

# Systematic analysis to identify a key role of CDK1 in mediating gene interaction networks in cervical cancer development

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## Abstract

**Objective** This study aims to identify corresponding differentially expressed genes in cervical cancer by comparing gene expression profiles between normal and cervical cancer samples.

**Method** To identify differentially expressed genes in cervical cancer, two groups of Affymetrix microarray data available online were analyzed. One group consisted of 43 carcinomatous cervical epithelial cell samples, and the other was composed of 17 healthy cervical epithelial cell samples, both from the Amerindian. R packages—GO.db, KEGG.db and KEGGREST were used to detect GO categories and KEGG pathways with significant overrepresentation in differentially expressed genes comparing with the whole genome. Cytoscape was utilized to construct biological networks.

**Results** By comparing gene expression profile of normal and cervical cancer samples, 122 differentially expressed genes were identified including 46 up-regulated genes and 76 down-regulated genes. Using the identified differentially expressed genes, a large and a small biological network was constructed. In addition, 402 GO biological

processes and 9 KEGG pathways were over-represented. Top significant biological processes included cell cycle and cell proliferation. Moreover, top significant KEGG pathways were oocyte meiosis, cell cycle and progesterone-mediated oocyte maturation. Most importantly, *CDK1* frequently appeared in these processes and pathways, which indicated its significant role in the progression of cervical cancer.

**Conclusion** *CDK1* plays a comprehensive role in mediating genetic networks implicated in the progression of cervical cancer. Novel therapeutics targeting *CDK1* or its related pathways might help improve prognosis of advanced stage cervical cancer.

**Keywords** Cervical cancer development · Systematic analysis · Gene expression profiles

## Introduction

As the second most common cancer among women worldwide, cervical cancer is a major threat to our health, especially in developing countries where active screening and human papillomavirus (HPV) vaccines have not yet become available to every individual. Although most patients can be cured if diagnosed at an early stage, those with advanced stage disease are at a high risk of secondary metastatic cancer and tumor relapse [1]. Discovery of biomarkers for early diagnosis and novel effective therapeutic modalities against advanced stage cervical cancer is urgently required. To this end, uncovering the underlying mechanisms of tumorigenesis and progression of cervical cancer is of great importance. Tumorigenesis is a complicated pathological process which involves multiple genetic alterations such as overexpression of oncogenes

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and inactivation of tumor-suppressor genes [2]. Expression profiling studies using microarrays have been extensively used in identifying biomarkers for early cancer diagnosis, classifying cancer subtypes, and predicting response to anticancer therapies [3]. Identification of genes with multiple roles in dysregulated cancer pathways shed light on tumorigenesis and cancer progression, thus helping invent novel anticancer strategies. In terms of cervical cancer, earlier studies comparing expression profiles between normal cervix and cervical cancer samples identified many differentially expressed genes (DEGs). However, DEGs reported in different studies vary greatly with only a few consistently detected among various studies [3–5]. Although these studies provide evidence of several DEGs as valuable potential biomarkers for diagnosis and prognosis, due to their limited role in disease progression, few of the identified DEGs led to the development of novel therapeutics. The objectives of this study were to verify DEGs in cervical cancer and identify significantly changed biological networks and KEGG pathways, with a special focus on uncovering possible hub genes which play multiple roles in tumorigenesis and disease progression of cervical cancer. In the study, to detect DEGs in cervical cancer, two groups of Affymetrix microarray data were analyzed. One group consisted of carcinomatous cervical epithelial cell samples, and the other had healthy cervical epithelial cell samples, both from the Amerindian. In addition, gene ontology (GO) biological process and KEGG pathway analysis using identified DEGs were conducted, and corresponding heat maps and biological networks were constructed.

## Methods

### Data collection

Gene expression omnibus (GEO) database (<http://www.ncbi.nlm.nih.gov/geo/>) was searched and Affymetrix microarray expression data (GSE 29570), which contained two groups, were obtained [6]. One group with 43 samples consisted of carcinomatous cervical epithelial cells and the other with 17 samples of healthy cervical epithelial cells, both from the Amerindian. Unprocessed data sets (.cel files) were collected for further analysis. The Affymetrix Human Gene 1.0 ST Array [transcript (gene) version] was applied in the experiments. The probe annotation files were downloaded accordingly for further research. This study was approved by the Ethics Committee of the Affiliated Hospital of North Sichuan Medical College and the Ethics Committee concluded that the study would not cause harm or risk to the subjects.

### Data processing and filtering

Considering the array platform, we applied RMA in this research to quantify microarray signal [7]. The normalization process mainly consisted of three steps: model-based background correction, quantile normalization and summarizing. To filter out uninformative data such as control probesets and other internal controls as well as removing genes which were expressed uniformly close to background detection levels, the *genefilter* package in R language provides *nsFilter* function to do this. However, the filter does not remove probesets without Entrez Gene identifiers or with identical Entrez Gene identifiers.

### DEGs analysis

Statistical comparison between normal samples and carcinomatous samples was conducted. Limma in R language was applied to identify the differential expression of the comparison. For probes which had identical Entrez Gene identifiers, only the probe occupying the biggest variance was kept for further DEG analysis. Only those genes with absolute  $\log_2$  (fold change)  $>2$  and adjusted  $p$  value  $<0.01$  were recognized as statistically differentially expressed between two sample groups. The adjusted  $p$  value was obtained by applying Benjamini and Hochberg's (BH) false discovery rate correction on the original  $p$  value, and fold change threshold was selected based on our purpose of focusing on significantly differentially expressed genes.

### Hierarchical clustering

Hierarchical clustering was performed to classify the analyzed samples based on gene expression profiles [8]. Hierarchical clustering was carried out using DEGs to observe the global gene expression patterns. Besides, the DEGs, which were classified in specific GO biological processes and KEGG pathway analysis, were further extracted and the expression pattern of those DEGs was characterized, and the heat maps for the DEGs classified in targeted biological processes or KEGG pathways using R package were also constructed.

### GO and KEGG pathway analysis

R packages—GO.db, KEGG.db and KEGGREST were used to detect GO categories and KEGG pathways with significant overrepresentation in DEGs comparing with the whole genome. The significantly enriched biological processes were identified as  $p$  value less than threshold value 0.01. As to KEGG pathway,  $p$  value was set to less than 0.05.

### Construction of biological network

Network database was downloaded from HPRD, BIOGRID, and PIP [9–11]. The pair interactions, which were included in any of the three databases, were chosen to be in our curated database. Cytoscape was utilized to construct biological network [12]. Interacted gene pairs existed in the curated database was imported as stored network. After the functional enrichment analysis, DEGs specified in dramatically altered biological processes and KEGG pathways were mapped to corresponding network, respectively, to analyze the interactions.

## Results

### Differential expression analysis

Comparative analysis was performed between normal and carcinomatous samples to identify genes with significantly

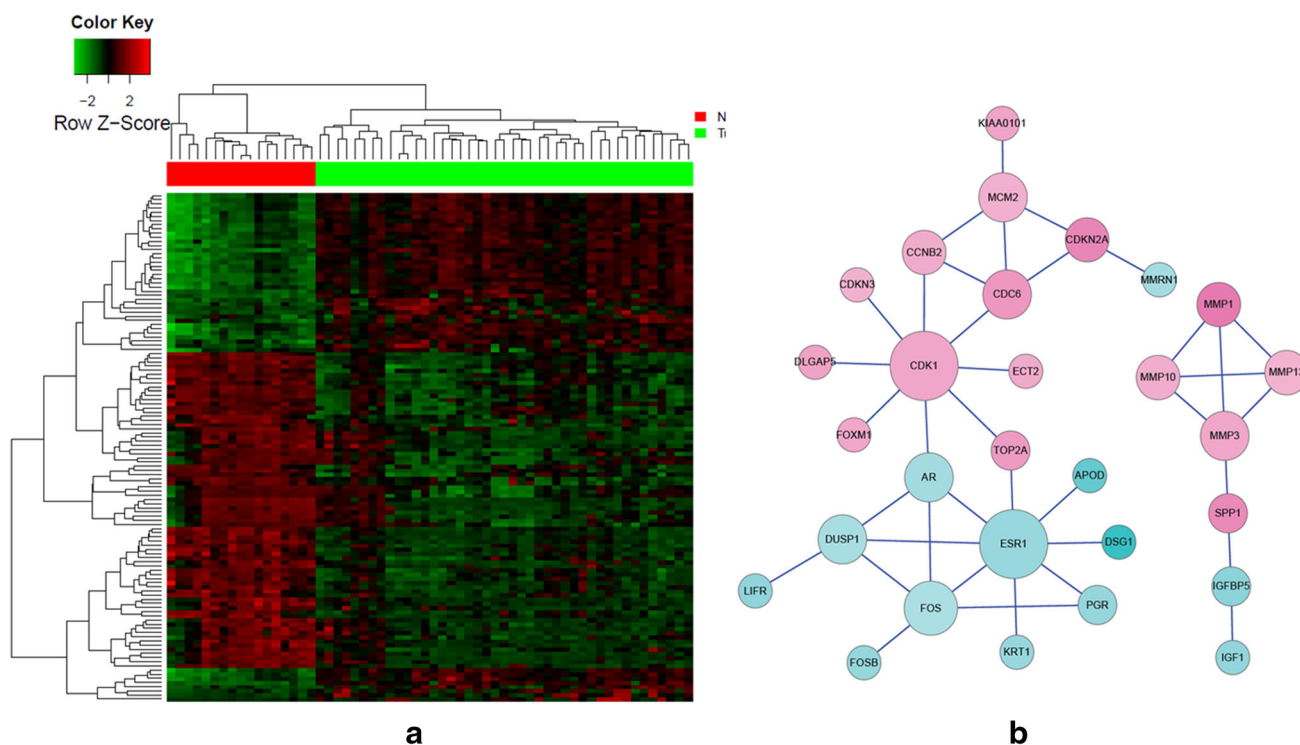
differential expression levels. Setting the threshold as absolute  $\log_2(\text{fold change}) > 2$  and adjusted  $p$  value  $< 0.01$ , 122 DEGs were identified, among which 46 were up-regulated and 76 were down-regulated (Table 1, Supplementary Table 1, 2). The top 5 up-regulated genes were *MMP12*, *SYCP2*, *SMC1B*, *MMP1* and *CDKN2A*. The top 5 down-regulated genes were *SPINK7*, *CRISP3*, *TMPRSS11B*, *CRNN* and *DSG1*.

### Construction of biological network

DEGs identified were utilized to construct biological network (Fig. 1), since the size of DEGs was relatively small. The interactions of genes could be observed clearly. Two separated sub-networks were indicated as shown and the larger one centered on *CDK1* and *ESR1*. Moreover, the differential expression was considerably consistent. It was interesting to see that most of the genes connected with *CDK1* such as *CDC6*,

**Table 1** Statistical distribution of differentially expressed genes in normal and tumor samples

	Probe	Gene	
All	9991	9571	
log FC  > 2 and adj. p value < 0.01	125	46 (up)	122
		79 (down)	76 (down)



**Fig. 1** Heat map of DEGs (a) and corresponding biological network (b). **a** Heat map of hierarchical clustering of all DEGs (17 normal samples and 43 tumor samples). Red indicates high relative expression, and green indicates low relative expression. **b** Biological

network was constructed according to the direct connections among all DEGs. Red indicates high relative expression, and green indicates low relative expression

*CDKN3*, *ECT2* and *TOP2A* were up-regulated, whereas those connected with *ESR1* such as *AR*, *FOS*, *DSG1* and *KRT1* were down-regulated. Genes connected with *CDK1* are mainly involved in cell cycle and cell proliferation. The smaller biological network was mainly composed of genes of *MMP* family such as *MMP1* and 3.

### GO and KEGG pathway analysis

402 GO biological processes and 9 KEGG pathways were over-represented by DEGs (Table 2). Top significant biological processes included cell cycle process, cell proliferation, tissue development, nuclear division, and cell division (Table 3). Moreover, top significant KEGG pathways were oocyte meiosis, cell cycle, p53 signaling pathway, pathways in cancer, and progesterone-mediated oocyte maturation (Table 4). Heat maps and biological networks of significantly enriched GO biological processes or KEGG pathways were constructed (Figs. 2, 3, 4, 5). Obviously, *CDK1* frequently appeared in these processes, which indicated its significant role in the progression of cervical cancer.

**Table 2** Obtained GO biological processes and KEGG pathways

	<i>p</i> value	Number
GO biological process	<0.01	402
KEGG pathway	<0.05	9

**Table 3** Several significantly changed GO biological processes

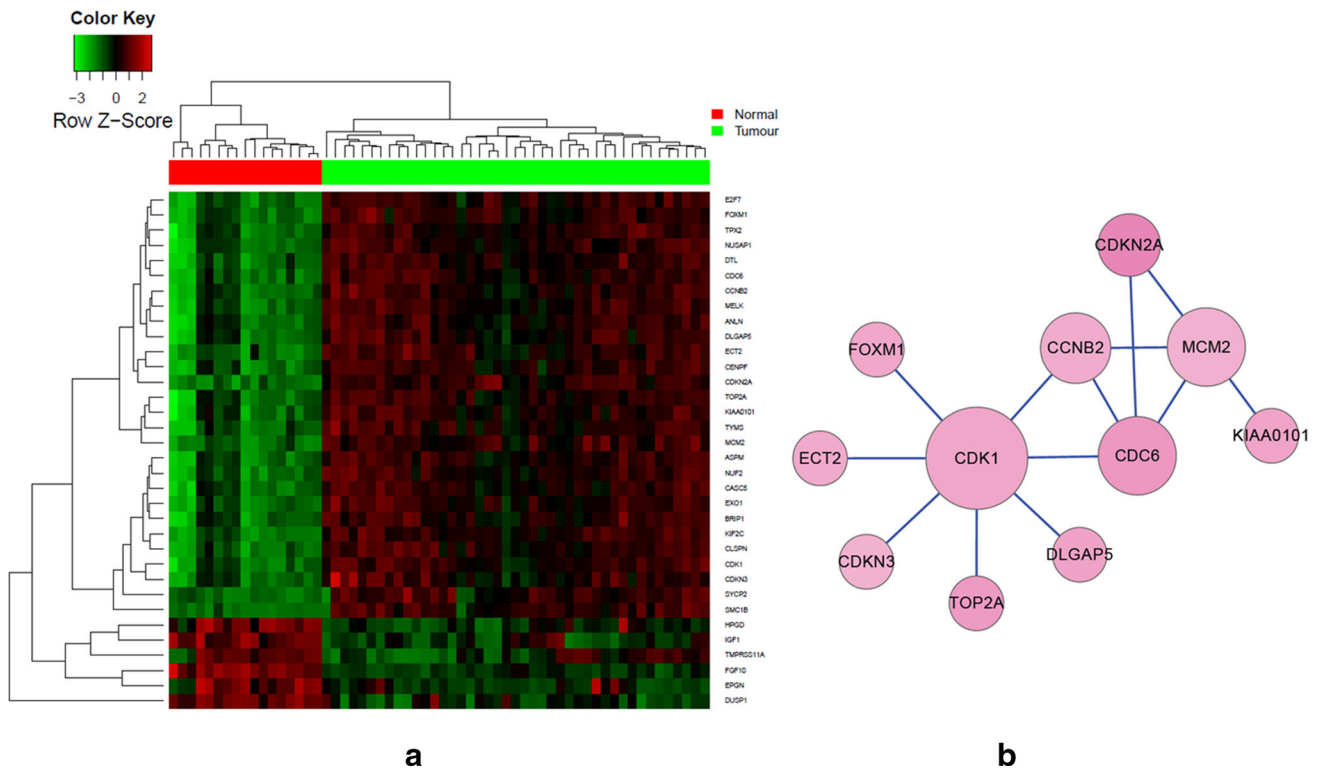
GO-BP-ID	<i>p</i> value	Count	Term
GO:0022402	5.91E-11	30	Cell cycle process
GO:0000278	9.13E-11	26	Mitotic cell cycle
GO:0007049	1.66E-10	34	Cell cycle
GO:0009888	5.24E-10	32	Tissue development
GO:0008283	3.00E-09	34	Cell proliferation
GO:0000280	6.68E-09	18	Nuclear division
GO:0030855	1.13E-08	18	Epithelial cell differentiation
GO:0051301	1.47E-08	21	Cell division

**Table 4** Several significantly changed KEGG pathways

KEGGID	<i>p</i> value	Count	Term
04114	9.54E-05	6	Oocyte meiosis
04110	1.68E-04	6	Cell cycle
04115	1.10E-03	4	P53 signaling pathway
04914	2.63E-03	4	Progesterone-mediated oocyte maturation
05200	5.82E-03	7	Pathways in cancer

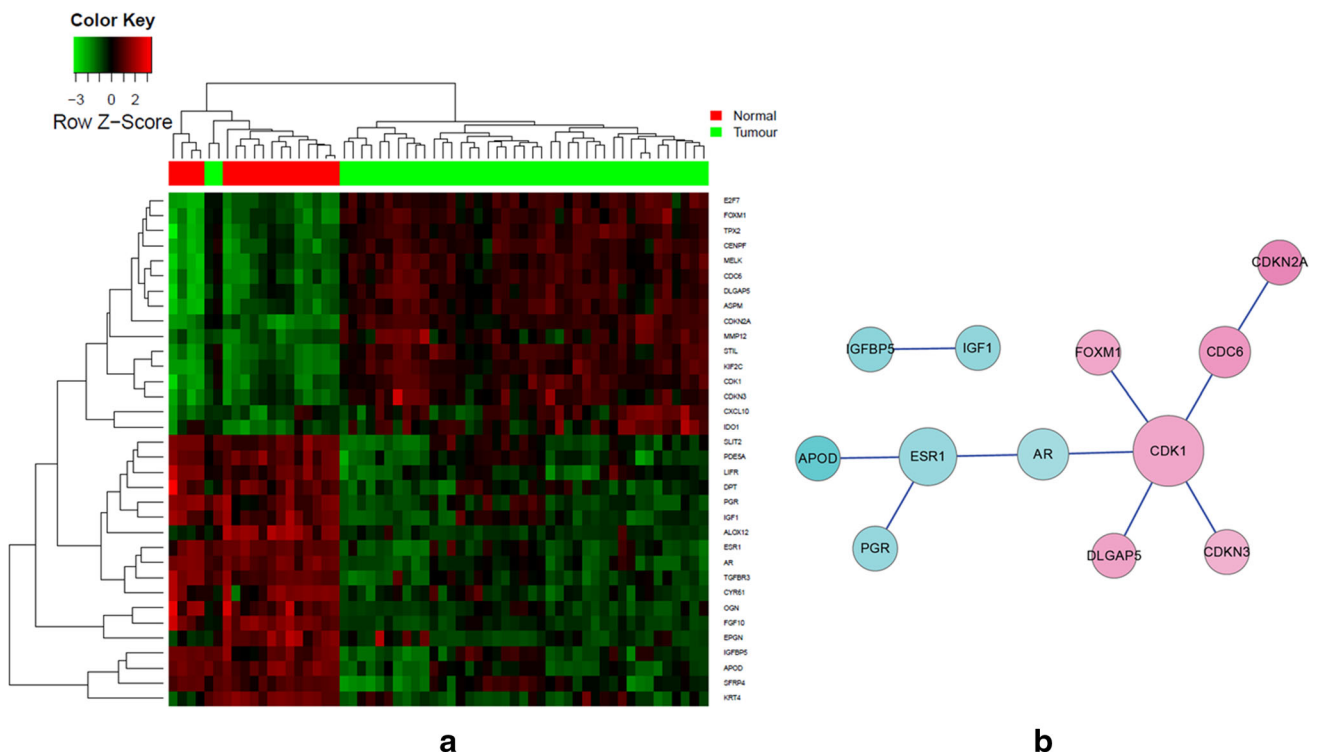
### Discussion

One objective of this study was to identify corresponding DEGs between normal cervix and cervical cancer. By comparing gene expression profiles of 17 normal cervical epithelial samples and 43 carcinomatous cervical samples, 122 DEGs were identified including 46 up-regulated and 76 down-regulated genes. Referring to results of previous profiling studies in cervical cancer, many DEGs identified in this study such as *SPPI*, *MMP12*, *CDKN2A*, and *CDK1* were previously reported [3, 4, 13, 14]. In the study, 402 GO biological processes and 9 KEGG pathways were over-represented by DEGs. While cell cycle process, cell proliferation, tissue development, nuclear division, and cell division were revealed as the most significantly up-regulated biological processes, the top KEGG pathways were pathways in cancer, oocyte meiosis, cell cycle, p53 signaling pathway, and progesterone-mediated oocyte maturation. Our results were consistent with previous findings. A previous study conducted by Seo and colleagues using mRNA differential display and GO analysis demonstrated that cervical cancer was present with complete up-regulation of cell cycle, transport, protein biosynthesis, and RNA metabolism [15]. On the other hand, up-regulation of pathways in cancer, metabolic pathways, and progesterone-mediated oocyte maturation in cervical cancer was also previously reported [3]. In fact, dysregulation of cell cycle, cell proliferation, and p53 signaling pathway are present in most types of cancer. Under normal circumstances, cell cycle progression is tightly regulated by groups of proteins that interact with each other in a very specific sequence of events. In cancer cells, some of these proteins no longer function properly leading to dysregulated cell cycle progression and cell proliferation. Described as the guardian of the genome, the p53 signaling pathway contains an array of genes whose protein products are very important in maintaining cellular homeostasis. Mutations in p53 signaling pathways are very frequent in cancers. Using the identified DEGs, a large and a small biological network were constructed. The up-regulated genes detected in the smaller network are members of *MMP* family and *SPPI*. Matrix metalloproteinases (MMPs), are known to help induce tumor metastasis by involving in tumor angiogenesis, local invasion and establishment of metastatic lesions at distant



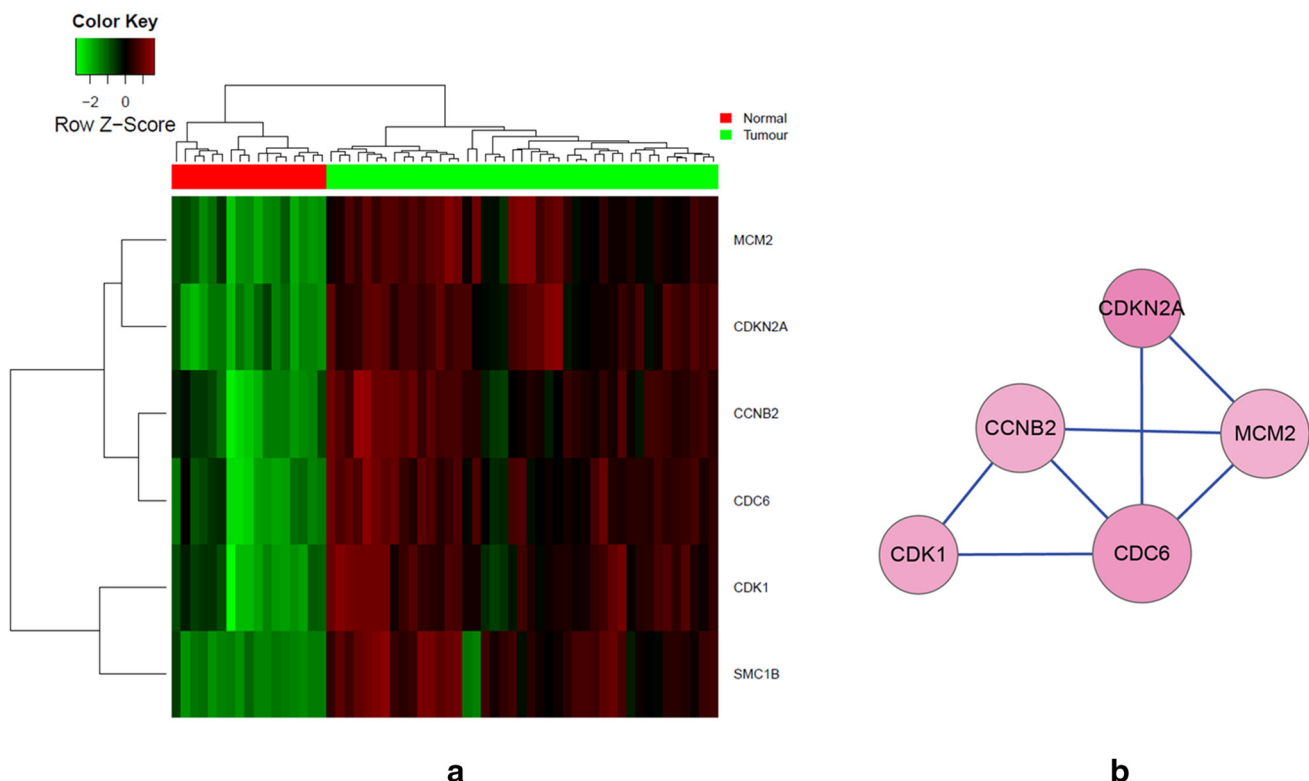
**Fig. 2** Heat map of GO: 0007049/cell cycle (a) and corresponding biological network (b). a Heat map of hierarchical clustering of all data sets (17 normal samples and 43 tumor samples) using 34 DEGs

in “cell cycle”. b Biological network was constructed according to the direct connections among corresponding DEGs



**Fig. 3** Heat map of GO: 0008283/cell proliferation (a) and corresponding biological network (b). a Heat map of hierarchical clustering of all data sets (17 normal samples and 43 tumor samples) using 34 DEGs in “cell proliferation”. Red indicates high relative

expression, and green indicates low relative expression. b Biological network were constructed according to the direct connections among corresponding DEGs

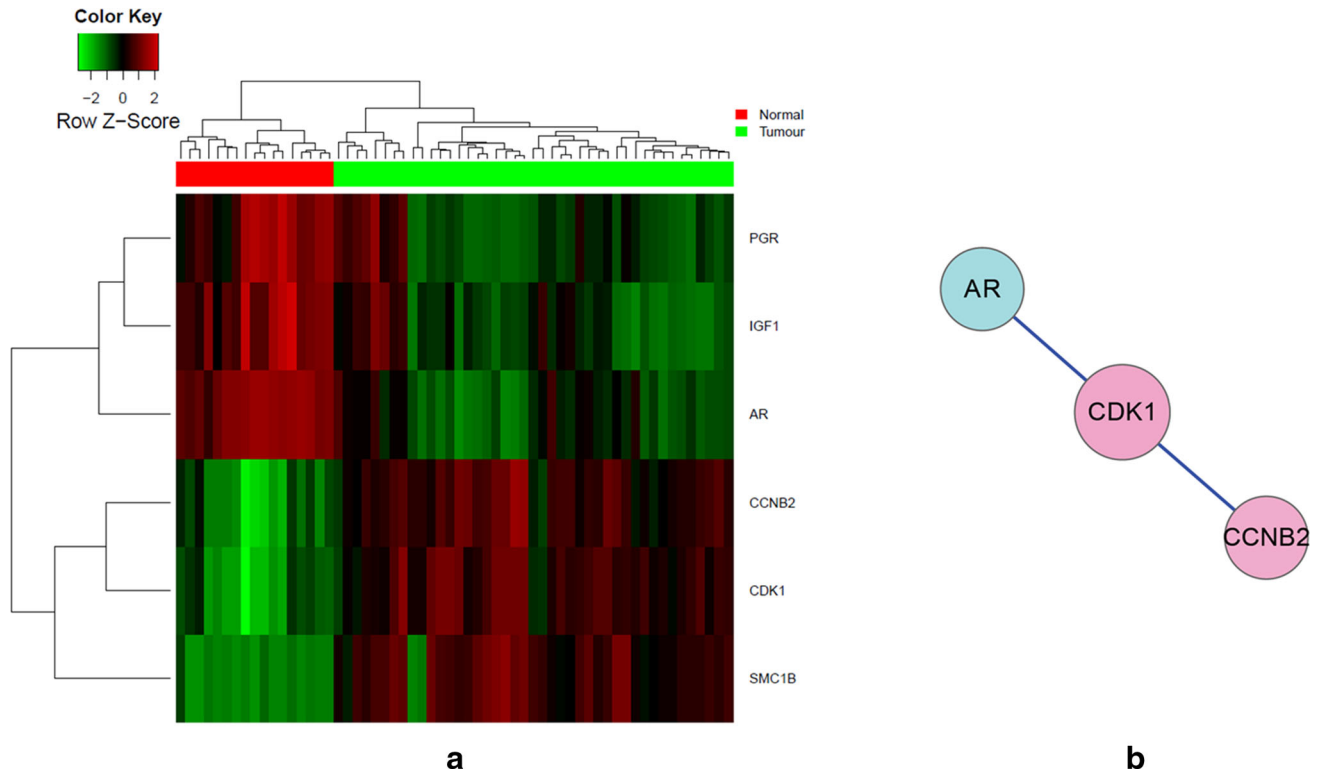


**Fig. 4** Heat map of KEGG: 04110/cell cycle (**a**) and corresponding biological network (**b**). **a** Heat map of hierarchical clustering of all data sets (17 normal samples and 43 tumor samples) using 6 DEGs in

“cell cycle”. **b** Biological network were constructed according to the direct connections among corresponding DEGs

secondary site [16]. For example, research has shown that membrane type 1 MMP (MT1-MMP) is involved in cervical cancer carcinoma progression and invasion [17]. In a separate study, overexpression of *MMP-1*, *MMP-2*, *MMP-9*, *MMP-14* and *MMP-15* were detected in more than 50 % of primary cervical cancer samples. More importantly, progressively up-regulated expression of *MMP-2* and *MMP-9* is closely associated with progression and recurrence of human cervical cancer [18]. Our results further confirmed that MMP family is closely associated with progression and invasion of cervical cancer. In addition to the MMPs, *SPP1* (secreted phosphoprotein 1), also known as osteopontin, which encodes an extracellular glycosylated bone phosphoprotein, was another up-regulated gene identified in the smaller biological network. In fact, up-regulation of *SPP1* in cervical cancer was detected in a number of previous studies [3, 5, 19]. In a study conducted by Cho and colleagues, diagnostic and prognostic significance of increased plasma *SPP1* level in cervical cancer patients were evaluated. Their results suggest that it is a potential diagnostic and prognostic biomarker for cervical cancer [20]. In a separate study, *SPP1* was also suggested as an effective biomarker of disease progression of cervical cancer [3].

There were 11 up-regulated genes in the larger biological network. The functions of these genes are mainly involved in regulation of cell cycle, cell proliferation, and oocyte meiosis. Dysregulation of most of these genes has been reported in cervical cancer studies [21, 22]. Intriguingly, up-regulation of *CDK1* was found to be involved in all up-regulated biological networks with direct interactions with most of the 11 DEGs. In the cell cycle network, *CDK1* was directly linked to *CCNB2* and *CDC6*. In the cell proliferation network, direct interactions of *CDK1* with *CDC6*, *CDKN3*, *DLGAP5*, *FOXMI* were revealed. In oocyte meiosis, we found interconnections among *CDK1*, *CCNB2*, and *AR*. Taken together; these results suggest that *CDK1* plays a pivotal role in mediating cervical cancer genetic networks in tumorigenesis and disease progression. The finding is not surprising. It is well known that HPV infection is one of the most important factors in carcinogenesis of cervical cancer. After infection, E6 and E7 viral oncogenic proteins inhibit p53-mediated apoptosis and disrupt regulations of cell cycle by inducing rapid proteasomal degradation of p53 [23]. As a tumor-suppressor protein, p53 regulates the transcription of regulatory genes involved in apoptosis and cell cycle arrest. Under normal circumstances, p53 negatively regulates *CDK1*



**Fig. 5** Heat map of KEGG: 04114/oocyte meiosis (a) and corresponding biological network (b). **a** Heat map of hierarchical clustering of all data sets (17 normal samples and 43 tumor samples) using 6 DEGs in “oocyte meiosis”. *Red* indicates high relative

expression, and *green* indicates low relative expression. **b** Biological network were constructed according to the direct connections among corresponding DEGs

transcription [24]. In cervical cancer, abrogation of p53 function due to HPV infection leads to overexpression of a number of its downstream genes such as *SPP1* [5, 25, 26]. Results from a number of studies have suggested that therapeutics which can effectively restore p53 level in cancer cells have significant anticancer effects. As a direct downstream gene of *p53*, overexpression of *CDK1* in cervical cancer was previously reported. Cyclin-dependent kinase 1 (Cdk1), a highly conserved protein, functions as a serine/threonine kinase. It plays a pivotal role in the control of cell cycle with more than 70 regulatory targets. In response to various stimuli, Cdk1 directly phosphorylates a variety of target substrates that are involved in the control of transcription and cell cycle progression [27].

Studies have shown that aberrant activation of CDKs and their modulators are present in many cancers. Dysregulation of CDKs causes abnormal cell proliferation and genomic instability [28]. In fact, the D-cyclin-cdk4/6-INK4-Rb pathway is disrupted in all human cancers [29]. A study comprised of both in vitro and in vivo anticancer assays showed IPP5, a novel inhibitor of protein phosphatase, successfully inhibited growth of Hela cells by regulating expression levels of cell cycle regulators

including Cdk1, cyclin A1, cyclinB1, p21 and p53 [30]. Their results further indicated that aberrant cell cycle regulation is a major pathological event in the progression of cervical cancer. Previous studies have proposed a number of genes such as *p53*, *SPP1*, *MMP1*, and *CDKN2A* as very important genes in cervical cancer with great diagnostic and therapeutic values [3, 13, 19]. Based on our data, due to its extensive involvement in aberrantly regulated pathways present in cervical cancer, *CDK1* is obviously another important hub gene in cervical cancer, which might help develop novel therapeutics. In fact, studies have indicated that anticancer drugs targeting aberrant cell cycle regulation have great therapeutic potentials. For example, treatment of JNJ-7706621, a cell cycle inhibitor, inhibited CDK1 activity, changed its phosphorylation status, and arrested cell cycle at the G2/M phase. Data from both in vitro and in vivo studies showed that the CDK inhibitors have significant anticancer effect against a number of cancers including cervical, colon, and breast cancer [31, 32]. A recent study showed that oridonin, an active compound extracted from the plant *Rabdosia rubescens*, inhibited proliferation of gastric cancer cell by inducing cell cycle arrest in the G2/M phase. It was

suggested that the cell cycle arrest might be induced by decreased expression of CDK2 and CyclinB1 [33].

In summary, in the current study, 122 DEGs were detected by comparing gene expression files between normal and cervical cancer samples. Among the 122 DEGs, a number of genes were previously reported such as *SPP1*, *CDKN2A*, and *CDK1*. In addition, several significantly changed biological networks and KEGG pathways were identified. The most important finding was that *CDK1* plays a comprehensive role in modulating genetic networks implicated in tumorigenesis, disease progression, and metastasis of cervical cancer. To date, only a few genes have been reported to have multiple impacts on progression of cervical cancer such as *p53*, *pRb*, and *SPP1*. Our finding suggests that novel therapeutics targeting *CDK1* or its related pathways might help improve prognosis of advanced stage cervical cancer. However, due to *CDK1*'s global regulatory functions in cell cycle, inhibition of *CDK1* is likely to induce severe toxicities and side effects in patients. As a result, strategies focusing on its downstream regulation pathways might be more promising options.

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**Conflict of interest** This manuscript represents original work that has not been published nor is in press elsewhere. All authors have approved the submission of the manuscript, and no conflict of interest is declared.

## References

- Petignat P, Roy M (2007) Diagnosis and management of cervical cancer. *BMJ* 335(7623):765–768
- Vogelstein B, Kinzler KW (2004) Cancer genes and the pathways they control. *Nat Med* 10(8):789–799
- Thomas A et al (2013) Expression profiling of cervical cancers in Indian women at different stages to identify gene signatures during progression of the disease. *Cancer Med* 2(6):836–848
- Cheng Q et al (2002) Identification and characterization of genes involved in the carcinogenesis of human squamous cell cervical carcinoma. *Int J Cancer* 98(3):419–426
- Wong YF et al (2006) Genome-wide gene expression profiling of cervical cancer in Hong Kong women by oligonucleotide microarray. *Int J Cancer* 118(10):2461–2469
- Guardado-Estrada M et al (2012) The Amerindian mtDNA haplogroup B2 enhances the risk of HPV for cervical cancer: deregulation of mitochondrial genes may be involved. *J Hum Genet* 57(4):269–276
- Carvalho BS, Irizarry RA (2010) A framework for oligonucleotide microarray preprocessing. *Bioinformatics* 26(19):2363–2367
- Tavazoie S et al (1999) Systematic determination of genetic network architecture. *Nat Genet* 22(3):281–285
- Keshava Prasad TS et al (2009) Human protein reference database–2009 update. *Nucleic Acids Res* 37(Database issue):D767–D772
- Chatr-Aryamontri A et al (2013) The BioGRID interaction database: 2013 update. *Nucleic Acids Res* 41(Database issue):D816–D823
- McDowall MD, Scott MS, Barton GJ (2009) PIPs: human protein-protein interaction prediction database. *Nucleic Acids Res* 37(Database issue):D651–D656
- Smoot ME et al (2011) Cytoscape 2.8: new features for data integration and network visualization. *Bioinformatics* 27(3):431–432
- Santin AD et al (2005) Gene expression profiles of primary HPV16- and HPV18-infected early stage cervical cancers and normal cervical epithelium: identification of novel candidate molecular markers for cervical cancer diagnosis and therapy. *Virology* 331(2):269–291
- Rajkumar T et al (2011) Identification and validation of genes involved in cervical tumorigenesis. *BMC Cancer* 11:80
- Seo MJ et al (2005) New approaches to pathogenic gene function discovery with human squamous cell cervical carcinoma by gene ontology. *Gynecol Oncol* 96(3):621–629
- Deryugina EI, Quigley JP (2006) Matrix metalloproteinases and tumor metastasis. *Cancer Metastasis Rev* 25(1):9–34
- Zhai Y et al (2005) Expression of membrane type 1 matrix metalloproteinase is associated with cervical carcinoma progression and invasion. *Cancer Res* 65(15):6543–6550
- Sheu BC et al (2003) Increased expression and activation of gelatinolytic matrix metalloproteinases is associated with the progression and recurrence of human cervical cancer. *Cancer Res* 63(19):6537–6542
- Song JY et al (2008) Microarray analysis of normal cervix, carcinoma in situ, and invasive cervical cancer: identification of candidate genes in pathogenesis of invasion in cervical cancer. *Int J Gynecol Cancer* 18(5):1051–1059
- Cho H et al (2008) Clinical significance of osteopontin expression in cervical cancer. *J Cancer Res Clin Oncol* 134(8):909–917
- Conesa-Zamora P (2013) Role of cell cycle biomarkers in human papillomavirus related uterine lesions. *Curr Pharm Des* 19(8):1412–1424
- Lau E et al (2007) The role of pre-replicative complex (pre-RC) components in oncogenesis. *FASEB J* 21(14):3786–3794
- Burd EM (2003) Human papillomavirus and cervical cancer. *Clin Microbiol Rev* 16(1):1–17
- Yun J et al (1999) p53 negatively regulates cdc2 transcription via the CCAAT-binding NF-Y transcription factor. *J Biol Chem* 274(42):29677–29682
- Cho NH et al (2006) Elevation of cyclin B1, active cdc2, and HuR in cervical neoplasia with human papillomavirus type 18 infection. *Cancer Lett* 232(2):170–178
- Cerciello F et al (2005) G2/M cell cycle checkpoint is functional in cervical cancer patients after initiation of external beam radiotherapy. *Int J Radiat Oncol Biol Phys* 62(5):1390–1398
- Malumbres M, Barbacid M (2005) Mammalian cyclin-dependent kinases. *Trends Biochem Sci* 30(11):630–641
- Hall M, Peters G (1996) Genetic alterations of cyclins, cyclin-dependent kinases, and Cdk inhibitors in human cancer. *Adv Cancer Res* 68:67–108
- Shapiro GI (2006) Cyclin-dependent kinase pathways as targets for cancer treatment. *J Clin Oncol* 24(11):1770–1783
- Zeng Q et al (2012) IPP5, a novel inhibitor of protein phosphatase 1, suppresses tumor growth and progression of cervical carcinoma cells by inducing G2/M arrest. *Cancer Genet* 205(9):442–452



31. Emanuel S et al (2005) The in vitro and in vivo effects of JNJ-7706621: a dual inhibitor of cyclin-dependent kinases and aurora kinases. *Cancer Res* 65(19):9038–9046
32. Sutherland RL, Musgrove EA (2009) CDK inhibitors as potential breast cancer therapeutics: new evidence for enhanced efficacy in ER + disease. *Breast Cancer Res* 11(6):112
33. Gao SY et al (2014) Downregulation of Cdk1 and CyclinB1 Expression Contributes to Oridonin-induced Cell Cycle Arrest at G2/M Phase and Growth Inhibition in SGC-7901 Gastric Cancer Cells. *Asian Pac J Cancer Prev* 15(15):6437–6441