Original Article

Overexpression of UHRF1 and its potential role in the development of invasive ductal breast cancer validated by integrative bioinformatics and immunohistochemistry analyses

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Background: Increasing evidence has highlighted the role of ubiquitin-like PHD and RING finger domain-containing protein 1 (UHRF1) in the development of cancers, including hepatocellular carcinoma, pancreatic cancer, and bladder cancer. However, the correlation between UHRF1 and breast cancer remains unclear. The present study aimed to analyze the expression of UHRF1 and its role in the development of invasive ductal breast cancer (IDC) by integrating multilevel expression data and immunohistochemistry analysis.

Methods: The Gene Expression Omnibus (GEO) and The Cancer Genome Atlas (TCGA) databases were used to gather UHRF1 expression data on IDC. Additionally, immunohistochemistry analysis was used to investigate the correlations between UHRF1 expression and the clinical characteristics of IDC.

Results: The GEO and TCGA databases indicated that UHRF1 was up-regulated in IDC. Consistently, the immunohistochemical specimens showed that the significant overexpression of UHRF1 in IDC, and its expression level showed an increasing trend from ductal carcinomas *in situ* to IDC. Notably, the increased levels of UHRF1 were closely correlated with estrogen receptor expression, pathological grade, and the prognosis of the disease. In addition, patients with a high UHRF1 expression had a poorer prognosis.

Conclusions: In conclusion, our findings suggested that UHRF1 plays a promoting role in breast tumorigenesis, and the over-expression of UHRF1 could serve as a biomarker for the prognosis in invasive ductal carcinomas in breast cancer.

Keywords: Breast cancer; ER; bioinformatics analysis; tumorigenesis; ubiquitin-like PHD and RING finger domain-containing protein 1 (UHRF1)

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Introduction

Breast cancer has long been a threat to women's health for its high rates of morbidity and mortality. According to the data from the World Health Organization (WHO), breast cancer is the second most deadly cancer among women in the US (1). Although diagnostic approaches and treatments have improved in recent years, the 5-year survival rate is still less than 25% in breast cancer patients with distant metastasis (2). The pathogenesis of breast cancer is complex and involves multiple factors, including cholesterol metabolism (3), hormone levels (4), and diet (5), among others. Understanding the pathogenesis and seeking for

biomarkers is urgently needed for the diagnosis, therapy, and prognosis of breast cancer patients.

Ubiquitin-like PHD and RING finger domain-containing protein 1 (UHRF1) [also named nuclear protein 95 (Np95), or inverted CCAAT box-binding protein of 90 kDa (ICBP90) (6)] plays an essential role in cell proliferation and DNA methylation (7,8). Currently, a plethora of research has demonstrated that UHRF1 participates in the development and progression of many cancers (9). It could inactivate tumor suppressor genes by methylation in many cancers, such as non-small lung carcinoma (9), gastric cancer (10), and endometrial carcinoma (11). High expression of UHRF1 is associated with a poor prognosis in many cancers, including hepatocellular carcinoma (10), pancreatic cancer (11), and bladder cancer (12). Previous research has described a differential role of UHRF1 in breast cancer. Some studies found that UHRF1 inhibits the transcription of *MDR1* gene, and enhances the chemotherapeutic effects (13), while other studies found that UHRF1 can act as an oncogene, promoting tumorigenesis and metastasis through complicated mechanisms, such as silencing DNA repair genes and inhibiting apoptosis (14). In the breast cancer cell lines, the mRNA (7,15) and protein (16) expression of UHRF1 was upregulated. The UHRF1 DNA levels in the plasma of breast cancer patients were also increased (17). Clinically, the expression of UHRF1 is gradually increased from normal breast tissue to low-grade breast cancer tissue to high-grade breast cancer tissue (6).

Breast cancer is a heterogeneous disease with many pathological types and the different pathological types have different prognoses (18). It is better to detect the different pathological types of breast cancer separately than it is to do so together. In this study we mainly investigated the expression of UHRF1 in invasive ductal carcinomas of breast cancer, which is the major pathological type of breast cancer. This study aimed to study the role of UHRF1 in invasive ductal carcinoma based on bioinformatics analysis, and to investigate its associations with its clinical pathological characteristics and prognostic significance.

Methods

Gene Expression Omnibus (GEO) data analysis

To identify the critical candidate genes in the development and progression of breast cancer, all breast cancer datasets were collected and assembled from the GEO database. The search strategy was formulated was as follows: (malignan* OR cancer OR tumor OR tumor OR neoplas* OR carcinoma) AND (breast OR mammary). Inclusion criteria included the following: (I) the dataset sample organism was from Homo sapiens; (II) both breast cancer and normal breast tissue with more than two samples were included in the datasets; and (III) provided enough mRNA expression data of normal breast tissue or breast cancer were provided for analysis. Meanwhile, exclusion criteria included the following: (I) data based on cell lines; (II) breast cancer or normal breast tissue treated with some specific treatments such as chemotherapeutic drugs, gene knockout etc.; (III) non Homo sapiens test subjects; and (IV) breast cancer patients with other coincident cancers. The quality-controlled and standardized chip data used in the chosen databases were analyzed by Bayesian Analysis performed by R (version: 2.15.3). Probes without complete gene expression data were filtered, and the probes were transformed manually into gene symbols according to GPL570 manually. Genes with an adjusted P value <0.01 and log2-transformed expression fold change >1.5 or <–1.5 were considered differentially expressing genes (DEG) and chosen for further analysis. In this study, GSE10780, which was submitted by Chen *et al.* (19) to the GEO database and based on the GPL570 platform (Affymetrix Human Genome U133 Plus 2.0 Array) was chosen for analysis. The GSE10780 included 143 histologically normal breast tissues and 42 invasive ductal carcinoma (IDC) tissues, and was analyzed using the Naive Bayes methods after initial quality control, background correction, and standardization. The top 100 DEGs in normal breast tissues and IDC tissues were used to drawn a heatmap by using R (version: 2.15.3). The GSE29044, which was submitted by Colak *et al.* (20) and was based on GPL570 platform, contained 36 IDC tissue and 67 normal breast tissue, and were analyzed by the same method to validate the results.

The Cancer Genome Atlas (TCGA) data analysis

For cross-validating mRNA overexpression of target genes, Counts were downloaded and extracted from TCGA (http://cancergenome.nih.gov/) during April 2018 by using the data transfer tool (downloaded from https://gdc.cancer.gov/access-data/gdc-data-transfer-tool). After downloading the count data which is presented as the Roshal Archive, the gene expression matrix with ensemble ID was extracted by decompressing packages and extracting the script manually. Downloading of the TCGA symbol IDs for sampling was used to correlate gene expression matrices using R (version:

2.15.3). These samples were classified into two groups: a tumour group (n=1,102) and a normal group (n=113). Gene expression data were compared between the two groups by Student's *t*-test. Approval by an ethics committee was not needed as the data were obtained from TCGA. This study meets the TCGA publication guidelines.

Patients and tumor specimens

The study was approved by the Medical Ethics Committee of Xinxiang Central Hospital. Paraffin-embedded tissue blocks from 96 IDC patients who underwent biopsy at initial diagnosis between April 2001 and August 2004 without treatment at initial presentation were retrieved. 67 normal breast tissues and 37 breast tissues of ductal carcinoma *in situ* (DCIS) were also included in the study. Three pathologists re-appraised the histological subtypes according to the current WHO classification. Tumor staging was re-appraised using the 7th edition of the American Joint Committee on Cancer (AJCC) system.

Survival analysis

All 96 IDC patients underwent chemotherapy and radiotherapy according to NCCN guidelines. The 37 patients with DCIS underwent a conventional surgical excision. Patient follow-up was defined as the time between surgery and the last hospital contact (scheduled follow-up or telephone contact) or a recurrence of the disease. The endpoints analyzed were disease-free survival (DFS) and overall survival (OS) and were calculated using the date of surgical resection to the date of the event. The event was defined as the time of hospital contact (scheduled follow-up or telephone contact), disease recurrence, or patient death according to the definition of DFS and OS. Patients lost to follow-up were recorded as the latest follow-up date. Ninety-six IDC tissues were divided into a high-transcription group and low-transcription group by the median value of FPKM of UHRF1 and analyzed by chi-square or Fisher's exact test. The follow-up deadline was July 2014.

Immunohistochemical staining and assessment of UHRF1

Immunohistochemical staining was carried out as follows. Sections (4 μm thick) were cut from paraffin-embedded blocks and mounted on glass slides. The slides were deparaffinized with xylene and rehydrated with ethanol. The slides were heated in a pressure cooker with 10 mM citrate buffer solution (pH 6) for 2 minutes to retrieve antigen epitopes. Endogenous peroxidase was quenched by a 3% H_2O_2 treatment solution. The slides were washed with phosphate buffered saline (PBS) and incubated with blocking buffer at room temperature for 30 minutes, and then with UHRF1 primary antibodies (cat no: ab194236, 1:100, Abcam, UK) at 4 ℃ overnight followed by incubation with the secondary antibody at room temperature (Neobioscience, China) for 30 minutes. The slides were developed with 3,3-diaminobenzidine for 5 minutes and then counter-stained with hematoxylin. We used lymphocytes from human tonsils, which are known to express UHRF1, as the positive control and breast cancer tissue, incubated with PBS instead of the primary antibody, as the negative control.

Three pathologists, with no prior knowledge of the clinical and follow-up information, scored UHRF1 immunoexpression using a multi-head microscope to reach a consensus. Nuclear UHRF1 staining scores were calculated using H-scores. The H-score was calculated using the following equation: H-score = $\Sigma Pi \times (i+1)$, where i stands for the intensity score of the slides (which ranged from 0 to 3), and Pi stands for the percentage of stained cells at each intensity (which ranged from 0 to 100%). The H-score ranges from 0 to 4.0, where 0 indicates that 100% of cells were negative (0), and 4.0 indicates that 100% of the cells were strongly stained (3+). H-scores higher than the median value were construed as UHRF1 overexpression.

Statistical analysis

R (version: 2.15.3) was used to analyze the DEGs between the normal breast tissues and IDC tissues and to match symbol ID to the TCGA Ensembl ID. SPSS 19 software package (SPSS Inc., Chicago, IL, USA) was used for statistical analysis. We compared the data between groups by *t*-test and evaluated the association between UHRF1 expression status and various clinicopathological parameters by chi-square or Fisher's exact test (if the theoretical frequency was smaller than 5). For survival analysis, we performed log-rank tests to evaluate the prognostic differences between the groups and plotted the survival curves using the Kaplan-Meier method with GraphPad Prism 5.0 (GraphPad Software, San Diego, CA, USA). For all analyses, $P<0.05$ was considered statistically significant.

Table 1 Summary of top 10 differentially expressed genes between breast normal tissue and breast cancer tissue

Probe	Gene symbol	LogFC	Adj P value	Molecular function
225655_at	UHRF1	2.66	1.79×10^{-55}	Transferase activity, ubiquitin protein ligase activity, metal ion binding, hemi-methylated DNA-binding, hemi-methylated DNA-binding proximal promoter sequence-specific DNA binding, nucleosomal histone binding, methylated histone binding, methyl-cpg binding, identical protein binding, histone binding
210052_s_at	TPX2	1.90	2.83×10^{-49}	ATP binding, GTP binding, importin-alpha family protein binding, protein binding, protein kinase binding
229357_at	ADAMTS5	–2.70	4.92×10^{-49}	Extracellular matrix binding, heparin binding, integrin binding, metalloendopeptidase activity, metallopeptidase activity, protein binding, zinc ion binding
218039_at	NUSAP1	2.68	5.20×10^{-49}	DNA binding, RNA binding, microtubule binding, protein binding
205941_s_at	COL10A1	3.12	9.98×10^{-49}	Metal ion binding, protein binding, extracellular matrix structural constituent, molecular_function
235368_at	ADAMTS5	–2.68	1.39×10^{-48}	Extracellular matrix binding, heparin binding, integrin binding, metalloendopeptidase activity, peptidase activity, metallopeptidase activity, protein binding, zinc ion binding
218585_s_at	DTL	2.26	1.39×10^{-48}	Protein binding, contributes_to ubiquitin-protein transferase activity
223229_at	UBE2T	2.32	1.45×10^{-48}	ATP binding, chromatin binding, protein binding, nucleotide binding, transferase activity, ubiquitin conjugating enzyme activity, ubiquitin protein ligase activity, ubiquitin protein ligase binding, ubiquitin-protein transferase activity
203213_at	CDK1	2.46	1.90×10^{-48}	ATP binding, Hsp70 protein binding, RNA polymerase II carboxy-terminal domain kinase activity, RNA polymerase II carboxy-terminal domain kinase activity, chromatin binding, cyclin binding, cyclin-dependent protein serine/threonine kinase activity, RNA polymerase II carboxy-terminal domain kinase activity, protein serine/threonine kinase activity
204641_at	NEK2	2.47	3.63×10^{-48}	ATP binding, metal ion binding, protein binding, protein kinase activity, protein phosphatase binding, protein serine/threonine kinase activity, transferase activity

Results

UHRF1 is upregulated in IDC

To identify the role of *UHRF1* in the development and progression of breast cancer, the breast cancer datasets GSE10780 and GSE29044 were obtained from the GEO. In the GSE10780 dataset, we identified 4928 genes with adjusted P values <0.01, including 513 genes with |log FC| >1.5. Furthermore, the 513 DEGs identified consisted of 129 upregulated and 384 downregulated genes. *UHRF1*, *TPX2*, *ADAMTS5*, *NUSAP1*, *COL10A1*, *ADAMTS5*, *DTL*, *UBE2T*, *CDK1*, and *NEK2* were the top 10 differentially expressed genes between IDC and normal tissues (*Table 1*). Among the statistically significant genes, *UHRF1* had the smallest adjusted P value (1.79×10^{-55}), and a |log FC| of 2.66, making it the most differentially expressed gene. We plotted the heat map of the top 100 differently expressed genes (*Figure 1*). In GSE29044, *UHRF1* was also identified as a DEG with an adjusted P value of 2.07×10^{-6} and a |log FC| of 2.03.

Compared with the normal breast tissues, expression of *UHRF1* in GSE10780 and GSE29044 was significantly upregulated (*Figure 2A*,*B* respectively). We subsequently performed cross-validation using TCGA datasets and found that the *UHRF1* expression in IDC was dramatically higher (10.65-fold) than that in paracarcinoma tissues (*Figure 2C*).

To further observe the expression of UHRF1 in breast cancer, immunohistochemistry was utilized to detect the expression of UHRF1 expression in 96 IDC samples, 37 DICS samples and 67 normal breast tissue samples. Positive

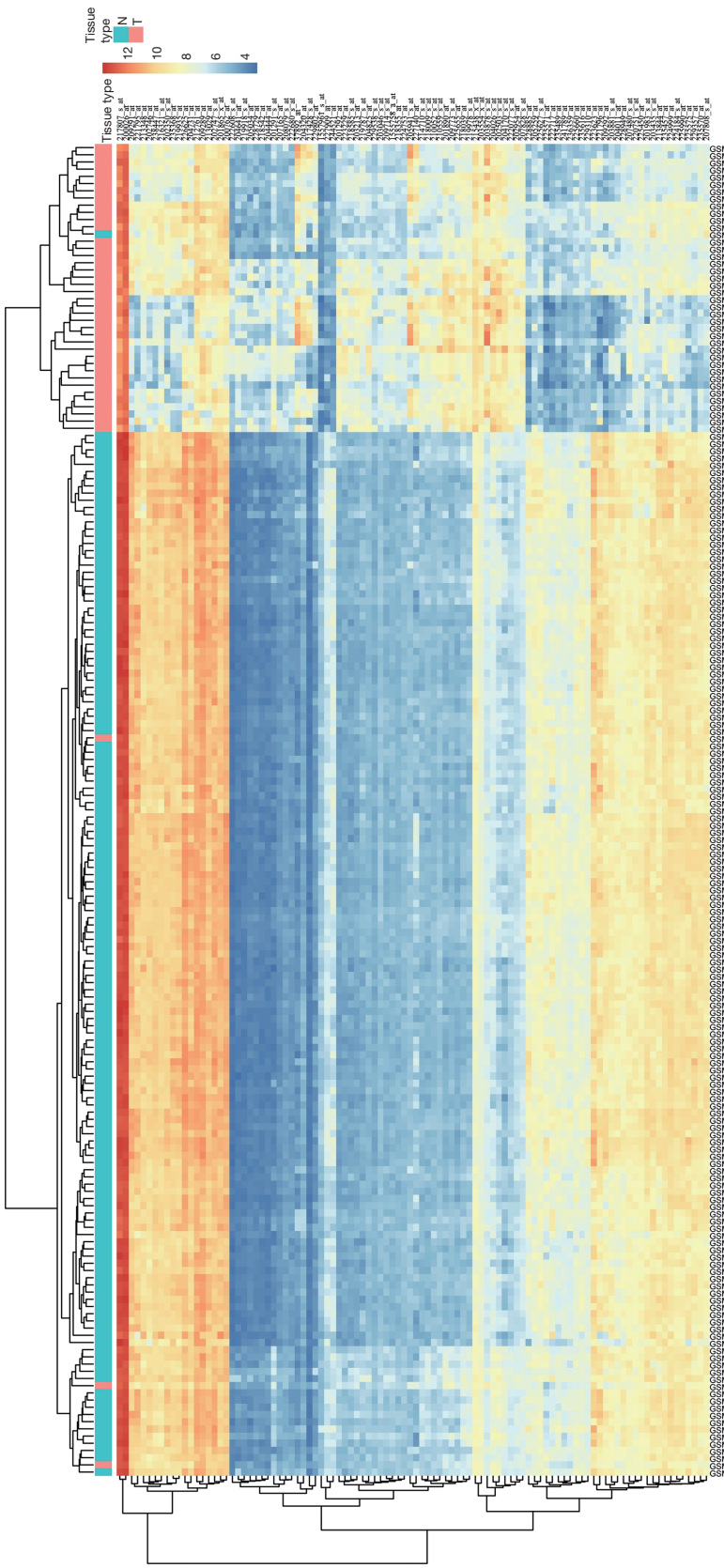

Figure 1 Heatmap composed of the top 100 DEGs in normal breast tissues and IDC from the GSE10780 dataset. GSE1078, which was submitted by Chen *et al.*, included 143 histologically normal breast tissues and 42 IDC tissues. Each column represents tissue from breast cancer or normal breast tissue, while each row represents a probe of the top 100 DEGs. N: normal breast tissue; T: breast tumor tissue. The dendrogram represents the similarity of expression profiles between the tissue samples based on average linkage clustering. Blue indicates a low level of expression, green and red are intermediate levels, and yellow indicates maximal levels of gene expression. DEGs, differentially expressed genes; IDC, invasive ductal carcinoma.

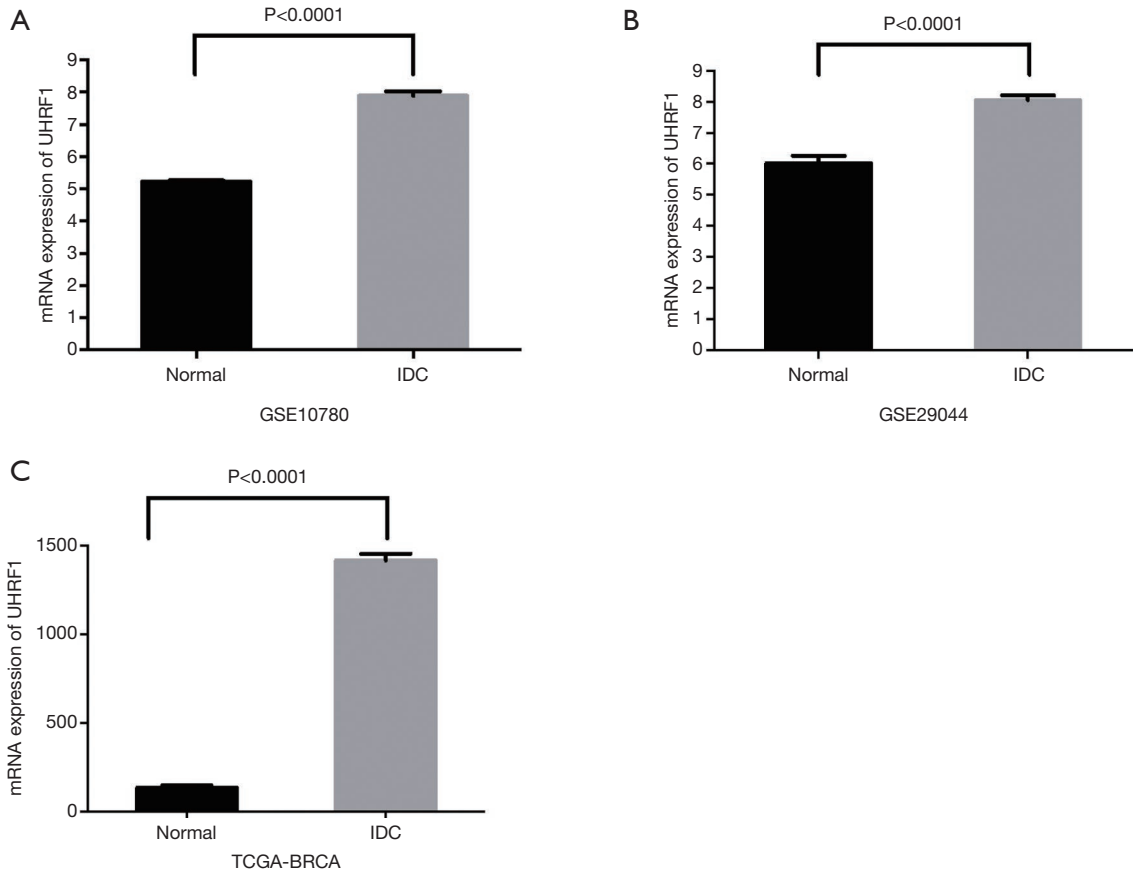

Figure 2 UHRF1 was up-regulated in IDC tissues. There are 143 histologically normal breast tissues and 42 IDC tissues; 67 histologically normal breast tissues and 36 IDC tissues; 113 histologically normal breast tissues and 1,102 invasive ductal carcinoma tissues in the GSE10780, GSE29044, and TCGA databases respectively. (A) The expression of UHRF1 was significantly increased in breast invasive ductal carcinomas compared to the normal breast tissues based on the Gene Expression Omnibus database GSE10780 (P<0.0001); (B) UHRF1 was highly expressed in breast cancer tissues compared with the normal breast tissues based on the GSE29044 database (P<0.0001); (C) UHRF1 was highly expressed in breast invasive ductal carcinomas compared with the non-cancerous breast tissues based on The Cancer Genome Atlas database (P<0.0001). The data were expressed as mean ± SEM, and were analyzed using Student's *t*-test. Normal, normal breast tissues; UHRF1, ubiquitin-like PHD and RING finger domain-containing protein 1; IDC, invasive ductal carcinoma.

staining of UHRF1 was mainly distributed in the nucleus of breast cancer. Representative staining of UHRF1 in IDC, DICS, and normal tissues are summarized in *Figure 3A*. In 96 cases of IDC, 60 (62.5%) showed UHRF1 overexpression. In 37 cases of DCIS, 20 showed UHRF1 overexpression (54.1%). In 67 cases of normal breast tissues, 21 showed UHRF1 overexpression (30.9%).

Subsequently, we compared the UHRF1 expression in the normal breast tissues and in IDC and DICS breast cancer samples. Our results showed that UHRF1 expression was the lowest in the normal breast tissues (average protein expression values =0.22). DCIS showed intermediate expression (average protein expression values =0.52), and IDC had the highest expression of UHRF1 (average protein expression values =0.76). Statistical analyses revealed a significant increase of UHRF1 in DCIS and IDC tissues compared to normal tissues (*Figure 3B*).

Relationship between UHRF1 and clinicopathological features of IDC

To dissect the roles of UHRF1 in the development of breast cancer, we analyzed the associations of UHRF1 expression with clinicopathological features of breast cancer using

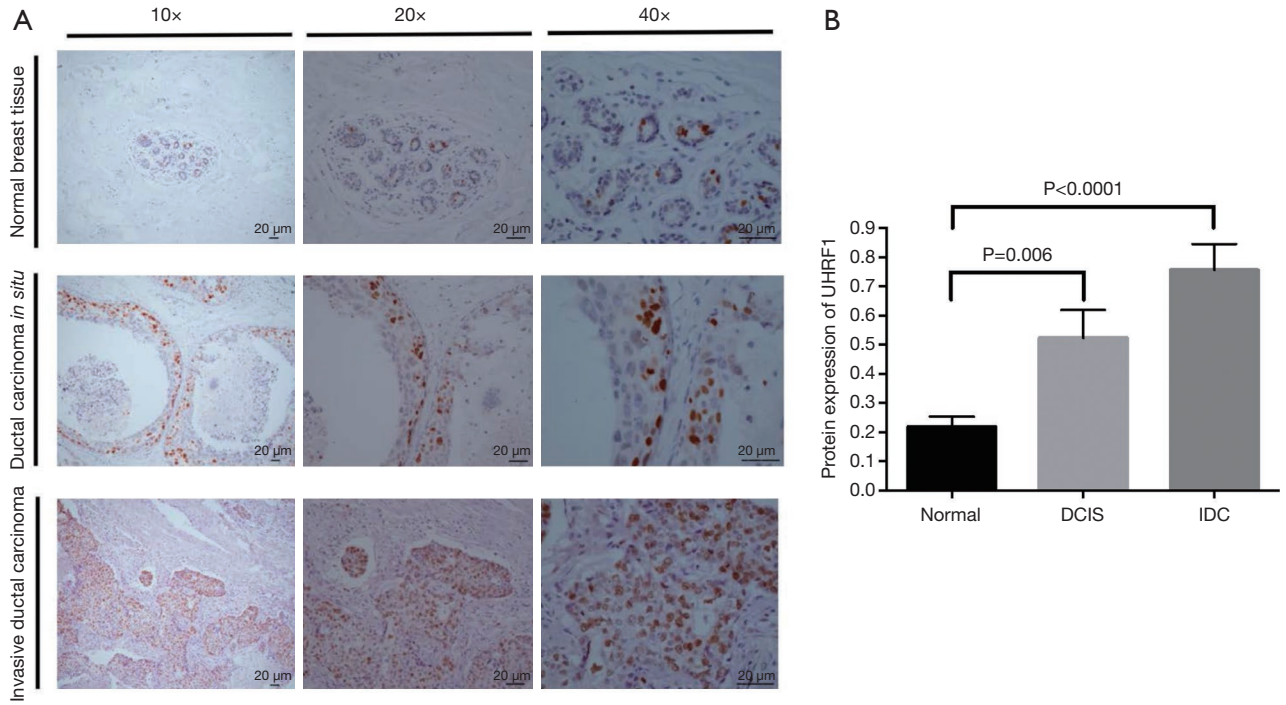

Figure 3 The immunohistochemistry analyses of UHRF1 expression in DCIS and IDC tissues. Expression of UHRF1 protein in invasive ductal carcinomas (n=96), ductal carcinoma *in situ* (n=37), and normal tissues (n=67) was analyzed by immunohistochemistry. Positive staining of UHRF1 was mainly distributed in the nucleus of breast cancer. Compared to normal breast tissues, the expression of *UHRF1* in DCIS and IDC was elevated. (A) The representative staining of UHRF1 in breast invasive ductal carcinomas, ductal carcinoma *in situ*, and normal tissues. (B) Statistical analysis of the UHRF1 expression in different breast cancer tissues. The data were expressed as mean ± SEM, and the statistical significance was tested using Student's *t*-test. Normal, normal breast tissues; UHRF1, ubiquitin-like PHD and RING finger domain-containing protein 1; DCIS, ductal carcinoma *in situ*; IDC, invasive ductal carcinoma.

SPSS 19.0 software. We classified the breast cancer patients into UHRF1 high and low expression groups based on the median expression level of UHRF1.

As shown in *Table 2*, UHRF1 protein expression was correlated with estrogen receptor (ER) (χ^2=4.200, P=0.040) and pathological grade (χ^2=4.798, P=0.028) of breast tumor, but not related with other clinical features, such as age and sex (P>0.028).

High UHRF1 expression is associated with poor survival in IDC

To explore the prognostic value of UHRF1 in breast cancer, Kaplan-Meier survival analysis was employed to analyze the DFS and OS. Among the total IDC patients, the average follow-up time was 97.3 (range, 8–147) months. As shown in *Figure 4*, patients with high UHRF1 expression had a worse OS (47.92%) than those with low UHRF1 expression (29.17%). The DFS of UHRF1 high expression and low expression groups were 31.25% and 54.17%, respectively, while the UHRF1 high expression group showed statistically significantly lower DFS (*Figure 4A*) and OS (*Figure 4B*) than that of the low expression group.

Discussion

Recent studies have demonstrated that UHRF1 is related to the progression (21), drug resistance (13), and radiotherapy (22) of breast cancer. Most of these studies were focused on dissecting the mechanism of how UHRF1 regulates breast cancer progression (23). However, very few studies have analyzed the correlation of the UHRF1 expression in breast cancer tissue by using large-scale patient population. In the current study, we investigated the role of

Table 2 Correlation analysis between UHRF1 protein level and the clinical characteristics of breast cancer patients

Clinical Pathological Character	UHRF1 protein level*		χ^2	P value
	Low [49]	High [47]		
Age			1.841	0.175
≤60	29	34		
>60	20	13		
Location			3.414	0.065
Left	28	18		
Right	21	29		
ER			4.200	0.040
Negative	14	23		
Positive	35	24		
PR			1.570	0.210
Negative	24	29		
Positive	25	18		
HER-2			0.626	0.429
Negative	33	28		
Positive	16	19		
T stage			0.011	0.917
T1	10	10		
T2–T3	39	37		
N stage#			0.048	0.826
N0–N1	32	31		
N2–N3	15	16		
Clinical stage#			0.047	0.829
I–II	31	30		
III–IV	16	17		
Pathological grade			4.798	0.028
I–II	44	34		
III	5	13		

#, there are two missing values in the UHRF1 low expression group; *, 96 IDC tissues were divided into high-transcription group and low-transcription group by the median value of FPKM of UHRF1. UHRF1, ubiquitin-like PHD and RING finger domain-containing protein 1; IDC, invasive ductal carcinoma.

UHRF1 in the development of breast cancer by integrating the GEO, TCGA datasets, and immunohistochemistry analysis. In this study, we analyzed not only the mRNA expression but also the protein expression of UHRF1 by multiple analyses, which can provide a comprehensive understanding of the role of UHRF1 in breast cancer.

In the GSE10780 and GSE29044 database, we observed that UHRF1 was significantly overexpressed in invasive ductal carcinoma tissue which is the most common pathological type of breast cancer (24). The data from the TCGA dataset showed that UHRF1 expression was increased in invasive ductal carcinoma compared to the

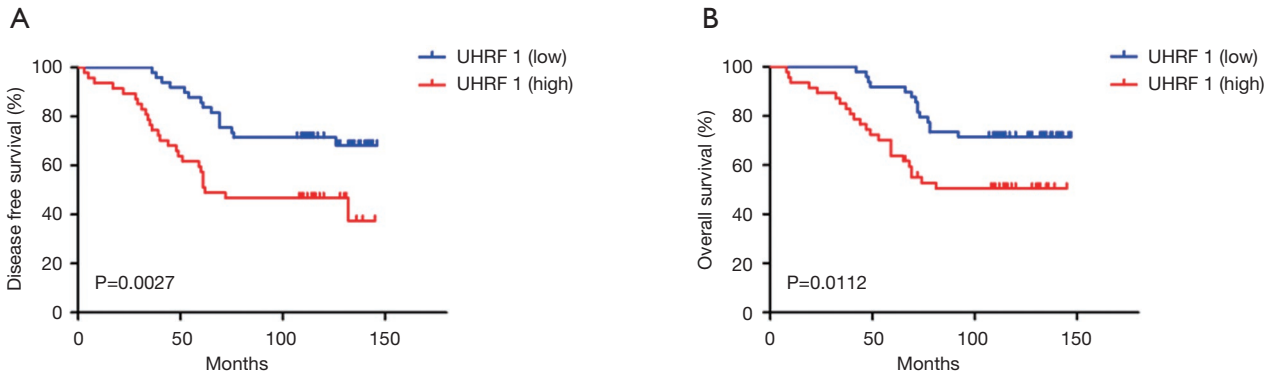

Figure 4 High UHRF1 expression associated with poor survival in IDC. All 96 cases of IDC patients were divided into a UHRF1 low expression group (n=49) and a UHRF1 high expression group (n=47) by the median value of FPKM of UHRF1. (A) Comparison of disease-free survival between the UHRF1 low expression group (n=49) and the UHRF1 high expression group (n=47); (B) comparison of overall survival (OS) between the UHRF1 low expression group (n=49) and the UHRF1 high expression group (n=47). Statistical analysis was performed using log-rank tests.

normal breast tissue samples. Notably, we further explored the expression of UHRF1 in breast cancer tissues via immunohistochemistry to validate the results observed from the data garnered from the GEO and TCGA databases. The results demonstrated that UHRF1 was increased in breast cancer tissues, particularly in IDC. Furthermore, UHRF1 expression in DICS, which is the interim stage in the development of IDC was also analyzed. We found an increasing trend of UHRF1 expression across the DCIS and IDC tissues. However, there is no statistical significance between DCIS and IDC, and this might have been caused by insufficient specimens of DCIS. These integrated analyses indicated that UHRF1 is highly expressed and may be linked to the development of breast cancer.

ERs play an important role in breast cancer development. ER, progesterone receptor (PR), and human epidermal growth factor receptor-2 (HER2) are crucial biomarkers for predicting the response to hormone treatments for breast cancer (25). Compared to ER-positive tumors, triple-negative (ER-negative, PR negative, and Her-2 not overexpressed) breast cancers do not respond to some treatments and tend to be more aggressive (26). Macaluso *et al.* (27) found that the triple-negative breast cancer cell lines, MDA-MD-231 and MDA-MB-361, exhibited a higher expression of UHRF1 (ICBP90) protein levels than MCF-7 in western blotting. No study has investigated the correlation of UHRF1 expression in breast cancer tissue and ER expression by using large amounts of cancer tissues. In the present study, high UHRF1 levels in tissue were significantly associated with an ER-negative status. However, the mechanism by which UHRF1 regulates ER expression remains unclear. However, *UHRF1* gene expression is closely related to the gene methylation levels (28). UHRF1 (ICBP90) can cooperate with pRb2/p130 and regulate the ER-α gene expression (27). Similar to previous findings, we also found that UHRF1 expression is highly correlated to the pathological grade of breast cancer (6).

High expression of UHRF1 is associated with poor prognosis in many cancers (10-12,21,29). In the survival analysis, we found that UHRF1 expression is distinctly related to DFS and OS in breast cancer patients. Similar results were reported in other cancers, including esophageal squamous cell carcinoma (30) and hepatocellular carcinoma (29). In breast cancer, elevated UHRF1 DNA levels in plasma were reported to be directly related with short, progression-free survival (20). The mechanism of UHRF1 leading to poor prognosis might be caused by be promotion of cell proliferation and metastasis since, in one published report, elevated UHRF1 expression contributed to poor prognosis by promoting cell proliferation and metastasis in hepatocellular carcinoma in a published report (10). Gao *et al.* (21) found that the overexpression of UHRF1 was linked to breast cancer progression and poor prognosis by suppression of *KLF17* which plays a pivotal role in the epithelial-mesenchymal transition.

Conclusions

In conclusion, our study assessed the relationship between the expression of UHRF1 and the clinical outcomes of breast cancer patients by analyzing the data from the GEO and TCGA databases and validating those results via an immunohistochemistry analysis. Our results demonstrated that the levels of UHRF1 were increased in breast cancer and were associated with the ER expression and pathological tumor grade. More significantly, highly expressed UHRF1 predicted a poor prognosis. Although further studies are needed, our findings indicate that UHRF1 might promote tumorigenesis and the development of breast cancer.

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Footnote

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at http://dx.doi.org/10.21037/tcr.2019.06.19). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. This study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The study was approved by the Medical Ethics Committee of Xinxiang Central Hospital. Individual informed consent was waived.

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