Role of CITED2 in stem cells and cancer (Review)

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Abstract. Cbp/P300 interacting transactivator with Glu/Asp-rich carboxy-terminal domain 2 (CITED2) is a transcription co-factor that interacts with several other transcription factors and co-factors, and serves critical roles in fundamental cell processes, including proliferation, apoptosis, differentiation, migration and autophagy. The interacting transcription factors or co-factors of CITED2 include LIM homeobox 2, transcription factor AP-2, SMAD2/3, peroxisome proliferator-activated receptor γ, oestrogen receptor, MYC, Nucleolin and p300/CBP, which regulate downstream gene expression, and serve important roles in the aforementioned fundamental cell processes. Emerging evidence has demonstrated that CITED2 serves an essential role in embryonic and adult tissue stem cells, including hematopoietic stem cells and tendon-derived stem/progenitor cells. Additionally, CITED2 has been reported to function in different types of cancer. Although the functions of CITED2 in different tissues vary depending on the interaction partner, altered CITED2 expression or altered interactions with transcription factors or co-factors result in alterations of fundamental cell processes, and may affect stem cell maintenance or cancer cell survival. The aim of this review is to summarize the molecular mechanisms of CITED2 function and how it serves a role in stem cells and different types of cancer based on the currently available literature.

1. Introduction

Cancer is one of the leading causes of mortality worldwide, and has been the second highest cause of mortality in the USA in recent years (1,2). Cancer is a group of diseases that is characterised by aberrant and uncontrolled growth of tissues or cells. The development of cancer is attributed to dysregulation of cell proliferation, apoptosis, differentiation, migration and autophagy (3). Transcription factors and their co-factors are the basic machinery controlling cell processes. Activity of several transcription factors is altered in a number of types of cancer through numerous and varying mechanisms, including chromosomal translocations, gene amplification or deletion, point mutations and dysregulated expression, and indirectly through non-coding DNA mutations that affect transcription factor binding (4,5). These transcription factors are also considered candidate oncogenic genes. Furthermore, transcription co-factors alter the activity of transcription factors by interacting with them, serving critical roles in cancer (5).

Cbp/P300 interacting transactivator with Glu/Asp-rich carboxy-terminal domain 2 (CITED2) is a protein encoded by the Cited2 gene and is a transcription co-factor. The Cited2 gene was cloned around two decades ago (6,7), and was reported to promote the development of cancer when overexpressed in cells in vitro (7). CITED2 is essential for mouse embryonic development, as deletion of CITED2 results in embryonic lethality around embryonic day 10.5 (8). CITED2 is also essential for the development of the liver, lungs, heart, neural tube, left-right patterning and eye development (8-13). CITED2 modulates gene transcription by directly or indirectly interacting with transcription factors or co-factors without a DNA binding motif (14-22). Cited2-null mouse embryonic fibroblasts exhibited premature arrest of proliferation (senescence) (23), which suggests that CITED2 is essential for cell proliferation. Taken together, these findings demonstrated that CITED2 serves a critical role in several fundamental cellular processes.

In addition to the critical role of CITED2 in several fundamental cell processes, CITED2 has been reported to serve roles in numerous different types of cancer. For example, lung cancer (21), gastric cancer (24) and breast cancer (19,25,26). Cancer stem cells (CSCs) are hypothesized to be a population of cells with multipotent stem cell-like properties, and can cause cancer relapse, metastasis, multidrug resistance and radiation resistance through their ability to arrest in the G0 phase, giving rise to new tumours when they finally leave
cell-cycle arrest (27-29). CSCs exhibit strong self-renewal capacity, in a similar way to normal stem cells (30). As the function of CITED2 in CSCs has not been extensively studied, reviewing the function of CITED2 in stem cells may provide directions for future studies. In the present review, the molecular mechanisms of CITED2 function, and the role of CITED2 in stem cells and different types of cancer are discussed.

2. Molecular mechanisms of CITED2 function

CITED2 is a transcriptional co-regulator without a DNA binding domain, that can directly interact with a host of transcription factors and co-factors, including LIM homeobox 2, transcription factor AP-2 (TFAP2), SMAD2/3, peroxisome proliferator-activated receptor (PPAR)-γ, oestrogen receptor, MYC, Nucleolin and p300/CBP, thus regulating the ability of these binding partners to activate or inactivate gene transcription (14-19,21,22). The molecular mechanism of CITED2 function varies based on the type of tissue and the binding partner. In this section, the molecular function of CITED2 in general is briefly summarized. The transcription factors interacting with CITED2 or regulated by CITED2 are listed in Table I. CITED2 was originally found to displace p300/CBP from hypoxia-inducible factor (HIF)-1α, thereby negatively regulating HIF-1α function (14). HIF-1α functions as a master regulator of cellular and systemic homeostatic response to hypoxia by activating transcription of a number of genes, including those involved in energy metabolism, angiogenesis, apoptosis, and other genes whose protein products increase oxygen delivery or facilitate metabolic adaptation to hypoxia (31). HIF-1α also serves an important role in stem cells and cancer (32-34). The molecular mechanisms through which CITED2 interacts with p300/CBP to inhibit HIF-1α function has been intensively studied (35-39). CITED2 acts as a bridge, directly interacting with and co-activating TFAP2 and the p300/CBP transcriptional co-activator complex to stimulate TFAP2-mediated transcriptional activation (8,12,16,40,41). CITED2 positively regulates TGF-β signalling through its association with the SMAD2/3-mediated transcriptional co-activator complex, and upregulates the expression of downstream targets, including matrix metalloproteinase (MMP)-9 and vascular endothelial growth factor (VEGF) (18,42). CITED2 co-activates PPAR-α and PPAR-γ transcriptional activities (17,43-45). CITED2 also functions as a transcriptional co-activator of the oestrogen receptor in breast cancer cells (19). Notably, CITED2 participates in sex determination and early gonad development through its combined action with WT1 and SF1, and regulates transcription activation of the genes located in the sex-determining region of the Y chromosome (46,47). By regulating the Nodal-Pitx2c pathway, CITED2 serves a role in controlling left-right gene transcription in the left lateral plate mesoderm (12,48-50). CITED2 is also reported to serve an essential role in the differentiation of the adrenal cortex from the adrenogonadal primordium, which stimulates WT1-mediated transcription activation, thereby increasing the promoter activity of the nuclear hormone receptor NR5A1 (47,51). CITED2 functions as a transcriptional co-repressor by interfering with the binding of HIF1-α or STAT2 with their transcription co-activator, p300/CBP (9,35,36,52-54). By displacing p300 from binding with ETS-1, CITED2 was shown to co-repress expression of MMPs, including MMP-1 and MMP-13 (55). Through downregulation of MMP expression, CITED2 was revealed to exhibit a chondroprotective role and is considered a potential drug target for treatment of osteoarthritis (56,57). A previous study demonstrated that by physically interacting with the DNA-binding transcription activator ISL1, CITED2 enhanced embryonic stem cell (ESC) cardiac differentiation (58). ISL1 has been shown to serve a role in several different types of cancer, including gastric cancer (59) and breast cancer (60,61). Therefore, whether CITED2 serves a role in cancer through its interaction with ISL1 will require further investigation in future studies.

3. Function of CITED2 in stem cells

CITED2 is essential for differentiation of hematopoietic stem cells (HSC) in the foetal liver and adult bone marrow, as Cited2-null mice exhibit impaired HSC function in the foetal liver and adult bone marrow (62-64). In the foetal liver, expression of self-renewal and survival-associated genes in HSCs (including Bmi-1, Wnt5a, LEF-1, Notch-1 and GATA2) were revealed to be significantly downregulated in Cited2-null HSCs (62). Studies have further revealed that PU.1 co-operates with CITED2 to maintain HSC (65,66). The role of CITED2 in hematopoietic stem cells has been previously reviewed (64). Thus, in the present review, a focus is placed on adult tissue stem cells, ESCs and induced pluripotent stem cells (iPSC).

Adult tissue stem cells: Tendon-derived stem/progenitor cells (TSPC) are the adult stem cells resident in the tendon tissue and are responsible for regeneration of tenocytes and healing of injuries to the tendons (67). A tendon’s healing capacity is gradually reduced with age, which may be due to decreased CITED2 expression in aged TSPCs, which results in defective self-renewal and altered differentiation fates (68). The proliferation rate is decreased, cell cycle progression is delayed and cell fate patterns are also altered in aged TSPCs (68). In particular, expression of tendon lineage marker genes is decreased, while adipocytic differentiation is increased in aged TSPCs (68). Another study suggested that CITED2 prevented TGF-β-induced TSPC senescence through downregulation of SP1 and P21, and upregulation of Myc (69). Whether increasing CITED2 expression can restore proliferation, cell cycle progression and determination of differentiation fate in aged TSPC, or even enhance tendon healing in vivo remains to be determined and should be the focus of future studies.

Although Cited2 is ubiquitously expressed (70) in other types of adult tissue stem cells, including adipose tissue-derived stem cells, intestinal stem cells, mammary stem cells and neural stem cells, whether CITED2 serves a role in the self-renewal and apoptosis of these cells remains to be determined.

ESCs: Interactions between transcription factors and transcription co-factors determine the fate of ESCs (71). Chromatin immunoprecipitation-seq analysis revealed that p300, a transcription co-factor, was mapped in the circuit of ESC stemness via its co-occupancy with stem cell marker transcription factor OCT4 on target genes (71). p300 is directly involved in regulating Nanog expression in mouse ESCs (72),
whereas CITED2 directly interacts with p300. By performing genome-wide screening of CITED2-overexpressing mouse ESCs, CITED2 was shown to be able to rescue the ESC phenotype following removal of leukaemia inhibitory factor (LIF) from the growth media (73). Loss-of-function of Cited2 in mouse ESC does not affect ESC proliferation and does not alter the undifferentiated state of ESCs in the presence of LIF (74). However, knockout of Cited2 delayed ESC differentiation due to delayed silencing of the genes involved in the maintenance of pluripotency and self-renewal of stem cells (including Oct4, Klf4, Sox2 and c-Myc) (74). A recent study revealed that Cited2-depleted stem cells retain higher expression levels of pluripotency-related transcription factors, including Nanog and Klf4, and that loss of Cited2 in differentiating ESCs delayed differentiation (75). However, Kranc et al (76) demonstrated that CITED2 is required for ESC proliferation, survival and self-renewal, by directly regulating the transcriptional expression of Nanog, Klf4 and Tbx3. Spontaneous differentiation of Cited2-knockdown ESCs occurred through downregulation of stem cell markers (including Nanog, Oct4, Sox2 and Tbx3), and upregulation of mesoderm gene markers (including Brachyury and Cdx2) and the ectoderm gene marker (Foxa2) (76). A possible explanation for the discrepancy between these previous studies is a result of the different methods of deleting the Cited2 gene used in the ESCs. Li et al (74) used a sequential targeting method; deleting the floxed Cited2 allele first by using Cag-cre, and then using a knockout targeting vector to delete the other allele. It is possible that the selected ESC clones were adapted to loss of Cited2 and thus survived. In fact, Kranc et al (76) observed a small portion of ESCs (~3%) that were able to self-renew without CITED2. Perhaps this small portion of self-renewing ESCs may be used to elucidate the mechanisms of how ESCs survive and self-renew.

### Table I. Transcription factors interacting with CITED2 or regulated by CITED2.

| Transcription factor(s) | Association with CITED2 | Function | (Refs.) |
|------------------------|-------------------------|----------|---------|
| TFAP2                  | Directly interacts with CITED2 | Normal neural tube and cardiac development, left-right patterning | (8,12,16,40,41) |
| SMAD2/3                | Directly interacts with CITED2 | Upregulates TGFβ downstream targets, such as MMP9, VEGF | (18,42) |
| PPAR-α, PPAR-γ         | Directly interacts with CITED2 | Together with CITED2, participates in signalling cascades of hypoxic response and angiogenesis | (17,43-45) |
| WT1                    | Directly interacts with CITED2 | Regulates SFI expression, sex determination and early gonad development | (46,47) |
| Ptx2c                  | CITED2 with TFAP2 regulates Ptx2c expression | Controls left-right gene transcription | (12,48-50) |
| HIF-1α                 | CITED2 inhibits HIF-1α activity | Displace p300/CBP from HIF-1α represses HIF-1α downstream targets expression | (14,35-39) |
| ETS1                   | CITED2 binds to ETS1 | Displaces p300/CBP from ETS1, represses MMP expression | (55) |
| ISL1                   | Directly interacts with CITED2 | Promotes stem cells cardiac differentiation | (58) |
| Myc                    | Directly interacts with CITED2 | Recruits p300 and Myc to E2F3 promoter and transactivates E2F3 expression and increases G1/S cell cycle progression | (21) |
| Nanog, Klf4, Tbx3      | CITED2 regulates Nanog, Klf4 and Tbx3 expression | ESC proliferation, survival and self-renewal | (76) |
| Nucleolin              | CITED2 regulates Nucleolin expression | By chaperone PRMT5 and p300 to Nucleolin promoter, CITED2 activates Nucleolin transcription, involved in prostate cancer metastasis | (22) |

CITED2, Cbp/P300 interacting transactivator with Glu/Asp-rich carboxy-terminal domain 2; TFAP2, transcription factor AP-2; SMAD2/3, mothers against decapentaplegic homolog 2/3; TGFβ, transforming growth factor β; MMP9, matrix metallopeptidase 9; VEGF, vascular endothelial growth factor; PPARα/γ, peroxisome proliferator-activated receptor α/γ; WT1, Wilms tumor 1 transcription factor; SFI, splicing factor 1; Ptx2c, paired like homeodomain 2C; HIF1α, hypoxia inducible factor 1 subunit α; ETS1, avian erythroblastosis virus E26 (V-Ets) oncogene homolog-1; ISL1, ISL LIM homeobox 1; Myc, MYC proto-oncogene, BHLH Transcription Factor; Nanog, homeobox transcription factor Nanog; Klf4, Kruppel-like factor 4; Tbx3, T-Box transcription factor 3; ESC, embryonic stem cell; PRMT5, protein arginine methyltransferase 5.
overexpression of these transcription factors in somatic cells give rise to pluripotent stem cells, or iPSCs (79,80). As described earlier, CITED2 regulates expression of stem cell marker transcription factors. Charneca et al (81), assessed whether overexpression of Cited2 alone was sufficient for generation of iPSCs. Notably, overexpression of Cited2 in mouse embryonic fibroblasts did activate expression of certain stem cell marker transcription factors, including Nanog, Sox2 and Rex1, but this was not sufficient for generation of iPSCs (81). Furthermore, overexpression of CITED2 in the pre-senescent fibroblasts significantly increased the efficiency of iPSC generation using the combination of the four transcription factors that Yamanaka established (79-81). It has been reported that aged cells are more difficult to transform into iPSCs compared with younger cells (82,83), and improving our understanding of the differences between aged and younger cells may highlight a possible solution for the generation of iPSCs from aged cells.

4. Function of CITED2 in cancer

Overexpression of Cited2 in Rat1 fibroblasts results in a loss of cell contact inhibition in in vitro cell cultures and in nude mice in vivo, highlighting the potential role of Cited2 in the transformation of certain types of cells (7). A recent study used system-level approaches to analyse the genome-wide transcriptionome of the protein-coding genes of 17 major types of cancer with respect to clinical outcome (84). A general pattern of shorter patient survival times was associated with upregulation of genes involved in cell growth (84). CITED2, a protein involved in cell growth and other fundamental cell processes, has been reported to serve a critical role in different types of cancer, which are further discussed below.

CITED2 in breast cancer. Breast cancer accounts for 30% of all cancer cases in females in the USA and is the leading cause of cancer-associated mortality worldwide (2). Cited2 expression was shown to be upregulated in primary breast cancer specimens and bone metastatic tumours compared with the normal mammary epithelium (25,85). Notably, cell lines with bone metastatic capacity in animal models exhibit the highest expression levels of Cited2 compared with less metastatic cell lines (25). A recent study also confirmed that human metastatic tumours express higher mRNA levels of Cited2 compared with primary tumours and normal epithelium (26). Additionally, expression levels of Cited2 are negatively associated with survival (19,25). Cited2 expression levels are associated with disease-free survival in patients with breast cancer, and has been proposed to serve as potential prognostic marker (85,86). However, van Aghthoven et al (87) reported that Cited2 mRNA expression levels were significantly increased in human breast cancer cell lines, and an analysis of data obtained from the Genomic Spatial Event database showed that Cited2 levels are lower in breast cancer tissue compared with normal tissues, albeit not statistically significant (22). Despite the discrepancy between studies, CITED2 is able to modulate oestrogen receptor transcriptional activity in breast cancer cells, and thus, elevated Cited2 expression may lead to oestrogen-independent oestrogen receptor activation, resulting in a reduction in oestrogen dependence and thus a reduced response to anti-oestrogen therapy (19). CITED2 may also modulate the capability of breast cancer metastases by positively regulating IKKα (26). Using breast cancer cell lines, it was demonstrated that knockdown of Cited2 expression resulted in reduced expression of the NF-κB signalling pathway regulator IKKα, and of the NF-κB signalling pathway downstream targets, OPN, MMP9, μPA, SPARC, IL-11 and IL-1β, which are known to serve roles in metastasis (26). Furthermore, knockdown of Cited2 expression in breast cancer cell lines attenuates breast tumour growth in mice (42), further highlighting the role of CITED2 in breast cancer progression. Therefore, it was proposed that CITED2 regulated vascularization of breast tumours through TGF-β dependent VEGF expression (42). Tumour-associated macrophages are important immune cells that serve a role in promoting primary tumour growth, metastatic progression, poor overall survival and therapeutic resistance (88-91). Notably, silencing Cited2 expression in breast cancer cell lines decreased the expression of the macrophage chemoattractant, CCL2, and thus attenuated macrophage recruitment (92). In fast growing tumours, cells usually encounter hypoxic stress, which results in induction of HIF expression, and apoptosis can be induced by HIF (93). Bakker et al (94) reported that in MCF7 breast cancer cells, HIF-1α induced FOXO3a expression and upregulated the downstream CITED2 expression. Increased expression of CITED2 inhibits HIF-1α-induced cell apoptosis (94). Although the mechanism of CITED2 function in breast cancer is not fully understood, the results of the aforementioned studies have demonstrated that CITED2 may be a potential therapeutic target for treatment of breast cancer.

Cited2 in leukaemia. CITED2 serves a critical role in the maintenance of hematopoietic stem cells (63,64,66,95), and loss of Cited2 in adult mice results in a loss of HSCs and bone marrow failure (63). Although leukaemia primarily rises from the hematopoietic progenitor level, different types of leukaemia have different causes (96). In acute myeloid leukaemia cells, CITED2 regulates P53 activity by regulating the AKT signalling pathway at multiple levels (97). When Cited2 expression was knocked down, expression of the AKT signalling pathway positive regulator transcription factor, SOX4, was decreased; however, expression of the negative regulator Thioredoxin-interacting protein was upregulated. Expression of the p53 target gene, PHLD3A, was downregulated, and expression of BCL2 gene was upregulated. As a consequence, loss of Cited2 expression in acute myeloid leukaemia cells increased P53-mediated apoptosis (97). Therefore, targeting Cited2 expression or CITED2 function may be a potential therapeutic method for treating patients with AML.

Cited2 in lung cancer. CITED2 is essential for foetal lung maturation in mice; Cited2-null lung exhibit reduced terminal sac space at embryonic day 18.5 (11). However, the function of CITED2 in lung cancer is not fully understood. Our unpublished preliminary data based on data mining from the Oncomine database (98) and data analysis using The Cancer Genome Atlas (22) all demonstrated that Cited2 expression levels in lung cancer are lower than in normal lung tissues from the same patients. The downregulation of Cited2 expression in
lung cancer may be explained by the lower expression of Foxa2 directly reducing Cited2 expression (98). However, another study reported that higher expression of CITED2 in lung cancer was associated with a less favourable prognosis (21). The proposed molecular mechanism in lung cancer is: CITED2 enhances E2F3 transcription factor expression by interacting with Myc, leading to increased G1/S cell cycle progression. On the other hand, CITED2 enhances Myc-mediated suppression of p21(CIP1), inhibiting cellular quiescence (21). Notably, knockdown of CITED2 expression can sensitize lung cancer cells to chemotherapy (Cisplatin), primarily through stabilization of p53 (99). Therefore, CITED2 may potentially serve as a therapeutic target in the treatment of lung cancer. A recent data mining study further found that CITED2 expression is significantly different between lung cancer specimen from smokers and non-smokers, suggesting that CITED2 may be used as a biomarker for smoking-related lung cancer (100).

Cited2 in prostate cancer. Prostate cancer is the most frequently diagnosed type of cancer in males, and the second leading cause of cancer-associated mortality among males (2). Despite the huge advances in prostate cancer therapy in recent years, the prognosis of patients with advanced prostate cancer remains generally poor due to metastasis (101), as the molecular and cellular mechanisms of prostate metastasis are not well understood. A recent study revealed that CITED2 is highly expressed in prostate cancer tissue from patients compared with normal tissues. Notably, metastatic prostate tumours express even higher CITED2 mRNA levels than non-metastatic prostate tumours (22). This study found that high CITED2 expression is significantly correlated with the overall survival of patients with prostate cancer. The authors further elucidated the mechanisms of this and proposed that CITED2 recruits Protein Arginine Methyltransferase 5 and p300 to nucleolin, which promotes epithelial-mesenchymal transition and prostate cancer metastasis (22). Therefore, CITED2 may be a potential therapeutic target for treatment of metastatic prostate cancer (22).

CITED2 in colon cancer. Colon cancer or colorectal cancer is the third most frequently diagnosed cancer type, in males and females (2). The function of CITED2 in colon cancer has not yet been extensively studied. One study using a colon cancer cell line reported that knockdown of Cited2 increased cancer cell invasiveness in vitro by upregulating MMP-13 expression (102). The histone deacetylase (HDAC) inhibitor, butyrate, resulted in upregulated expression of Cited2 in colon cancer cells and consequently downregulated MMP-13 expression. Ectopic expression of Cited2 induced colon cancer cell growth arrest (102). RNA-seq analysis showed that Cited2 expression was correlated with resistance to the chemotherapeutic reagent, irinotecan (103). The role of CITED2 in irinotecan resistance and the underlying mechanisms remain to be determined.

CITED2 in gastric cancer. Gastric cancer (colloquially referred to as stomach cancer) is a major category of cancer and is a leading cause of mortality in cancer-associated diseases (2). A few recent studies investigated the possible roles of CITED2 in gastric cancer. Data mining using existing gene expression data revealed that Cited2 was a signature prognostic gene among 16 genes (104). A Human Protein Atlas program used systems biology analysis and revealed that CITED2 is a prognostic predictor of stomach cancer (proteinatlas.org) (84). High expression of Cited2 is associated with a worse prognosis in patients with stomach cancer (Fig. 1; patient information in Table II; data derived proteinatlas.org) (84). Notably, low expression of Cited2 in gastric cancer cells is associated with chemoresistance (24). By overexpressing Cited2 or inducing Cited2 expression using an HDAC inhibitor in gastric cell lines, the cells were sensitized to the chemotherapeutic reagent, anthracycline, both in vitro and in vivo (24). There was also a small subset of patients with higher Cited2 expression levels in gastric cancer with a more complete response to chemotherapy, including epirubicin, although the number of patients was too small to draw any conclusions from (24). One possible explanation for this is that usually, high expression levels of Cited2 results in increased rates of cell proliferation and DNA synthesis, which in turn results in increased sensitivity to chemotherapy or radiotherapy. Mycophenolic acid (MPA), a metabolized product and active element of mycophenolate mofetil was revealed to inhibit gastric cancer cell invasion and migration (105). Based on microarray analysis, MPA may have downregulated the expression of a large number of pro-migratory genes and upregulated the expression of a number of anti-migratory genes, including Cited2 (105).
An in vitro study by Tang et al (106) reported that knockdown of Cited2 expression in gastric cells resulted in decreased cell proliferation, mitochondrial membrane potential and colony formation. Our understanding of the role of CITED2 in gastric cancer is limited and requires further study.

5. Conclusion and future perspectives

As CITED2 serves critical roles in numerous fundamental cell processes, it may be a suitable target for treatment of several types of cancer. Cited2-null normal tissue cells, including mouse embryonic fibroblast cells, HSCs, foetal liver cells, midbrain cells and neuroepithelium cells, exhibit premature senescence or an increase in the levels of apoptosis (8,10,23,107,108). Although resistance to apoptosis is a hallmark of cancer cells, induction of senescence or apoptosis, is a hypothesized means of controlling cancer growth. Notably, several studies have reported that CITED2 inhibits P53 activation in cancer cells (97,99,109,110), and upregulated expression of Cited2 in cancer cells inhibits P53 activation and apoptosis. Therefore, targeting CITED2 by silencing Cited2 gene expression and increasing cancer cell apoptosis (99,106) may be a possible treatment for cancer. Knocking down CITED2 expression in lung cancer cells resulted in a shrinkage of tumour size in nude mice and increased host mouse survival rates (21). Other methods, including inducing expression of microRNAs targeting and downregulating Cited2 expression, also results in apoptosis of cancer cells (111). Relapse and metastasis are the major hurdles of cancer therapy, and, whether CITED2 serves a role in cancer relapse or metastasis will be a topic of interest. As mentioned earlier, CITED2 directly or indirectly regulates expression of key stem cell transcription factors, OCT4, Nanog, Klf4 and Tbx3. These transcription factors are also known to be key players in cancer stem-like cells (112-115). Therefore, it is possible that CITED2 serves a key role in cancer stem cell function. Indirectly downregulating Cited2 expression in chronic myeloid leukaemia using PPAR-γ agonists resulted in an erosion of the leukaemia stem cell pool (116), which suggests that targeting CITED2 expression in cancer stem cell in general may be a therapeutic method for treatment of cancer, and preventing relapse. In conclusion, CITED2 is an essential transcription co-factor and may serve as a therapeutic target for the treatment of cancer.

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Availability of data and materials

All datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors’ contributions

YG conceived the presented idea and supervised the project. BA and XJ collected all the references and data. BA wrote the manuscript. All authors discussed, contributed toward and approved the final manuscript.

Ethics approval and consent to participate

This study does not contain any studies with human participants or animals performed by any of the authors.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Siegel RL, Miller KD and Jemal A: Cancer statistics, 2018. CA Cancer J Clin 68: 7-30, 2018.
2. Siegel RL, Miller KD and Jemal A: Cancer statistics, 2019. CA Cancer J Clin 69: 7-34, 2019.
3. Lampe JW: Dairy products and cancer. J Am Coll Nutr 30 (5 Suppl 1): 464S-470S, 2011.
4. Lambert M, Jambou S, Depauw S and David-Cordonnier MH: Targeting transcription factors for cancer treatment. Molecules 23: 1479, 2018.
5. Bushweller JH: Targeting transcription factors in cancer—from undruggable to reality. Nat Rev Cancer 19: 611-624, 2019.
6. Shioda T, Fener MH and Isselbacher KJ: MsiG1 and its related protein MRGI share a transcription activating domain. Gene 204: 235-241, 1997.
7. Sun HB, Zhu YX, Yin T, Sledge G and Yang YC: MRG1, the product of a melanocyte-specific gene related gene, is a cytokine-inducible transcription factor with transformation activity. Proc Natl Acad Sci USA 95: 13555-13560, 1998.
8. Bamforth SD, Braganca J, Eloranta JJ, Murdoch JN, Marques FIl, Kranc KR, Farza H, Henderson DJ, Hurst HC and Bhattacharya S: Cardiac malformations, adrenal agenesis, neural crest defects and exencephaly in mice lacking Cited2, a new Tfap2 co-activator. Nat Genet 29: 469-474, 2001.
9. Yin Z, Haynie J, Yang X, Han B, Kiatchoosakun S, Restivo J, Yuan S, Prabhakar NR, Herrup K, Conlon RA, et al: The essential role of Cited2, a negative regulator for HIF-1alpha, in heart development and neurodevelopment. Proc Natl Acad Sci USA 99: 10488-10493, 2002.
10. Xu B, Qu X, Gu S, Doughman YQ, Watanabe M, Xu B, et al: Cited2, a coactivator of HNF4alpha, is essential for liver development. EMBO J 26: 4445-4456, 2007.
11. Xu B, Qu X, Gu S, Doughman YQ, Watanabe M, Dunwoodie SL, and Yang YC: Cited2 is required for fetal lung maturation. Dev Biol 317: 95-105, 2008.
12. Bamforth SD, Braganca J, Farthing CR, Schneider JE, Broadbent C, Michell AC, Clarke K, Neubauer S, Norris D, Brown NA, et al: Cited2 controls left-right patterning and heart development through a Nodal-Pitx2c pathway. Nat Genet 36: 1189-1196, 2004.
13. Huang TQ, Wang Y, Ebrahim Q, Chen Y, Cheng C, Doughman YQ, Watanabe M, Dunwoodie SL and Yang YC: Cited2 is required for fetal lung maturation. Dev Biol 317: 95-105, 2008.
14. Bhattacharya S, Michels CL, Leung MK, Arany ZP, Kung AL and Livingston DM: Functional role of p35rsg, a novel p300/CBP binding protein, during transactivation by HIF-1. Genes Dev 13: 64-75, 1999.
15. Glenn DJ and Maurer RA: Mrg1 binds to the LIM domain of Lhx2 and may function as a coactivator to stimulate glycoprotein hormone alpha-subunit gene expression. J Biol Chem 274: 36139-36147, 1999.
16. Braganca J, Eloranta JJ, Bamforth SD, Ibbit JC, Hurst HC and Bhattacharya S: Physical and functional interactions among AP-2 transcription factors, p300/CREB-binding protein, and CITED2. J Biol Chem 278: 16021-16029, 2003.
Functional significance of SRJ domain

Berlow RB, Martinez-Yamout MA, Dyson HJ and Wright PE: The activation domain of HIF-1α interacts with the TAZ1 domain of CBP from molecular simulations. J Biol Chem 279: 3042-3049, 2004.

Matt T, Martinez-Yamout MA, Dyson HJ and Wright PE: The binding sites for CITED2 activate hypoxia-inducible transcriptional activity by perturbing left-right patterning of the body axis. Hum Mol Genet 19: 3394-3401, 2010.

Lau WM, Wei KL, Duan YT, Chen Y, and Sun YT: The transcription co-factor TGF-β induces Smad and CITED2 to the TAZ1 domain of CBP from molecular simulations. Stem Cells Dev 19: 107-113, 2010.

Chen CM, Benthem J, Cosgrove C, Braganca J, Cuenca A, Bamforth SD, Schneider JE, Watkins H, Keaveny B, Davies B and Bhattacharya S: Functional significance of SRJ domain interactions in CITED2. PLoS One 7, e46232, 2012.

MacDonald ST, Bamforth SD, Braganca J, Chen CM, Broadbent C, Schneider JE, Schwartz RJ and Bhattacharya S: A cell-autonomous role of CITED2 in controlling myocardial and coronary vascular development. Eur Heart J 34: 2557-2565, 2013.

Jayaaraman S, Drouet M and Kominsky SL: Down-regulation of CITED2 attenuates breast tumor growth, vessel formation and TGF-β-induced expression of VEGFA. Oncotarget 8: 6169-6178, 2017.

Gonzalez YR, Zhang Y, Belzadpoosh D, Cregan S, Bamforth S, Sanger RS and Park DS: CITED2 signals through peroxisome proliferator-activated receptor-gamma to regulate death of cortical neurons after DNA damage. J Neurosci 28: 5559-5569, 2008.

Kim GD, Das R, Rao X, Zhong J, Deulius JA, Ramirez-Bergeron DL, Rajagopalan S and Mahalebeshwar GH: CITED2 restrains pathological macrophage activation and response. Mol Cell Biol 38: e00452-17, 2018.

Liu Z, Wang Y, Dou C, Sun L, Li Q, Wang L, Xu Q, Yang W, Liu Q and Tu K: MicroRNA-1468 promotes tumor progression by activating PPAR-γ-mediated AKT signaling in human hepato-melanoma carcinoma. J Exp Clin Cancer Res 37: 49, 2018.

Buasa FW, Val P and Swain A: The transcription co-factor CITED2 functions during sex determination and early gonad development. Hum Mol Genet 18: 2989-3001, 2009.

Val P, Martinez-Barbera JP and Swain A: Adrenal development is initiated by CITED2 and required both for heart morphogenesis and establishment of the left-right axis in mouse development. Development 132: 1337-1348, 2005.

Bentham J, Michell AC, Lockstone H, Andrew D, Schneider JE, Brown NA and Bhattacharya S: Maternal high-fat diet interacts with embryonic Cited2 genotype to reduce Pitx2c expression and enhance penetrance of left-right patterning defects. Hum Mol Genet 19: 3394-3401, 2010.

Lopes Floro K, Arapt ST, Preis JJ, Fatkin D, Chapman G, Furtado MB, Harvey RP, Hamada H, Sparrow DB and Dunwoodie SL: Loss of Cited2 causes congenital heart disease by perturbing left-right patterning of the body axis. Hum Mol Genet 19: 3394-3401, 2010.

Combes AN, Spiller CM, Harley VR, Sinclair AH, Dunwoodie SL, Wilkinson D and Koppelmann P: Gonadal defects in Cited2-mutant mice indicate a role for SF1 in both testis and ovary differentiation. Int J Dev Biol 54: 683-689, 2010.

Berlow RB, Dyson HJ and Wright PE: Hypersensitive termination of the hypoxic response by a disordered protein switch. Nature 543: 447-451, 2017.

Shin DH, Li SH, Chun YS, Huang LE, Kim MS and Park JW: CITED2 mediates the paradoxical responses of HIF-1α to proteasome inhibition. Oncogene 27: 1593-1604, 2013.

Freedman SJ, Sun ZY, Kung AL, France DS, Wagner G and Eck MJ: Structural basis for negative regulation of hypoxia-inducible factor-1α by CITED2. Nat Struct Mol Biol 10: 504-512, 2003.

Combs AN, Spiller CM, Harley VR, Sinclair AH, Dunwoodie SL, Wilkinson D and Koppelmann P: Gonadal defects in Cited2-mutant mice indicate a role for SF1 in both testis and ovary differentiation. Int J Dev Biol 54: 683-689, 2010.
CITED2 and NCOR2 in the Ink4/Arf locus is a barrier for disorders. Dialogues Clin Neurosci 18: 267‑276, 2016.

Gurwitz D: Human iPSC‑derived neurons and lymphoblastoid cells for personalized medicine: What will it take for Africa? Trends Mol Med 15: 697‑704, 2019.

59. Guo T, Wen XZ, Li ZY, Han HB, Zhang CG, Bai YH, Xing XF, Cheng XJ, Du H, Hu Y, et al: ISL1 predicts poor outcomes for patients with gastric cancer and drives tumor progression through binding to the ZEB1 promoter together with SETD7. Cell Death Dis 10: 33, 2019.

60. Zhang Y, Wang L, Gao P, Sun Z, Li N, Lu Y, Shen J, Sun J, Yang Y, Dai H and Cai H: ISL1 promotes cancer progression and inhibits cisplatin sensitivity in triple‑negative breast cancer cells. Int J Mol Med 32: 2332‑2352, 2018.

61. Li X, Sun F, Chen G and Zhang M: ISL1 is upregulated in breast cancer and promotes cell proliferation, invasion, and angiogenesis. Onco Targets Ther 11: 781‑789, 2018.

62. Chen Y, Havriëk P, Bunting KD and Yang YC: Cited2 is required for normal hematopoiesis in the murine fetal liver. Blood 110: 2987‑2997, 2007.

63. Kranc KR, Schepers H, Rodrigues NP, Bamforth S, Villadés E, Ferry H, Bourrie‑Jones T, Sigvardsson M, Bhattacharya S, Jacobsen SE and Enver T: Cited2 is an essential regulator of adult hematopoietic stem cells. Cell Stem Cell 5: 659‑663, 2009.

64. Du J and Yang YC: Cited2 in hematopoietic stem cell function. Curr Opin Hematol 20: 301‑307, 2013.

65. Korthuis PM, Berger G, Bakker B, Rozenveld‑Geugien M, Jaques J, de Haan G, Schuringa JJ, Vellenga E and Schepers H: CITED2‑mediated human hematopoietic stem cell maintenance is critical for acute myeloid leukemia. Leukemia 29: 625‑635, 2015.

66. Mattes K, Geugien M, Korthuis PM, Brouwers‑Vos AZ, Fehrman RSN, Todorova TI, Steidl U, Vellenga E and Schepers H: Transcriptional regulators CITED2 and PU.1 cooperate in maintaining hematopoietic stem cells. Exp Hematol 73: 38‑49,e7, 2019.

67. Bi Y, Ehirchiou D, Kilts TM, Inkson CA, Embree MC, Sonoyama W, Li L, Bouriez‑Jones T, Sigvardsson M, Fehrmann RSN, Todorova TI, Steidl U, Vellenga E and Schepers H: Identification of tendon/stem/progenitor cells and the role of the extracellular matrix in their niche. Nat Med 13: 1219‑1227, 2007.

68. Zhou Z, Akinbiyi T, Xu L, Ramcharan M, Leong DJ, Ros SJ, Colvin AC, Schaffler MB, Majeska RJ, Flatow EL and Yang YC: Identification of human cancer‑specific transcription factors FOXA2‑centered transcriptional regulation network in breast cancer. Cancer Sci 101: 1824‑1832, 2009.

69. De Palma M and Lewis CE: Cancer: Macrophages limit chemotherapy. Nature 473: 303‑304, 2011.

70. Nielsen SR and Schmid MC: Macrophages as key drivers of cancer progression and metastasis. Mediators Inflamm 2017: 9624760, 2017.

71. Pollard JW: Tumour‑educated macrophages promote tumour progression and metastasis. Nat Rev Cancer 4: 71‑78, 2004.

72. Ruffell B and Coussens LM: Macrophages and therapeutic resistance in cancer. Cancer Cell 27: 462‑472, 2015.

73. Jayaraman S, Doucet M and Kominsky SL: CITED2 attenuates macrophage recruitment concordant with the downregulation of CCL2 in breast cancer cells. Oncol Lett 2019: 5292787, 2019.

74. Greider AE and van der Wal T: The role of hypoxia inducible factor 1 (HIF‑1) in hypoxia induced apoptosis. J Clin Pathol 57: 1009‑1014, 2004.

75. Bakker WJ, Harris IS and Mak TW: FOXO3a is activated in response to hypoxic stress and inhibits HIF‑1‑induced apoptosis via suppression of anti‑oestrogen resistance and progression of breast cancer. Br J Cancer 101: 1824‑1832, 2009.

76. Blasco MA and Serrano M: The two‑way street. Curr Opin Cell Biol 24: 744‑756, 2012.

77. Mahmoudi S and Brunet A: Aging and reprogramming: A one‑way street. Curr Opin Cell Biol 24: 744‑756, 2012.

78. Zhang Y, Wang L, Gao P, Sun Y, Qin L, Li Z, Liu J, Li J and Sun C: Competitive endogenous RNA network construction and application for cancer subtype discovery and screening. Bioinformatics 35: 4989‑4995, 2019.
103. Li XX, Zheng HT, Peng JJ, Huang LY, Shi DB, Liang L and Cai SJ: RNA-seq reveals determinants for irinotecan sensitivity/resistance in colorectal cancer cell lines. Int J Clin Exp Pathol 7: 2729-2736, 2014.

104. Zhao X, Cai H, Wang X and Ma L: Discovery of signature genes in gastric cancer associated with prognosis. Neoplasma 63: 239-245, 2016.

105. Dun B, Sharma A, Teng Y, Liu H, Purohit S, Xu H, Zeng L and She JX: Mycphenolic acid inhibits migration and invasion of gastric cancer cells via multiple molecular pathways. PLoS One 8: e81702, 2013.

106. Tang Z, He G, Xu J and Zhongfu L: Knockdown of Cbp/P300-interacting transactivator with Glu/Asp-rich carboxy-terminal domain 2 inhibits cell division and increases apoptosis in gastric cancer. J Surg Res 211: 1-7, 2017.

107. Barbera JP, Rodriguez TA, Greene ND, Weninger WJ, Simeone A, Copp AJ, Beddington RS and Dunwoodie SL: Folic acid prevents exencephaly in Cited2 deficient mice. Hum Mol Genet 11: 283-293, 2002.

108. Du J, Chen Y, Li Q, Han X, Cheng C, Wang Z, Danielpour D, Dunwoodie SL, Bunting KD and Yang YC: HIF-1α deletion partially rescues defects of hematopoietic stem cell quiescence caused by Cited2 deficiency. Blood 119: 2789-2798, 2012.

109. Liu YC, Chang PY and Chao CC: CITED2 silencing sensitizes cancer cells to cisplatin by inhibiting p53 trans-activation and chromatin relaxation on the ERCC1 DNA repair gene. Nucleic Acids Res 43: 10760-10781, 2015.

110. Yoshida T, Sekine T, Aisaki K, Mikami T, Kanno J and Okayasu I: CITED2 is activated in ulcerative colitis and induces p53-dependent apoptosis in response to butyric acid. J Gastroenterol 46: 339-349, 2011.

111. Jiang Y, Zhou Z, Fei R, Zhou X, Wang J, Tao Y, Li J and Chen T: Role of miR-182-5p overexpression in trichloroethylen-induced abnormal cell cycle functions in human HepG2 cells. J Toxicol Environ Health A 82: 920-927, 2019.

112. Rasti A, Mehrzama M, Madjd Z, Abolhasani M, Saeednejad Zanjani L and Asgari M: Co-expression of cancer stem cell markers OCT4 and NANOG predicts poor prognosis in renal cell carcinomas. Sci Rep 8: 11739, 2018.

113. Ponti D, Costa A, Zaffaroni N, Pratesi G, Petrangolini G, Coradini D, Pilotti S, Pierotti MA and Daidone MG: Isolation and in vitro propagation of tumorigenic breast cancer cells with stem/progenitor cell properties. Cancer Res 65: 5506-5511, 2005.

114. Jeter CR, Yang T, Wang J, Chao HP and Tang DG: Concise review: NANOG in cancer stem cells and tumor development: An update and outstanding questions. Stem Cells 33: 2381-2390, 2015.

115. Yu F, Li J, Chen H, Fu J, Ray S, Huang S, Zheng H and Ai W: Kruppel-like factor 4 (KLF4) is required for maintenance of breast cancer stem cells and for cell migration and invasion. Oncogene 30: 2161-2172, 2011.

116. Priest S, Relouzat F, Spetchian M, Ouzegdouh Y, Saliba J, Massonet G, Beressi JP, Verhoeyen E, Ragueneau V, Maneglier B, et al: Erosion of the chronic myeloid leukaemia stem cell pool by PPARγ agonists. Nature 525: 380-383, 2015.

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