Octamer-binding transcription factor 4 correlates with complex karyotype, FLT3-ITD mutation and poorer risk stratification, and predicts unfavourable prognosis in patients with acute myeloid leukaemia

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ABSTRACT
Objective: To investigate the correlation of octamer-binding transcription factor 4 (OCT4) expression with clinicopathological features and its predictive value for treatment response as well as survival profiles in de novo acute myeloid leukaemia (AML) patients.
Method: One hundred fifty-two de novo AML patients and 52 non-hematologic malignancy patients were recruited in this prospective cohort study. OCT4 expression was determined in bone marrow sample collected before treatment. Complete response (CR), event free survival (EFS) and overall survival (OS) were evaluated.
Results: Compared with the controls, OCT4 mRNA expression was higher in AML patients (P < .001), and higher OCT4 expression was correlated with presence of complex karyotype (CK) (P = .037), FLT3-ITD mutation (P = .012) and poorer risk stratification (P < .001) in AML patients. As to predictive value, OCT4 mRNA expression was decreased in patients achieved CR compared to non-CR patients (P = .022). Kaplan–Meier (K–M) curves showed that shorter EFS (9.0 (95% CI (7.7–10.3)) months vs. 25.0 (95% CI (17.5–32.5)) months, P < .001) and shorter OS (20.0 (95% CI (17.8–22.2)) months vs. 33.0 months, P < .001) were observed in OCT4 mRNA high expression patients compared to OCT4 mRNA low expression patients. Multivariate Cox’s proportional hazards regression analyses revealed that OCT4 mRNA high expression was an independent predictive factor for shorter EFS and OS in AML patients.
Conclusion: OCT4 correlates with presence of CK, FLT3-ITD mutation and poorer risk stratification, and it could be served as a convincing biomarker for predicting unfavourable prognosis in AML patients.

Introduction
Acute myeloid leukaemia (AML), one of the hematologic malignancies, originates from the proliferative, clonal, abnormally differentiated or occasionally poorly differentiated cells penetrating into the bone marrow (BM), blood and other tissues, and it is characterized by fatal infection, bleeding or organ infiltration [1,2]. With the advances in treatment for AML, 35–40% of adult AML patients with age less than 60 years and 5–15% AML patients with age more than 60 years are cured, while 20–70% patients relapse after achieving complete remission (CR) with initial induction chemotherapy, and treatment outcome in some patients remains dismal, such as the older patients who are unable to undergo intensive chemotherapy [1,3]. Emerging evidences have revealed that understanding of prognostic markers for AML would improve prevention of aggressive disease or treatment outcomes at early-stage disease [4–6].

Cancer stem cells (CSCs), a small subset of cancer cells, are characterized by stem cell features including self-renewal and differentiation capacity [7]. According to previous studies, CSCs involve in the processes of development, infiltration, metastasis and recurrence of tumours, and contribute to resistance to several chemo- or radio-therapies [8,9]. Octamer-binding transcription factor 4 (OCT4), which belongs to the family of POU transcription factor, is considered as a critical role in mediating tumour proliferation and differentiation. Plenty of studies have demonstrated that OCT4 is aberrantly expressed in various cancers, such as non-small cell lung cancer, bladder cancer, and cervical cancer [10–13]. In addition, abnormal OCT4 expression in above-mentioned cancers is correlated with aggressive tumourigenic activity and poor prognosis. Therefore, OCT4 has been identified as a predictive biomarker in some cancers, while little is known about the role of OCT4 in AML. Hence, this prospective cohort study was conducted to investigate the correlation of OCT4 expression with clinicopathological features and its predictive value for treatment response as well as survival profiles in de novo AML patients.
**Methods**

**Participants**

In this prospective cohort study, a total of 152 de novo AML patients from July 2014 to June 2017 were consecutively enrolled. Inclusion criteria included: (1) Newly diagnosed as AML according to World Health Organization (WHO) criteria (version 2008); (2) Aged 18 years or older; (3) Afford to complete the conventional therapy and able to be followed up regularly. Patients who (1) were diagnosed as acute promyelocytic leukaemia, (2) had previously received radiotherapy, chemotherapy or allogeneic hematopoietic stem cell transplantation (allo-HSCT) for other diseases, (3) had severe infection, severe heart dysfunction, severe lung dysfunction, severe hepatic dysfunction or renal dysfunction, (4) had history of any malignant tumour and (5) were in pregnancy or lactation, or planed for pregnancy were excluded from this study. In the same duration, 52 age and gender matched non-hematologic malignancy patients who received BM biopsy were consecutively enrolled as controls.

The present study had been approved by Institute’s Review Board of Xiangyang Central Hospital, The Affiliated Hospital of Hubei University of Arts and Science, and conducted according to the Helsinki declaration. All patients and controls provided written informed consents.

**Treatments**

All AML patients enrolled in this study underwent the Ara-C-based standard induction chemotherapy regimens (IA, DA, HA or MA) according to disease condition and patients’ willing. The induction chemotherapy regimens were as follows: (1) IA, Idarubicin 8–12 mg/m2/d (days 1–3), Ara-C 100–150 mg/m2/d (days 1–7); (2) DA, Daunorubicin 45–90 mg/m2/d (days 1–3), Ara-C 100–200 mg/m2/d (days 1–7); (3) HA, Homoharringtonine 2.0–2.5 mg/m2/d (days 1–7) (or 4 mg/m2/d (days 1–3)), Ara-C 100–200 mg/m2/d (days 1–7); (4) MA, Mitoxantrone 8–12 mg/m2/d (days 1–3), Ara-C 100–150 mg/m2/d (days 1–7). If the first course of induction treatment did not achieve complete remission (CR) for some AML patients, etoposide (100 mg/m2/d, d (days 1–5)), teniposide (100 mg/m2/d, day1, day 4) or an intermediate or intensive dose Ara-C was added to second course based on the original regimens and disease conditions. If CR was not achieved after two courses of treatment, it was considered to be treatment failure and other chemotherapy regimens should be reformulated. After remission, transplantation, micro transplantation or chemotherapy were selected according to real situations such as the risk stratification and the donor.

**Assessments and definitions**

For outcome analyses, we evaluated CR, which defined as BM with at least 20% cellularity and BM blasts below 5% at steady state after treatment, without cytological evidence of leukaemia, no transfusion requirement, leucocyte count above 1×10^9/L and platelet count above 100×10^9/L. Besides, event free survival (EFS) was defined as from the date of treatment to the time of recurrence, progression or death for any cause, and overall survival (OS) was calculated from the date of treatment to the time of death for any cause.

**Information collection and follow-up**

After enrollment in this study, demographic information of all patients and controls were collected, which included age and sex. For AML patients, FAB classification (according to French–American–British classification systems), cytogenetics (according to An International System for Human Cytogenetic Nomenclature (ISCN 2009)), the mutation status of internal tandem duplications in the FMS-like tyrosine kinase 3 (FLT3-ITD), isolated biallelic CEBPA mutation and nucleophosmin (NPM1) mutation, risk stratification (according to National Comprehensive Cancer Network (NCCN) Clinical Practice Guidelines in Oncology of AML (Version 2.2013)) and white blood cell (WBC) counts were recorded in detail. The median follow-up duration was 17.0 months (1/4–3/4 quartile: 12.0–24.0 months, range 2.0–36.0 months), and the last follow up month was June 2017.

**Samples and real-time quantitative Polymerase Chain Reaction (qPCR) assay**

Samples of the BM for OCT4 mRNA relative expression analysis were collected by BM biopsy before initiation of chemotherapy in AML patients and were also collected before treatment in non-hematologic malignancy controls. Total RNA was extracted by TRIzol Reagent (Invitrogen, U.S.A.) according to the manufacturer’s instructions. The RNA was quantified by Nano Drop 2000 (Thermo-Fisher, U.S.A.), and then 1 μg of total RNA from each sample was extracted and used for the synthesis of cDNA with reverse transcription kit (TaKaRa, Japan). Subsequently, cDNA product was subjected to qPCR with SYBR Green kit (TaKaRa, Japan). Amplification conditions of PCR were as follows: 95°C for 5 min, followed by 40 cycles of 95°C for 5 s, and then 61°C for 30 s. PCR results were calculated by 2^−ΔΔCt method, and glyceraldehyde phosphate dehydrogenase (GAPDH) was adopted as an internal reference. The primer sequences used for qPCR were as follows: OCT4, forward: 5’-GAGAAT TTTTCCCTGCGATGTC-3’; Reverse: 5’-ATCTCTCGTTGATCGCATAGTCG-3’; GAPDH,
forward: 5′-GAGTCCACTGGCGTCTTCC-3′; Reverse: 5′-ATCTTGAGGCTGGTGTCATACTTCT-3′.

Statistics

Statistical analysis was conducted by SPSS 21.0 software (IBM, U.S.A.) and Graphpad Prism 6 software (GraphPad Software Inc, U.S.A.). Data were mainly presented as mean value ± standard deviation, median value (1/4–3/4 quartiles), or count (percentage). Comparison between two groups was determined by Wilcoxon rank sum test; Comparison among three or above groups was detected by Kruskal–Wallis rank sum test. Kaplan–Meier (K–M) curves were used to present EFS and OS, and the log-rank test was applied to compare EFS and OS between/among groups. Univariate logistic regression and multivariate logistic regression were used to assess factors affecting CR, and Cox’s proportional hazards regression models were used to evaluate factors affecting EFS and OS. P < .05 was considered significant.

Results

Baseline characteristics

There were 152 AML patients (89 males and 63 females) in AML group, and 52 non-hematologic malignancy patients (27 males and 25 females) in control group. The mean ages of AML patients and control group were 37.05 ± 10.57 years and 38.44 ± 14.41 years, respectively. No difference in age (P = .459) or gender (P = .405) was observed between AML group and controls. As presented in Table 1, in AML patients, median value of WBC was 15.820 (7.266–26.827) × 10⁹ cell/L. According to FAB classification, 1 (0.7%), 56 (36.8%), 37 (24.3%), 45 (29.6%) and 13 (8.6%) patients were categorized as M1, M2, M4, M5 and M6 AML, respectively. As to risk stratification (NCCN 2013), 42 (27.6%), 60 (39.5%) and 50 (32.9%) AML patients were categorized as better-risk, intermediate-risk and poor-risk, respectively. Other detailed cytogenetic and molecular genetic information of AML patients were presented in Table 1.

Comparison of OCT4 mRNA relative expression in controls and AML patients

As shown in Figure 1, median value of OCT4 mRNA relative expression in AML patients (1.101 (0.692–1.927)) was remarkably higher compared with the controls (0.620 (0.300–1.262), P < .001).

Correlation of OCT4 mRNA relative expression with clinicopathological features

AML patients were divided into groups according to their clinicopathological features, and comparisons of OCT4 mRNA relative expression between/among different groups were performed (Table 2). Higher OCT4 mRNA expression was associated with poorer risk stratification (P < .001). Furthermore, OCT4 mRNA expression was positively correlated with complex karyotype (CK) (vs. no CK, P = .037) and FLT3-ITD mutation (vs. no FLT3-ITD mutation, P = .012). There was no correlation of OCT4 mRNA relative expression with age,

Table 1. Baseline characteristics of AML patients.

| AML patients (N = 152) |
|-----------------------|
| Age (years)           | 37.05 ± 10.57 |
| Gender (Male/Female)  | 89/63         |
| WBC (× 10⁹ cell/L)    | 15.820 (7.266–26.827) |
| FAB Classification    |               |
| M1 (n/%)              | 1 (0.7)       |
| M2 (n/%)              | 56 (36.8)     |
| M4 (n/%)              | 37 (24.3)     |
| M5 (n/%)              | 45 (29.6)     |
| M6 (n/%)              | 13 (8.6)      |
| Cytogenetics          |               |
| −5 or −7q− (n/%)      | 2 (1.3)       |
| t(9;22) (n/%)         | 2 (1.3)       |
| t(5;11) (n/%)         | 2 (1.3)       |
| inv(3) or t(3;3) (n/%)| 3 (2.0)       |
| 11q23 (n/%)           | 4 (2.6)       |
| +8 (n/%)              | 6 (3.9)       |
| −7 or −7q+ (n/%)      | 7 (4.6)       |
| t(8;21) (n/%)         | 7 (4.6)       |
| inv(16) or t(16;16) (n/%)| 13 (8.6)     |
| Others (n/%)          | 13 (8.6)      |
| CK (n/%)              | 15 (9.9)      |
| NK (n/%)              | 78 (51.3)     |
| Monosomol karyotype (n/%)| 12 (7.9)      |
| FLT3-ITD mutation (n/%)| 26 (17.1)     |
| Isolated biallelic CEBPA mutation (n/%)| 14 (9.2)    |
| NPM1 (n/%)            | 49 (32.2)     |
| Risk stratification (NCCN 2013) |               |
| Better-risk (n/%)     | 42 (27.6)     |
| Intermediate-risk (n/%)| 60 (39.5)     |
| Poor-risk (n/%)       | 50 (32.9)     |

Notes: Data were presented as mean value ± standard deviation, median value (1/4–3/4 quartiles), or count (percentage). AML: acute myeloid leukemia; FAB Classification: French-American-British classification systems; CK: complex karyotype; NK: normal karyotype; FLT3-ITD: internal tandem duplications in the FMS-like tyrosine kinase 3; CEBPA: CCAAT/enhancer-binding protein α; NPM1: nucleophosmin; NCCN: National Comprehensive Cancer Network; WBC: white blood cell.

Figure 1. OCT4 mRNA relative expression in AML patients and controls. OCT4 mRNA relative expression in AML patients was greatly higher compared with controls (P < .001). Wilcoxon rank sum test was conducted to compare the difference of OCT4 mRNA relative expression between the two groups. P < .05 was considered significant.
Notes: Data were presented as median value (1/4–3/4 quartiles) or median. Comparison was determined by Wilcoxon rank sum test or Kruskal-Wallis rank sum test. P < .05 was considered significant. Age was divided by median value (median = 37 years). AML: acute myeloid leukaemia; OCT4: Octamer-binding transcription factor 4; FAB Classification: French-American-British classification systems; CK: complex karyotype; NK: normal karyotype; FLT3-ITD: internal tandem duplications in the FMS-like tyrosine kinase 3; CEBPA: CCAAT/enhancer-binding protein α; NPM1, nucleophosmin; WBC, white blood cell.

### Table 2. Correlation of OCT4 mRNA relative expression with clinicopathological features.

| Clinicopathological features | OCT4 mRNA relative expression | P value |
|------------------------------|-------------------------------|---------|
| Age (years)                  |                              |         |
| >37                          | 1.130 (0.691–2.447)          | .417    |
| ≤37                          | 1.028 (0.694–1.688)          |         |
| Gender                       |                              |         |
| Male                         | 1.141 (0.699–2.109)          | .415    |
| Female                       | 1.000 (0.682–1.761)          | .269    |
| WBC                          |                              | .170    |
| >10 (× 10⁹ cell/L)           | 0.971 (0.691–1.559)          |         |
| ≤10 (× 10⁹ cell/L)           | 1.144 (0.703–2.201)          |         |
| FAB Classification           |                              |         |
| M1                           | 1.164                         |         |
| M2                           | 0.898 (0.560–1.668)          |         |
| M4                           | 1.093 (0.786–1.637)          |         |
| M5                           | 1.408 (0.918–3.048)          |         |
| M6                           | 1.078 (0.906–1.383)          |         |
| Cytogenetics                 |                              |         |
| −S or Sq−                    | 1.649                         |         |
| Yes                          | 1.097 (0.691–1.966)          |         |
| No                           | 2.056                         | .315    |
| t(9;22)                      | 1.101 (0.691–1.852)          |         |
| Yes                          | 1.420                         |         |
| No                           | 0.862                         | .587    |
| inv(3) or t(3;3)             | 1.101 (0.691–1.966)          |         |
| Yes                          | 1.311 (0.959–10.041)         | .303    |
| No                           | 1.096 (0.691–1.927)          |         |
| +8                           | 1.113 (0.969–4.103)          | .320    |
| Yes                          | 1.101 (0.691–1.852)          |         |
| No                           | 1.121 (0.996–1.670)          | .562    |
| −7 or 7q−                    | 1.094 (0.691–1.966)          |         |
| Yes                          | 0.896 (0.785–2.070)          | .604    |
| No                           | 1.109 (0.691–1.890)          |         |
| inv(16) or t(16;16)         | 0.896 (0.587–2.460)          | .280    |
| Yes                          | 1.116 (0.709–1.965)          |         |
| No                           | 1.184 (1.042–3.121)          | .037    |
| CK                           | 1.078 (0.676–1.788)          |         |
| Yes                          | 0.993 (0.646–1.978)          | .196    |
| No                           | 1.126 (0.876–1.852)          |         |
| Monosomal karyotype          | 1.190 (1.051–1.696)          | .223    |
| Yes                          | 1.086 (0.684–1.967)          |         |
| No                           | 1.579 (0.888–3.673)          | .012    |
| FLT3-ITD mutation            | 1.035 (0.677–1.594)          |         |
| Yes                          | 0.989 (0.661–2.478)          | .841    |
| No                           | 1.119 (0.695–1.966)          |         |
| NPM1                         | 0.978 (0.669–2.513)          | .780    |
| Isolated biallelic CEBPA mutation | 1.099 (0.691–1.705)             |         |
| Yes                          | 0.892 (0.588–1.242)          | <.001   |
| No                           | 1.035 (0.627–1.558)          |         |
| Risk stratification          | 1.470 (1.039–3.185)          |         |
| Better-risk                  |                              |         |
| Intermediate-risk            |                              |         |
| Poor-risk                    |                              |         |

**Correlation of OCT4 mRNA relative expression with CR and allo-HSCT achievement**

In patients achieved CR (N = 124), OCT4 mRNA relative expression was 1.035 (0.690–1.587), which was lower than that in non-CR patients (1.554 (0.904–3.096)) (Figure 2A, P = .022). Among patients achieved CR, there were 12 patients received allo-HSCT and the others (N = 112) did not (Figure 2B). No difference in OCT4 mRNA relative expression was found between allo-HSCT patients (1.076 (0.746–2.685)) and non-allo-HSCT patients (1.018 (0.684–1.564)) (P = .740).

**Analysis of factors affecting CR**

Univariate logistic regression analysis revealed that OCT4 mRNA high expression was numerically correlated with the absence of CR, but of no statistical significance (Table 3, P = .084). In addition, poorer risk stratification was associated with lower possibility of CR achievement (P = .001). In further multivariate logistic regression analysis, OCT4 mRNA high expression wasn’t an independent factor affecting CR, while risk stratification was an independent factor predicting worse CR in AML patients (P = .001).

**Comparison of survival profiles between patients with OCT4 mRNA high expression and patients with OCT4 mRNA low expression**

AML patients were classified as patients with OCT4 mRNA high expression (N = 76) and patients with OCT4 mRNA low expression (N = 76) according to the median value of OCT4 mRNA expression (median: 1.101). As disclosed in Figure 3(A), median EFS in OCT4 mRNA high expression patients was 9.0 (95% CI (7.7–10.3)) months, which was greatly shorter than that in OCT4 mRNA low expression patients (25.0 (95% CI (17.5–22.2)) months) patients, while risk stratification was an independent factor affecting EFS.

**Analysis of factors affecting EFS**

Factors affecting EFS were evaluated by univariate Cox’s regression, which displayed that OCT4 mRNA high expression was closely associated with shorter EFS (Table 4, P < .001). Besides, WBC > 10 (× 10⁹ cell/L) (P = .006) and higher risk stratification (P < .001) were
correlated with worse EFS, and there was a trend of poor OS in male (vs. female) patients ($P = .090$). Further analysis by multivariate Cox’s regression disclosed that OCT4 mRNA high expression ($P < .001$), WBC $> 10$ ($\times 10^9$ cell/L) ($P < .006$) and higher risk stratification ($P < .001$) were independent factors predicting reduced EFS in AML patients.

### Table 3. Factors affecting CR achievement by logistic regression model analysis

| Parameters            | Univariate logistic regression | Multivariate logistic regression |
|-----------------------|-------------------------------|---------------------------------|
|                       | $P$ value | OR | 95% CI   | $P$ value | OR | 95% CI   |
|-----------------------|-----------|----|-----------|-----------|----|-----------|
| OCT4 mRNA high expression | .084 | 0.473 | 0.202–1.106 | .211 | 0.625 | 0.253–1.532 |
| Age $>37$ years        | .797      | 0.896 | 0.388–2.071 | .306      | 0.632 | 0.262–1.522 |
| Male                  | .797      | 0.896 | 0.388–2.071 | .306      | 0.632 | 0.262–1.522 |
| WBC $>10$ ($\times 10^9$ cell/L) | .202 | 0.546 | 0.216–1.383 | .103 | 0.417 | 0.146–1.194 |
| Risk stratification   | .001      | 0.323 | 0.169–0.615 | .001      | 0.307 | 0.152–0.620 |

Notes: Data were presented as $P$ value, OR (odds ratio) and 95% CI (confidence interval). Factors affecting CR (complete remission) achievement were determined by univariate logistic regression and multivariate logistic regression analysis. $P$ value $< .05$ was considered significant. OCT4 mRNA high expression was divided by median value ($\text{median} = 1.101$). Age was divided by median value ($\text{median} = 37$ years). Risk stratification was scored as 0 – better risk, 1 – Intermediate risk, 2 – poor risk. OCT4: Octamer-binding transcription factor 4; WBC: white blood cell.

**Analysis of factors affecting OS**

Based on the analysis of univariate Cox’s regression, OCT4 mRNA high expression ($P < .001$) and higher risk stratification ($P < .001$) were correlated with decreased OS in AML patients, meanwhile, age $>37$ years ($P = .092$) and WBC $> 10$ ($\times 10^9$ cell/L) ($P = .083$) were numerically

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**Figure 2.** OCT4 mRNA relative expression in patients with different CR or allo-HSCT achievements. OCT4 mRNA relative expression in CR patients was lower than that in non-CR patients (A) ($P = .022$). No difference of OCT4 mRNA relative expression was found between allo-HSCT and non-allo-HSCT patients (B) ($P = .740$). Comparison between two groups was conducted by Wilcoxon rank sum test. $P < .05$ was considered significant.

**Figure 3.** Survival profiles in patients with OCT4 high expression and patients with OCT4 low expression. Median EFS in OCT4 mRNA high expression patients was reduced compared with OCT4 mRNA low expression patients (A) ($P < .001$). Median OS in OCT4 mRNA high expression group was shorter than that in OCT4 mRNA low expression group (B) ($P < .001$). K–M curves and log-rank test were used to compare OS between two groups. $P < .05$ was considered significant.
correlated with poor OS (Table 5). Furthermore, it was revealed by multivariate Cox’s regression that OCT4 mRNA high expression was an independent predictive factor for worse OS, as well as age > 37 years (P = .040) and higher risk stratification (P < .001).

**Risk stratification subgroup analysis of survival in patients with high/low OCT4 mRNA expression**

In better risk patients, high OCT4 mRNA expression was associated with lessened EFS (Figure 4(A), P < .001) and lessened OS (Figure 4(D), P < .001) compared with OCT4 mRNA low expression. As to patients with intermediate-risk, high OCT4 mRNA expression (vs. low OCT4 mRNA expression) was also correlated with poor EFS (Figure 4(B), P = .005) and poor OS (Figure 4(E), P = .003). Additionally, poor-risk patients with high OCT4 mRNA expression presented reduced EFS compared to poor-risk patients with low OCT4 mRNA expression (Figure 4(C), P = .016), while no difference in OS was found between poor-risk patients with high or low OCT4 mRNA expression (Figure 4(F), P = .343).

**Discussion**

In this study, we found that: (1) elevated OCT4 mRNA expression was presented in AML patients compared with controls; (2) elevated OCT4 mRNA expression was correlated with presence of CK and FLT3-ITD mutation as well as poor risk stratification; (3) OCT4 mRNA high expression was numerically associated with lower CR, and it independently predicted shorter EFS and OS in AML patients.

OCT4 belongs to the family of POU transcription factor, whose members bind to an octameric sequence motif involving the consensus sequence of AGTCAAAT, thereby regulating their target genes [14,15]. According to several previous studies, OCT4 possesses various functions as follows: (1) it serves as a suppressor for genes involving in differentiation, or a trans-activator for genes self-renew, through regulating multiple pathways such as canonical Wnt/β-catenin pathway (Wnt/β-catenin), transforming growth factor-β (TGF-β) and Janus kinase/signal transducer and activator of transcription (JAK/STAT) signal pathways [16–19]; (2) it contributes to tumourigenesis, and also plays an important role in the neoplastic behaviour, such as recurrence or resistance to therapy [20]. A large number of previous studies have confirmed the role of OCT4 in some cancers. Over-expression of OCT4 regulates cancer cell survival via a Survibin function in esophageal squamous cell carcinoma, and thus activates the antiapoptosis [14,21]. Up-regulated OCT4 expression is induced by modulation of hitone marks especially H3K4me3 and H3K9AcS10p in breast cancer cells, and finally promotes cancer cell proliferation and metastatic ability [22]. Therefore, these previous studies suggest that OCT4 serves as an oncogene in some carcinomas.

Based on the previous studies, over-expression of OCT4 has been observed in several cancers including cervical cancer, breast cancer and esophageal squamous
cell carcinoma [14,16,22–24]. However, little is known about the expression of OCT4 in AML patients, except for one previous study from Yin et al, which enrols 87 AML patients and 16 controls [25]. It displays that OCT4 expression in AML patients is dramatically higher compared to controls, also over-expression of OCT4 is correlated with elevated WBC, the presence of abnormal karyotypes and higher risk stratification in AML patients. Although this previous study has identified the negative role of OCT4 in AML patients, the sample size is relatively small, resulting in low statistics power. Thus, in this study, we enrolled 152 AML patients, which had a relatively larger sample size, and we found OCT4 expression in AML patients was greatly higher compared with controls and raised OCT4 expression correlated with the presence of abnormal karyotypes and higher risk stratification in AML patients. Although this previous study has identified the negative role of OCT4 in AML patients, the sample size is relatively small, resulting in low statistics power. Thus, in this study, we enrolled 152 AML patients, which had a relatively larger sample size, and we found OCT4 expression in AML patients was greatly higher compared with controls and raised OCT4 expression correlated with the presence of CK, FLT3-ITD mutation and poorer risk stratification. The possible reasons were that OCT4 over-expression could regulate several genes and pathways to induce abnormal karyotypes or molecular mutations, which led to worse risk stratification.

As to the predictive value of OCT4, the up-regulation of OCT4 has been regarded as a biomarker for poor prognosis in cancers such as non-small-cell lung cancer (NSCLC) and gastric cancer [26,27]. Li et al conduct a study and disclose that higher expression of OCT4 is associated with poor OS in NSCLC [26]. Another study reveals that elevated OCT4 level predicts shorter OS in human gastric cancer [27]. However, few studies exploring correlation of OCT4 expression with treatment outcomes in AML patients have been carried out. The aforementioned study performed by Yin et al displays that OCT4 high expression correlates with poor OS in AML patients who are less than 70 years, while no difference of OS was observed between total AML patients with OCT4 high expression (60.5 (range 15–81) years) and OCT4 low expression (49.5 (range 10–87) years) [25]. Although the previous study reveals the negative association of OCT4 expression and prognosis in AML, their treatment regimens are not clear (information is not shown), thus, these AML patients might be treated with different treatments, which might seriously affect their treatment outcomes. Whereas, in our study, we enrolled de novo AML patients receiving Ara-C based chemotherapy regimens, which decreased the effect on treatment outcomes resulting from diverse therapies, and we found OCT4 over-expression numerically correlated with the absence of CR and it served as an independent predictive factor for reduced EFS and OS in AML patients. The possible reasons for these results were as follows: (1) OCT4 interacts with several signalling pathways such as mitogen-activated protein kinase (MAPK) and JAK/STAT pathways to accelerate leukaemic cells proliferation and invasion, aggravating disease progress and increasing the recurrence, thereby resulting in worse EFS and OS in AML patients [28,29]; (2) OCT4 plays a crucial role in resistance to therapies by regulating several genes or pathways.
thus, in AML patients, it also activates multiple cell-survival pathways to promoting antiapoptosis of leukemic cells, thereby leading to poor treatment outcomes in AML patients [30].

Our study still had some limitations: (1) sample size of this study, which enrolled 152 de novo AML patients, was relatively small, thus, the statistical power was relatively low; (2) the median follow-up duration, which was 17.0 months (1/4–3/4 quartile: 12.0–24.0 months, range 2.0–36.0 months), was relatively short, therefore, the predictive value of OCT4 in long-term treatment outcomes was not investigated; (3) detailed mechanism of OCT4 in AML was not explored completely.

In conclusion, OCT4 correlates with the presence of CK, FLT3-ITD mutation and poorer risk stratification, and it could be served as a convincing biomarker for predicting unfavourable prognosis in AML patients.

Disclosure statement
No potential conflict of interest was reported by the author(s).

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