First case of AML with rare chromosome translocations: a case report of twins

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Abstract

Background: Leukemia is different from solid tumor by harboring genetic rearrangements that predict prognosis and guide treatment strategy. PML-RARA, RUNX1-RUNX1T1, and KMT2A-rearrangement are common genetic rearrangements that drive the development of acute myeloid leukemia (AML). By contrast, rare genetic rearrangements may also contribute to leukemogenesis but are less summarized.

Case presentation: Here we reported rare fusion genes ZNF717-ZNF37A, ZNF273-DGKA, and ZDHHC2-TTTY15 in a 47-year-old AML-M4 patient with FLT3 internal tandem duplication (ITD) discovered by whole genome sequencing (WGS) using the patient’s healthy sibling as a sequencing control.

Conclusion: This is, to our knowledge, the first case of AML with fusion gene ZNF717-ZNF37A, ZNF273-DGKA, and ZDHHC2-TTTY15.

Keywords: AML, Genetic rearrangement, FLT3/ITD, WGS

Background

Chromosome translocations are common genetic abnormality in leukemia. Multiple genetic fusions have been summarized and can be used to predict prognosis and for targeting. For example, in The 2016 revision of the World Health Organization (WHO) classification of myeloid neoplasms and acute leukemia, AML with recurrent genetic abnormalities were classified into AML with t(8;21)(q22;q22.1); RUNX1-RUNX1T1, AML with inv. (16)(p13.1q22) or t(16;16)(p13.1q22); CBFβ-MYH11, AML with PML-RARA, AML with t(9;11)(p21.3q23.3); MLL3-KMT2A, AML with t(6;9)(p23;q34.1); DEK-NUP214, AML with inv.(3)(q21.3q26.2) or t(3;3)(q21.3q26.2); GATA2, MECOM, AML (megakaryoblastic) with t(1;22)(p13.3;q13.3); RBBM15-MKL1 [1]. These AML subtypes account for most AML with recurrent genetic abnormalities. However, rare genetic translocations exist. For example, in spite of the fact that six partners of KMT2A gene account for majority KMT2A-rearranged leukemia, more than 135 partners of KMT2A have been identified so far [2]. Rare translocations may also contribute to leukemogenesis and be useful for personalized medicine of leukemia.

Thanks to the development of next generation sequencing, variations with low population frequency are able to be identified. The identification of susceptible or driver genes are also accelerated by twin or family study. Here we reported rare fusion genes ZNF717-ZNF37A, ZNF273-DGKA, and ZDHHC2-TTTY15 in AML with FLT3/ITD by a twin study.

Case presentation

Patient’s history, clinical and molecular features

A 47-year-old male (T3) presented to hospital at 21 June, 2015 reporting fever and being hypodynamic. Routine blood test showed high leukocyte count (115.27×10⁹/L), anemia (Hb 84 g/L), a total platelet count of 259×10⁹/L, and C-reactive protein (CRP) of 40.9 mg/L. Primitive and immature cells made up 90% of the peripheral blood cells. Acute leukemia was diagnosed and the patient was hospitalized. Bone marrow aspiration showed that primitive and immature cells made up 90% of the bone marrow cells and an AML-M4 was diagnosed. Immunophenotyping showed full expression of HLA-DR and CD33, partial expression of CD7, CD117, CD13, CD34, CD38, CD25, FMC-7, CD56, CD64, CD11C and MPO, no expression of CD5, CD10, CD19,
CD20, CD14, CD103, CD23, CD41a, GlyA, CD11b, CD15, CD138, kappa, lambda, CD79a, TdT, and cCD3. Chromosome karyotype of bone marrow showed 46, XY [3]. FLT3/ITD was identified. WT1/ABL ratio was 133.74%. Pirarubicin+cytarabine were administered but bone marrow depression occurred. Bone marrow aspiration showed active myeloproliferative activity and primitive and immature cells made up 54% of the bone marrow cells. The patient left hospital.

The patient presented to hospital second time at 24 July, 2015. Routine blood test showed leukocyte count of 18.70×10⁹/L, Hb of 69 g/L, platelet count of 30×10⁹/L. Bone marrow aspiration showed primitive and immature cells made up 79% of the bone marrow cells. IEA (Idarubicin +Etoposide+Aza-C) regimen was administered but bone marrow depression occurred. Bone marrow aspiration showed active myeloproliferative activity and primitive and immature cells made up 92% of the bone marrow cells. The patient left hospital.

The patient presented to hospital third time at 17 August, 2015. Routine blood test showed leukocyte count of 61.46×10⁹/L, Hb of 62 g/L, platelet count of 101×10⁹/L. Homoharringtonin+Etoposide were administered. The patient left the hospital at 19 October, 2015.

The patient presented to hospital fourth time at 1 November, 2015. Routine blood test showed leukocyte count of 145.72×10⁹/L, Hb of 71 g/L, platelet count of 15×10⁹/L. The patient deteriorated rapidly and passed away.

The patient had a twin brother (T4) who is healthy. Short tandem repeat (STR) genotyping based on 21 loci (D19S433, DSS818, D21S11, D18S51, D6S1043, AMEL, D3S1358, D13S317, D7S820, D16S539, CSFIPO, Penta D, D25S441, vWA, D8S1179, TPOX, Penta E, TH01, D12S391, D25I338, FGA) identified T3 and T4 were monozygotic twins.

Whole genome sequencing
Peripheral blood mononuclear cells (PBMCs) from the patient T3 (leukocyte count of 145.72×10⁹/L) and healthy sibling T4 were collected and genome DNA were isolated for WGS. Paired end 150 bp (PE150) sequencing on Illumina HiSeq X was performed at the Core Genomic Facility of Beijing Annoroad Genomics. All data were aligned to hg19 with BWA, arranged with samtools, marked with Picard, locally aligned with GATK. The coverage rate at 30× is 89.82%.

Single nucleotide polymorphism (SNP) was annotated using ANNOVAR. A total 3,033,876 SNPs were shared between T3 and T4. T4 had 317,907 unique SNPs while T3 had 301,334 unique SNPs.

Insertion-deletion (Indel) was annotated using ANNOVAR. A total 515,603 InDels were shared between T3 and T4. T4 had 93,904 unique SNPs while T3 had 92,676 unique SNPs.

Structural variation (SV) was annotated using ANNOVAR. A total 444 SVs were shared between T3 and T4. T4 had 152 unique SVs while T3 had 136 unique SVs. Only 23 (3.97%) SVs are exonic and only 4 relevant genetic loci (AKR1C4, CLCNKB/FAM131C, NUP43, PPIL4AD/E/F) are unique for T3.

Copy number variation (CNV) was annotated using ANNOVAR. A total 316 CNVs were shared between T3 and T4. T4 had 35 unique CNVs while T3 had 48 unique CNVs (21 are exonic).

There are 3 unique fusion pairs in T3 which are ZNF717 (exon6) fused to ZNF37A (exon 8), ZNF273 (exon 15) fused to DGKA (exon 10), and ZDHHC2 (exon 1) fused to TTTY15 (exon 13) as shown in Fig. 1 and specified in Additional file 1.

Discussion and conclusions
Here, we reported the first case of AML-M4 in a 47 years old man bearing ZNF717-ZNF37A, ZNF273-DGKA, and ZDHHC2-TTTY15 fusions detected by WGS analysis. ZNF717 has been reported to be involved in hepatocellular carcinoma [4], gastric cancer [5], cervical cancer [6]. From TCGA fusion gene database (www.tumorfusion.org), ZNF717-FOXP1 fusion was found in one colon adenocarcinoma. From another fusion gene database (Dong lab’s database, http://donglab.ecnu.edu.cn/databases/FusionCancer/) to be fused with LOCI00132288 and ITGB1, respectively, in lung cancer.

ZNF273 was reported by the database (http://donglab.ecnu.edu.cn/databases/FusionCancer/) to be fused with TPRKB in two cases, one in melanoma and another in prostate cancer.

DGKA, the gene encoding diacylglycerol kinase alpha (DGKα) which is a negative regulator of oncogene Ras [7], has attracted much interests from cancer researchers recently due to its involvement in multiple signaling pathways. DGKα inhibition compromises cancer cell viability, impairs angiogenesis, and notably boost T cell activation and enhance cancer immunotherapies [8]. DGKA was reported to form fusions with ASB8 in prostate cancer and RAB5B in uterine carcinosarcoma by TCGA database. DGKA was also reported to be fused with STARD4 in 4 Burkitt’s lymphoma cases and with CD74 in one lung cancer from Dong lab’s database.

DGKA may play a role in leukemogenesis. DGKα was absent in non-differentiated human promyelocytic leukemia cell line HL-60 cells, but was robustly upregulated during differentiation. By contrast, the other DGK isoforms (δ, ε, γ, ζ) existed in undifferentiated HL-60 cells but were remarkably decreased throughout differentiation [9]. DGKα was also reported to be abundant in the nuclei of human
erythroleukemia cell line K562, and to be involved in cell cycle progression of K562 cells [10]. The information implies that DGKα may be involved in the differentiation and cell cycle progression of leukemia cells.

The fusion between ZNF273 and DGKα may result in the production of a new protein with changed localization that may in turn influence how the kinase activity of DGKα exerts. The fusion between ZNF273 and DGKα led to the replacement of N-terminal domain of DGKα by the whole Zn finger domain of ZNF273 (Fig. 2). Deletion of the N-terminal domain of DGKα was reported to confer no effect on enzyme activity but result in constitutive localization of DGKα at the plasma membrane in intact T cells [11]. ZNF273-DGKα fusion may lead to dysregulated signaling pathway in leukemia cells.

ZDHHC2, a palmitoyl acyltransferase [12], has been reported by multiple groups to be involved in gastric adenocarcinoma [13], hepatocellular carcinoma [14]. ZDHHC2 was reported to be fused with LTBPI in two breast cancer cases, with PPP2R2A in one ovarian cancer, with FGD6 in one sarcoma case from TCGA database.

**Fig. 1** ZNF717-ZNF37A, ZNF273-DGKA, and ZDHHC2-TTTY15 fusions. The ZNF717 exon 6 were fused in-frame with ZNF37A exon 8. The ZNF273 exon 15 were fused in-frame with DGKA exon 10. The ZDHHC2 exon 15 were fused in-frame with TTTY15 exon 1

**Fig. 2** The predicted ZNF273-DGKα fusion. Wild-type ZNF273 contains 13 C2H2 Zn finger domains. Wild-type DGKα contains N-terminal domain, EF hands domain (calcium binding), C1 domain (Phorbol esters/diacylglycerol binding), catalytic domain, and C-terminal domain. The fusion between ZNF273 and DGKα results in the replacement of N-terminal domain of DGKα by C2H2 Zn finger domains of ZNF273
TTTY15 fusions have been reported in prostate cancer [3]. TTTY15-USP9Y fusion has been found in multiple cases of hepatocellular cancer, lung cancer, melanoma, prostate cancer, implicating a potential driver function of TTTY15-USP9Y fusion in carcinogenesis.

To sum up, ZNF717-ZNF37A, ZNF273-DGKA, and ZDHHC2-TTTY15 fusions may contribute to the development of AML.

Additional file

Additional file 1: Fusion Report. (XLS 2 kb)

Abbreviations

AML: Acute myeloid leukemia; CNVs: Copy number variations; InDel: Insert-deletion; ITD: Internal tandem duplication; SNPs: Single nucleotide polymorphisms; SVs: Structural variations; WGS: Whole genome sequencing

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Availability of data and materials

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

Authors’ contributions

LW and XX performed analysis of the WGS data. YHS and YLS collected samples from the patient, isolated DNA. LM performed STR genotyping. XX and LW performed analysis of the WGS data. YHS and YLS collected data. The study was designed and supervised by YLS, YY, and YY. LW drafted the paper. All authors read and approved the final manuscript.

Ethics approval and consent to participate

The study received ethics approval from the Commission for Scientific Research in Weifang Medical University and was administered in accordance with the ethical standards of the Declaration of Helsinki, second revision. Informed consent was obtained from the patient and his healthy sibling included in the study (ethical approval reference number: WYFY2015-029).

Consent for publication

Written informed consent was obtained from the patient and his healthy sibling for publication.

Competing interests

The authors declare that they have no competing interests.

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