500, and the data were used to identify differentially expressed genes (DEGs) between groups. Then, enrichment analyses were performed for DEGs and a protein–protein interaction (PPI) network was constructed. Finally, the transcription factors (TFs)-DEGs regulatory network was visualized by Cytoscape software.

Results: A total of 121 DEGs were identified in the quercetin group. In the PPI network, Fos proto-oncogene (FOS, degree = 12), aryl hydrocarbon receptor (AHR, degree = 12), Jun proto-oncogene (JUN, degree = 11), and cytochrome P450 family 1 subfamily A member 1 (CYP1A1, degree = 11) with higher degrees highly interconnected with other proteins. Of the 5 TF-DEGs, early growth response 1 (*EGR1*), FOS like 1 (*FOSL1*), *FOS*, and *JUN* were upregulated, while *AHR* was downregulated. Moreover, *FOSL1*, *JUN*, and Wnt family member 7B (*WNT7B*) were enriched in the Wnt signaling pathway.

Discussion and conclusions: CYP1A1 highly interconnected with AHR in the PPI network. Therefore, *FOS*, *AHR*, *JUN*, *CYP1A1*, *EGR1*, *FOSL1*, and *WNT7B* might be targets of quercetin in gastric cancer.

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Differentially expressed genes; protein–protein interaction; transcriptional regulatory network

Introduction

Gastric/stomach cancer is a type of cancer that originates from the lining of the stomach (Piazuelo and Correa 2013). Its symptoms mainly include loss of appetite, heartburn, nausea, and upper abdominal pain in the early stages and weight loss, difficulty in swallowing, vomiting, and hematochezia in the later stages (Orditura et al. 2014). The major causes of gastric cancer are *Helicobacter pylori* infection, smoking, and dietary and genetic factors (González et al. 2013; Yang et al. 2014). Gastric cancer is more common in men (Jemal et al. 2015), suggesting that estrogen in women may confer protection from the disease (Jian et al. 2014). Gastric cancer accounts for 8.5% of all cancer cases in men, making it the fourth most common cancer in men in 2012 (Lozano et al. 2012). In 2012, there were 952,000 newly diagnosed cases of gastric cancer, and it was the fifth most common cancer globally (Peto et al. 2014). Therefore, investigating the pathological mechanisms of gastric cancer is of great significance.

As a natural ingredient abundant in grapes and red wine, quercetin plays antiproliferative roles in multiple malignant cell types (Russo et al. 2014). Previous studies have indicated that quercetin exerts antiproliferative effects on gastric cancer cells by induction of apoptosis and inhibition of telomerase activity (Wei et al. 2007; Borska et al. 2012). It was demonstrated that quercetin contributes to the apoptosis of BGC-823 gastric carcinoma cells through mitochondrial pathways (Wang et al. 2012). In 2011, it was revealed that quercetin can activate autophagy in gastric cancer cells via regulating hypoxia-inducible factor-1α and Akt-mammalian target of rapamycin signaling (Wang et al. 2011). It was reported that quercetin can promote the apoptosis of BGC-823 cells and arrest the cell cycle at S-phase by inhibiting the expression of proliferating cell nuclear antigen and *p53* (Xiang et al. 2006). Nevertheless, the mechanisms of action of quercetin against gastric cancer have not been comprehensively revealed.

Bioinformatics is an interdisciplinary field that develops methods and software tools for understanding biological data, which combines Computer Science, Biology, Mathematics, and Engineering to analyze and interpret biological data (Saeys et al. 2007). The results of bioinformatics will provide a scientific guidance for future study and increase the understanding of biological processes for quercetin against gastric cancer cells. Protein–protein interaction networks (PPIs) are the networks of

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Supplemental data for this article can be accessed here.

protein complexes formed by biochemical events and/or electrostatic forces and that serve a distinct biological function as a complex. The protein interactome describes the full repertoire of a biological system's PPIs (Kumar et al. 2017). In addition, the regulatory interactions between transcription factors and their target genes display a scale-free topology and indicate the presence of regulatory hubs (Babu et al. 2004).

In the current study, we sequenced DNA from human NCI-N87 gastric cancer cells treated with quercetin versus controls and screened for differentially expressed genes (DEGs), followed by enrichment analysis and construction of PPI and transcriptional regulatory networks.

Materials and methods

Cell culture and quercetin treatment

Human NCI-N87 gastric cancer cell lines were purchased from the Cell Bank of the Chinese Academy of Sciences (Shanghai, China). NCI-N87 cells were cultured in a mixture of 1% penicillin–streptomycin (Gibco, Grand Island, NY), 10% fetal bovine serum (Gibco), and Roswell Park Memorial Institute-1640 medium (Gibco) in an incubator at 37 °C with 5% CO_2. When the cells reached 80%–90% confluence, they were passaged with 0.25% trypsin (Gibco) The cells were then centrifuged and replaced with fresh medium on new Petri dishes. After counting, cells were seeded on Petri dishes (diameter: 6 cm) at a density of 2×10^6 cell/dish and cultured in 5 mL serum-free medium in an incubator at 37 °C with 5% CO_2 overnight. The next day, cells in the quercetin group were treated with 15 μM quercetin (Sigma, St. Louis, MO) for 48 h (Sekiguchi et al. 2008), whereas cells in the control group were treated with the same volume of dimethyl sulfoxide (Sigma).

RNA extraction and RNA-sequencing library construction

Total RNA was isolated from cells using TRIzol® (Invitrogen, Burlington, Ontario, Canada) according to the manufacturer's instructions; RNA integrity and purity were separately determined by 2% agarose gel electrophoresis and spectrophotometry. RNA-sequencing libraries were constructed using a NEBNext® Ultra™ RNA Library Prep Kit for Illumina® (NEB #E7530, New England Biolabs, Ipswich, MA) following the manufacturer's instructions. First, mRNA was isolated and broken into fragments of about 200 nucleotide (nt). Then, double-stranded cDNA was synthesized and amplified by polymerase chain reaction to construct the cDNA library. The quality of the cDNA library was evaluated on a Bioanalyzer 2100 (Agilent Technologies, Palo Alto, CA), and sequencing was performed on a HiSeq 2500 (Illumina, San Diego, CA). Sequencing data were uploaded to the National Center for Biotechnology Information Sequence Read Archive database (accession no. SRP091839).

Data preprocessing and DEG screening

Using the FASTX-Toolkit (version 0.0.13, http://hannonlab.cshl.edu/fastx_toolkit/) (Krueger et al. 2012), quality control was performed on sequencing data. After adapter removal, bases with a quality lower than 10 were eliminated, and then, reads larger than 50 nt were reserved. Reads with more than 80% bases having a quality greater than 20 were considered as clean reads. Using Top Hat software (Kim et al. 2013), clean reads were mapped to the hg19 human genome, allowing 2 mismatches. Based on annotation files of the hg19 human genome, gene expression values were calculated by Cufflinks software (http://cole-trapnell-lab.github.io/cufflinks/) (Ghosh and Chan 2016). The cuffmerge tool (Trapnell et al. 2010) in Cufflinks was utilized to integrate the gene expression values in different samples. Then, DEGs between quercetin and control groups were selected by the Cuffdiff tool (Trapnell et al. 2013) in Cufflinks. $p < 0.05$ was selected as the threshold.

Functional and pathway enrichment analysis

The Gene Ontology (http://www.geneontology.org) database aims to describe cellular components (CC), molecular functions (MF), and biological processes (BP) related to gene products (Tweedie et al. 2009). The Kyoto Encyclopedia of Genes and Genomes (http://www.genome.ad.jp/kegg) database is used for pathway analysis of genes or other molecules (Kanehisa and Goto 2000). Using the clusterProfiler package in R (http://bioconductor.org/packages/release/bioc/html/clusterProfiler.html) (Yu et al. 2012), functional and pathway enrichment analyses were separately conducted for both the upregulated and downregulated genes. Terms with $p < 0.05$ were considered to be significantly enriched.

PPI network analysis

The Search Tool for the Retrieval of Interacting Genes (http://string-db.org/) database includes direct and indirect PPIs in more than 1100 organisms (Franceschini et al. 2012). Using this database (Franceschini et al. 2012), PPIs among the identified DEGs were predicted, with a combined score >0.4 as the threshold. Subsequently, Cytoscape software (http://www.cytoscape.org) (Saito et al. 2012) was used to visualize the PPI network.

Transcriptional regulatory network analysis

Using the TRANSFAC® database (http://www.gene-regulation.com/pub/databases.html) (Matys 2006), transcription factors (TF) among the identified DEGs were screened. TF-DEG pairs were predicted using information on TF-binding sites obtained in the University of California-Santa Cruz genome browser database (http://genome.ucsc.edu) (Speir et al. 2015). Then, the TF-DEG regulatory network was constructed with Cytoscape software (Saito et al. 2012). Enrichment analysis was also performed on genes involved in the regulatory network using the clusterProfiler package in *R* (Yu et al. 2012), with $p < 0.05$ as the cutoff.

Results

DEG analysis

A total of 121 DEGs were selected between the quercetin and control groups, including 50 upregulated (e.g., early growth response 1 (*EGR1*), FOS like 1 (*FOSL1*), Fos proto-oncogene (*FOS*), and Jun proto-oncogene (*JUN*)) and 71 downregulated genes (e.g., aryl hydrocarbon receptor (*AHR*)) (Supplementary Table 1).

Functional and pathway enrichment analyses

Upregulated genes were mainly enriched within the apical plasma membrane (CC, $P = 8.49E-03$) in response to cAMP

Table 1. The top 5 functions and pathways enriched for the up-regulated genes.

Category	Description	*p* Value	Gene number	Gene symbol
GO_BP	GO:0051591~response to cAMP	3.54E-09	6	*DUSP1, EGR1, ALDH3A1, FOS, JUN, FOSL1*
GO_BP	GO:0046683~response to organophosphorus	2.12E-08	6	*DUSP1, EGR1, ALDH3A1, FOS, JUN, FOSL1*
GO_BP	GO:0014074~response to purine-containing compound	4.49E-08	6	*DUSP1, EGR1, ALDH3A1, FOS, JUN, FOSL1*
GO_BP	GO:0033993~response to lipid	5.09E-06	8	*CYP1A1, DUSP1, EGR1, ALDH3A1, FOS, JUN, WNT7B, FOSL1*
GO_BP	GO:0010033~response to organic substance	6.20E-06	14	*IFI30, CYP1A1, DAPK3, DUSP1, EGR1, ALDH3A1, FOS, PPP1R15A, IFI6, JUN, WNT7B, FOSL1, GDF15, FGFBP1*
GO_CC	GO:0016324~apical plasma membrane	8.49E-03	3	*SLC34A3, CYP4F12, VAMP3*
GO_CC	GO:0071944~cell periphery	1.51E-02	15	*IFI30, MISP, SLC34A3, IFI6, TTLL10, IHH, CEACAM6, FXYD3, PANX2, S100A6, CYP4F12, WNT7B, PSCA, VAMP3, FGFBP1*
GO_CC	GO:0045177~apical part of cell	1.79E-02	3	*SLC34A3, CYP4F12, VAMP3*
GO_CC	GO:0005886~plasma membrane	2.99E-02	14	*IFI30, SLC34A3, IFI6, TTLL10, IHH, CEACAM6, FXYD3, PANX2, S100A6, CYP4F12, WNT7B, PSCA, VAMP3, FGFBP1*
GO_CC	GO:0005737~cytoplasm	3.15E-02	24	*IFI30, TRIM31, MISP, TRIM16L, CYP1A1, DAPK3, EGR1, ALDH3A1, FOS, PPP1R15A, FTL, IFI6, TTLL10, GPX2, JUN, PANX2, S100A6, CYP4F12, PHLDA2, WNT7B, FOSL1, PIR, VAMP3,IER2*
GO_MF	GO:0016491~oxidoreductase activity	1.86E-04	7	*IFI30, CYP1A1, ALDH3A1, FTL, GPX2, CYP4F12, PIR*
GO_MF	GO:0070412~R-SMAD binding	5.08E-04	2	*FOS, JUN*
GO_MF	GO:0000977~RNA polymerase II regulatory region sequence-specific DNA binding	6.81E-04	3	*EGR1, JUN, FOSL1*
GO_MF	GO:0070330~aromatase activity	6.84E-04	2	*CYP1A1, CYP4F12*
GO_MF	GO:0001012~RNA polymerase II regulatory region DNA binding	7.43E-04	3	*EGR1, JUN, FOSL1*
Pathway	hsa04380~Osteoclast differentiation	3.76E-03	3	*FOSL1, JUN, FOS*
Pathway	hsa04310~Wnt signaling pathway	5.98E-03	3	*FOSL1, WNT7B, JUN*
Pathway	hsa05200~Pathways in cancer	7.80E-03	4	*DAPK3, WNT7B, JUN, FOS*
Pathway	hsa04340~Hedgehog signaling pathway	8.60E-03	2	*WNT7B, IHH*
Pathway	hsa05210~Colorectal cancer	1.05E-02	2	*JUN, FOS*

BP: biological process; CC: cell component; MF: molecular function.

signaling (BP, $P = 3.54E-09$), had oxidoreductase activity (MF, $P = 1.86E-04$), and were involved in osteoclast differentiation (pathway, $P = 3.76E-03$) (Table 1). The top five functions and pathways for downregulated genes included regulation of BPs (BP, $P = 1.45E-05$) within the endomembrane system (CC, $P = 1.75E-02$), MFs ($P = 1.10E-04$), and gap junction pathway involvement ($P = 3.01E-02$) (Table 2).

PPI network analysis

There were 43 nodes (29 upregulated and 14 downregulated genes) and 71 edges in the PPI network for the identified DEGs (Figure 1). In the PPI network, FOS (degree = 12), AHR (degree = 12), JUN (degree = 11), and cytochrome P450 family 1 subfamily A member 1 (CYP1A1, degree = 11) had higher degrees. A protein with a higher degree indicates that they are highly interconnected with other proteins in the PPI network (Supplementary Table 2).

Transcriptional regulatory network analysis

Based on the TRANSFAC® database, 4 upregulated (*EGR1*, *FOSL1*, *FOS*, and *JUN*) and 1 downregulated (*AHR*) genes were identified as TFs. After TF-DEG pairs were predicted (Supplementary Table 3), the transcriptional regulatory network was constructed and found to have 43 nodes (17 upregulated genes and 26 downregulated genes) and 71 edges (Figure 2). Moreover, all the genes involved in the transcriptional regulatory network were performed with pathway enrichment analysis. The enriched pathways included osteoclast differentiation ($P = 6.43E-03$), Wnt signaling ($P = 1.01E-02$; involving *FOSL1*, *JUN*, and *WNT7B*), and colorectal cancer ($P = 1.49E-02$) (Table 3).

Discussion

In the present study, a total of 121 DEGs (50 upregulated and 71 downregulated) were identified in gastric cancer cells treated with quercetin, and PPI network analysis showed that FOS (degree = 12), AHR (degree = 12), JUN (degree = 11), and CYP1A1 (degree = 11) had higher degrees and highly interconnected with other proteins. Previously, *c-Fos* downexpression was found to have tumor suppressor activity in gastric cancer, which may be associated with this protein's proapoptotic function (Jin et al. 2007; Zhou et al. 2010). In addition, *c-Fos* is overexpressed in human gastric adenocarcinoma metastasis involvement in the IL-1B/p38/AP-1/MMP2/MMP9 pathway and may be a new therapeutic target for the disease (Huang et al. 2014). *AHR* inhibition and calpain-10 activation have been shown to inhibit both peritoneal dissemination and growth of gastric tumors by suppressing epithelial-to-mesenchymal transition and inducing endoplasmic reticulum stress (Lai et al. 2014). Previous studies have also indicated that *AHR* facilitates growth and invasion of gastric carcinoma cells. Therefore, *AHR* may be a promising target for the treatment of gastric cancer (Yin et al. 2013; Powell and Ghotbaddini 2014). Suppression of c-Jun-N-terminal kinase/c-Jun/activator protein-1 has been shown to promote the antitumor activity of a cyclooxygenase 2-specific inhibitor, and suppression of c-Jun-N-terminal kinase activation may positively contribute to the treatment of gastric cancer (Jiang et al. 2004). Moreover, *CYP1A1* is a major enzyme in the carcinogen metabolizing pathway, and *CYP1A1* (rs4646422) polymorphism may be associated with gastric cancer development among Japanese individuals (Xue et al. 2012; Hidaka et al. 2016). Thus, these results suggest that quercetin functions against gastric cancer by regulating *FOS*, *AHR*, *JUN*, and *CYP1A1*.

Among DEGs, *EGR1*, *FOSL1*, *FOS*, *JUN*, and *AHR* were also TFs. By blocking nuclear factor-κB and EGR1 in gastric cancer AGS cells, chrysin has been shown to inhibit Recepteur

Table 2. The top 5 functions and pathways enriched for the down-regulated genes.

Category	Description	*p* Value	Gene number	Gene symbol
GO_BP	GO:0008150∼biological_process	1.45E-05	56	*CEBPD, MAP3K2, NUDT4, PSIP1, AKAP11, SNX18, GPR182, CHRNA5, OSBPL8, B3GALT6, UHMK1, EMB, LSM11, NEK7, SESN3, EGFR, AHR, STT3B, EXPH5, SLC44A1, DDAH1, FRK, BAMBI, RANBP6, AFF4, TMED8, FOXN2, HEPHL1, ITGA2, MARCKS, MAP1B, MBNL1, RLIM,HECA, CAB39, ANLN, FAR2, GPR126, LMBR1, RFX7, SLC12A2, SNTB1, SLC30A1, RNF128, SGPP1, ITCH, MAML2, EBPL, ENC1, HNRNPLL, PREPL, NFE2L3, NUP155, SOCS5, SLK, LPGAT1*
GO_BP	GO:0035413∼positive regulation of catenin import into nucleus	4.16E-04	2	*EGFR, BAMBI*
GO_BP	GO:0006376∼mRNA splice site selection	1.39E-03	2	*PSIP1, MBNL1*
GO_BP	GO:0046636∼negative regulation of alpha-beta T cell activation	1.39E-03	2	*ITCH, SOCS5*
GO_BP	GO:0046822∼regulation of nucleocytoplasmic transport	1.93E-03	4	*NUDT4, UHMK1, EGFR, BAMBI*
GO_CC	GO:0012505∼endomembrane system	1.75E-02	11	*SNX18, MAL2, B3GALT6, EGFR, STT3B, FAR2, RNF128, SGPP1, EBPL, NUP155, LPGAT1*
GO_CC	GO:0043235∼receptor complex	2.41E-02	3	*CHRNA5, AHR, ITGA2*
GO_CC	GO:0005575∼cellular_component	2.68E-02	60	*CEBPD, MAP3K2, NUDT4, PSIP1, AKAP11, SNX18, GPR182, CHRNA5, MAL2, B3GALT6, UHMK1, EMB, LSM11, NEK7, SESN3, EGFR, AHR, STT3B, EXPH5, SLC44A1, LRRC8B, DDAH1, FRK, EPHX4, BAMBI, RANBP6, AFF4, TMED8, FOXN2, HEPHL1, ITGA2, TMEM238, MARCKS, MAP1B, MBNL1, RLIM, HECA, CAB39, GFOD1, ANLN, FAR2, GPR126, LMBR1, RFX7, SLC12A2, SNTB1, POTEF, SLC30A1, RNF128, SGPP1, ITCH, MAML2, EBPL, ENC1, HNRNPLL, PREPL, NFE2L3, NUP155, SLK, LPGAT1*
GO_CC	GO:0016021∼integral to membrane	2.91E-02	25	*GPR182, CHRNA5, MAL2, B3GALT6, EMB, EGFR, STT3B, SLC44A1, LRRC8B, EPHX4, BAMBI, TMED8, HEPHL1, ITGA2, TMEM238, FAR2, GPR126, LMBR1, SLC12A2, SLC30A1, RNF128, SGPP1, EBPL, NUP155, LPGAT1*
GO_CC	GO:0042383∼sarcolemma	3.23E-02	2	*SNTB1, SLC30A1*
GO_MF	GO:0003674∼molecular function	1.10E-04	55	*CEBPD, MAP3K2, NUDT4, PSIP1, AKAP11, SNX18, GPR182, CHRNA5, MAL2, OSBPL8, B3GALT6, UHMK1, LSM11, NEK7, EGFR, AHR, STT3B, EXPH5, SLC44A1, DDAH1, FRK, EPHX4, BAMBI, RANBP6, AFF4, FOXN2, HEPHL1, ITGA2, MARCKS, MAP1B, MBNL1, RLIM, HECA, CAB39, GFOD1, ANLN, FAR2, GPR126, RFX7, SLC12A2, SNTB1, SLC30A1, RNF128, SGPP1, ITCH, MAML2, EBPL, ENC1, HNRNPLL, PREPL, NFE2L3, NUP155, SOCS5, SLK, LPGAT1*
GO_MF	GO:0004709∼MAP kinase kinase kinase activity	1.66E-03	2	*MAP3K2, EGFR*
GO_MF	GO:0004672∼protein kinase activity	1.82E-03	7	*MAP3K2, UHMK1, NEK7, EGFR, FRK, CAB39, SLK*
GO_MF	GO:0004674∼protein serine/threonine kinase activity	1.95E-03	6	*MAP3K2, UHMK1, NEK7, EGFR, CAB39, SLK*
GO_MF	GO:0003779∼actin binding	4.50E-03	5	*EGFR, MARCKS, ANLN, SNTB1, ENC1*
Pathway	hsa04540∼Gap junction	3.01E-02	2	*MAP3K2, EGFR*
Pathway	hsa04912∼GnRH signaling pathway	3.72E-029	3	*MAP3K2, EGFR*

BP: biological process; CC: cell component; MF: molecular function.

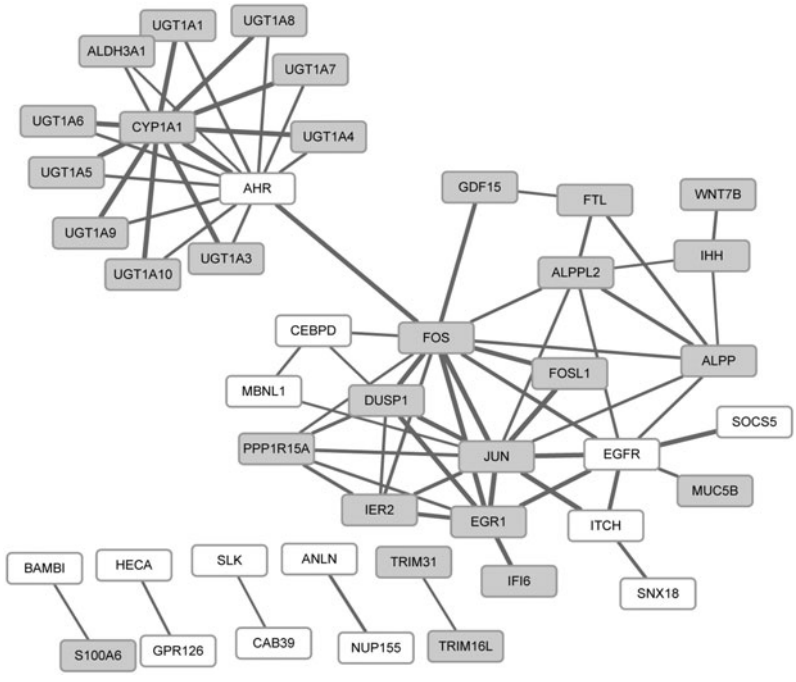

Figure 1. The protein–protein interaction network constructed for differentially expressed genes. Grey and white represent upregulated and downregulated genes, respectively.

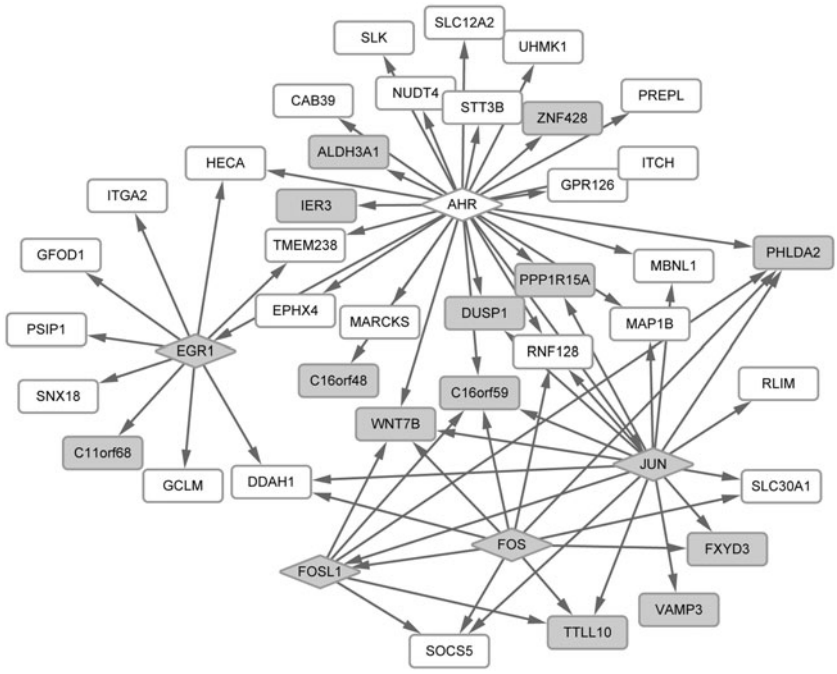

Figure 2. The transcriptional regulatory network. Grey and white represent upregulated and downregulated genes, respectively. Diamonds and rectangles indicate transcription factors and target genes, respectively.

Table 3. The pathways enriched for the genes involved in the transcriptional regulatory network.

Description	*p* Value	Gene number	Gene symbol
hsa04380~Osteoclast differentiation	6.43E-03	3	*FOS, FOSL1, JUN*
hsa04310~Wnt signaling pathway	1.01E-02	3	*FOSL1, JUN, WNT7B*
hsa05210~Colorectal cancer	1.49E-02	2	*FOS, JUN*
hsa05200~Pathways in cancer	1.53E-02	4	*FOS, ITGA2, JUN, WNT7B*
hsa05140~Leishmaniasis	2.04E-02	2	*FOS, JUN*
hsa04662~B cell receptor signaling pathway	2.14E-02	2	*FOS, JUN*
hsa05323~Rheumatoid arthritis	3.14E-02	2	*FOS, JUN*
hsa04620~Toll-like receptor signaling pathway	3.79E-02	2	*FOS, JUN*
hsa05142~Chagas disease (American trypanosomiasis)	3.93E-02	2	*FOS, JUN*
hsa04660~T cell receptor signaling pathway	4.21E-02	2	*FOS, JUN*
hsa04010~MAPK signaling pathway	4.57E-02	3	*DUSP1, FOS, JUN*

d'Origine Nantais expression, leading to anticancer effects (Xia et al. 2015). Through the extracellular signal-regulated kinases 1/2-EGR1 pathway, periplocin can suppress proliferation and induce the apoptosis of gastric cancer cells (Li et al. 2016), whereas though p53-independent EGR1/p21 signaling, genipin can induce the apoptosis of gastric cancer AGS cells (Ko et al. 2015). Moreover, *Fra-1* (*FOSL1*) has also been found to be overexpressed in gastric cancer, impacting phosphatidylinositol-3-kinase/Akt and p53 signaling (He et al. 2015). Overexpression of *Fra-1* may correlate with the development and progression of gastric carcinoma, making it another possible diagnostic marker and/or therapeutic target for the disease (Wang et al. 2013). These reports suggest that quercetin's mechanism of action against gastric cancer also correlates with *EGR1* and *FOSL1*.

Furthermore, current pathway enrichment analysis results showed that *FOSL1*, *JUN*, and *WNT7B* were enriched in the Wnt signaling pathway. Wnt signaling contributes to oncogenesis by suppressing c-Myc–induced apoptosis (You et al. 2002), and Wnt signaling also plays a role in gastric tumorigenesis (Nojima et al. 2007). As a Wnt signaling molecule, *WNT7B* is upregulated in gastric cancer cells and may play a critical role in the tumorigenesis of gastric cancer (Kim et al. 2003). Therefore, quercetin might also regulate gastric cancer by targeting *WNT7B* activity associated with Wnt signaling.

There are some limitations to the present study, and more research is needed to further confirm our findings. In future studies, expression of DEGs identified in the current study will be validated by real time-polymerase chain reaction, and their interactions within the PPI network as well as the regulatory relationships between TFs and DEGs will be confirmed.

The present in-depth bioinformatics analysis identified a total of 121 DEGs in human gastric cancer cells treated with quercetin versus controls. Five of these DEGs were determined to be TFs, including *EGR1*, *FOSL1*, *FOS*, and *JUN* (all upregulated) and *AHR* (downregulated). PPI network analysis demonstrated that CYP1A1 has a higher degree and interacts with AHR. In addition, FOSL1, JUN, and WNT7B were found to be enriched in the Wnt signaling pathway. Therefore, *FOS*, *AHR*, *JUN*, *CYP1A1*, *EGR1*, *FOSL1*, and *WNT7B* may be potential targets of quercetin in gastric cancer cells. Current results provide further understanding on the pathogenesis of gastric cancers treated with quercetin.

Disclosure statement

No potential conflict of interest was reported by the authors.

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