CASE REPORT

Infantile Acute Megakaryoblastic Leukaemia with T(1:22) in a Non-Down Syndrome Child

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ABSTRAK

Megakaryoblastik leukemia adalah sejenis myeloid leukemia akut yang sering dihidapi bayi penghidap sindrom Down. Megakaryoblastik leukemia dengan translokasi t(1;22)(p13;q13) adalah sejenis myeloid leukemia akut yang jarang ditemui dan meliputi hanya <1% bagi semua kes. Ia dilaporkan berlaku hanya kepada bayi penghidap megakaryoblastik leukemia yang tidak mengalami sindrom Down. Ia lebih melibatkan bayi perempuan dan translokasi sitogenetik ini dilaporkan mempunyai impak yang buruk kepada prognosis pesakit. Kami melaporkan kes yang jarang ditemui ini berlaku kepada seorang bayi perempuan berumur 9 bulan yang menunjukkan simptom tanda-tanda lebam, pembengkakan hati dan limpa yang serius dan pembengkakan kelenjar limfa servikal dan inguinal. Darah periferi menunjukkan anemia yang serius, ovalositosis, trombositopenia dan sel darah putih yang tinggi. Sum-sum tulang menunjukkan kepadatan sel-sel blast yang mempunyai ratio nukleus-sitoplasmic yang tinggi dan sitoplasm tanpa granul dan blebs. Ujian peroksidase adalah negatif. Ujian imunofenotip menunjukkan ekspresi penanda CD117, CD 13, CD33 dan CD61 yang mengesahkan diagnosa megakaryoblastik leukemia akut. Menariknya, ujian sitogenetik menunjukkan kehadiran translokasi t(1;22) yang dilaporkan berlaku kepada bayi yang tidak mengalami sindrom Down berlaku pada kes ini. Pesakit ini menerima rawatan rejim kemoterapi mengikut protokol AML trial 15 ADE dan sebagai kesannya telah mengalami neutropenik sepsis yang serius dan masalah pernafasan yang memerlukan bantuan pernafasan dan faktor stimulasi koloni granulosit (G-CSF). Walaubagaimanapun, pesakit tidak menghadiri sesi temujanji seterusnya setelah sembuh dan beberapa bulan kemudian kembali dengan penyakit myeloid lukemia akut berulang.

Kata kunci: Leukemia infantil,translokasi (1;22), non-Down Syndrome

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ABSTRACT

Megakaryoblastic leukaemia is the commonest form of leukaemia occuring in Down syndrome infants. However, it's subtype with translocation t(1;22)(p13;q13) is uncommon comprising <1% of all cases and reported to exclusively occur in infant without Down syndrome. It has a female predominance and carries a poor prognosis. We described this rare form of leukaemia in a 9-month-old girl who presented with bruises, massive hepatosplenomegaly and multiple cervical and inguinal lymphadenopathy. The blood film showed severe anaemia with ovalostomatocytosis, thrombocytopenia and mild leucocytosis. The bone marrow aspirate showed numerous blasts showing high nuclear-cytoplasmic ratio and agranular cytoplasm with cytoplasmic blebs. Peroxidase staining was negative. The immunophenotyping of the blasts showed positive expression of CD117, CD13, CD33 and CD61 which confirmed the diagnosis of acute megakaryoblastic leukaemia. Interestingly, the cytogenetic finding of translocation t(1;22) which is most common in acute megaloblastic leukaemia in infants without Down syndrome was found in this case. She received the AML trial 15 ADE protocol chemotherapy regime and developed severe neutropenic sepsis and respiratory distress requiring ventilatory support and granulocyte colony stimulating factor (G-CSF). She recovered well after the first course of chemotherapy and was discharged. Unfortunately, she was not brought in for follow-up chemotherapy and presented a few months later with relapsed AML. She was re-started on ADE protocol and currently is on oral thioguanine for maintenance therapy.

Keywords: Infantile leukaemia , translocation (1;22), non-Down Syndrome

INTRODUCTION

AML constitutes 15-20% of all acute leukaemia cases in children 15 years and below. According to the National Cancer Registry of Cancer Incidence in Malaysia 2007, the incidence of AML is 2.8 per 100,000 population. The age specific cancer incidence per 100,000 population for 0-9 years of age in Malaysia is 3-6 for males and 2-4 for females.

Acute megakaryoblastic leukemia (AML M7) is an uncommon subtype of AML, occuring in approximately 10% of paediatric and 5% of adult AML (Bernstein et al. 2000, Carroll et al. 1991). It was reported that its incidence is highest among infants with Down syndrome and is associated with mediastinal germ cell tumors (Swerdlow et al. 2008).

The t(1;22) is an uncommon abnormality in AML, constituting less than 1% of all AML cases. It is found most commonly among non-Down syndrome infants, predominantly in females. AML with t(1;22) is a de novo AML, restricted to infants and young children, with most cases occuring in the first six months of life. It is associated with t(1;22)/RBM15-MKL1 resulting in expression of the one twenty-two megakaryocytic acute leukaemia (OTT-

MAL) fusion gene (Carroll et al. 1991; Ma et al. 2001; Mercher et al. 2009). The expression of OTT-MAL fusion gene results in abnormal megakaryocyte development. The megakaryopoiesis changes include enhancement in the cells self-renewal ability. Apart from that there is activation of recombination signal binding protein for imunoglobulin J kappa region (RBPJ)-mediated transcription (Mercher et al. 2009). It is suggested that prenatal genetic factors are involved in leukaemogenesis in these AML M7 infants having t(1;22) (Carroll et al. 1991). These infants usually present with huge hepatosplenomegaly, anaemia, thrombocytopenia and moderate elevation of the white blood cell count. At presentation, they commonly come with cytopenias, particularly thrombocytopenia, however some may have normal or increased in platelet count.

The megakaryoblasts show similar morphological features in all AML-M7 with or without t(1;22). The blasts are heterogenous with some undifferentiated blast cells resembling lymphoblasts. These megakaryoblasts are medium to large in size with slight irregular or indented nucleus with fine reticular chromatin and prominent nucleoli. They have basophilic cytoplasm, often agranular and some showed distinct blebs or pseudopod formation (Swerdlow et al. 2008). The bone marrow biopsy may show hypercellular marrow with evidence of fibrosis (Carroll et al. 1991). These marrow fibrosis may be attributable to the secretion of fibrogenic cytokines by the megakaryoblasts (Carroll et al. 1991).

Immunophenotypically, the megakaryoblasts express one or more platelet glycoprotein CD41 (glycoprotein IIb/IIIa), and/or CD61 (glycoprotein IIIa) and other myeloid markers CD13 and CD33. The more mature platelet-associated marker CD42 (glycoprotein Ib) is less frequently expressed. CD34, CD45, MPO and HLA-DR are usually negative (Swerdlow et al. 2008).

Therapy of AML generally consists of aggressive multi-drug chemotherapy regimens consisting of cytarabine, anthracyclines (daunorubicin, idarubicin, and mitoxantrone) and etoposide (Bain 2007) aimed to eliminate the leukaemic cells and achieve a complete haematological remission (CR). Our centre used the AML 15 protocol similar to a study trial conducted in Birmingham.

AML M7 has a better prognosis among Down's syndrome infants, with a four year event free survival of 73%. The prognosis in both children and adults with AML M7 with t(1;22) is poor, primarily attributable to a resistant disease (Bain, 2007). Approximately 50% of patients will obtain an initial remission with cytarabine and anthracycline but with only 10.5 months median survival (Carroll et al. 1991). Bone marrow transplantation is indicated in all cases of AML M7 with t(1;22) (Lion et al. 1992). However, long term survivors following intensive therapies have been reported (Reinhardt et al. 2005).

CASE REPORT

A 9-month-old baby girl presented to our centre with pallor, lethargy,

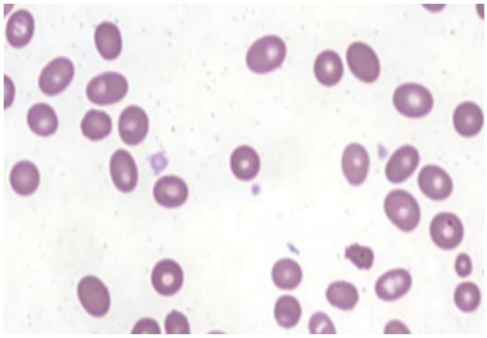

Figure 1: Full blood picture showing severe anaemia with ovalostomatocytes. (Wright's stain x40)

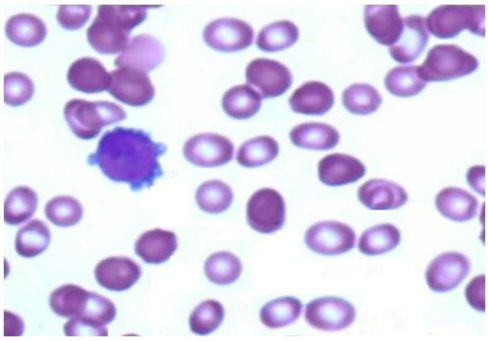

Figure 2: Presence of blast cell in the full blood picture. The blast show cytoplasmic blebs. (Wright's stain x40)

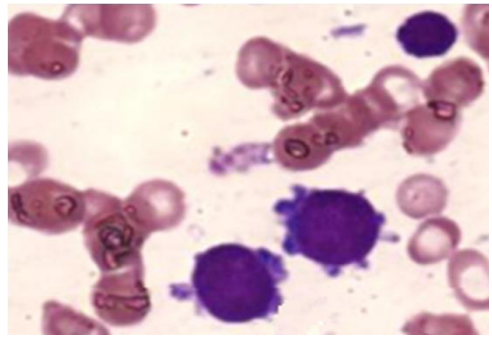

Figure 3: Blasts with cytoplasmic blebs. (MGG stain x40)

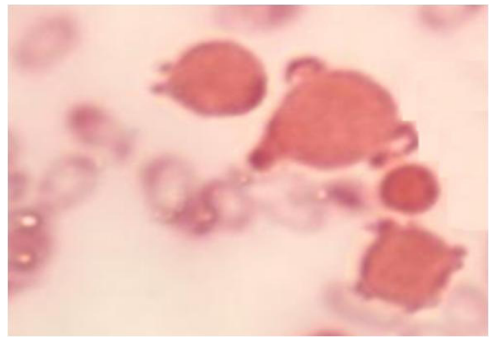

Figure 4: The megakaryoblasts are negative to Myeloperoxidase stain. (Peroxidase stain, x40)

abdominal distension, loose stool, vomiting and reduced oral intake of two weeks duration. On physical examination, she was noted to be underweight with failure to thrive, pale and tachypnoeic. There were huge hepatosplenomegaly and multiple lymphadenopathies involving the cervical, axillary, inguinal, and occipital region. Areas of ecchymoses were noted on her back. A soft systolic murmur was present with displaced apex beat at the 6th intercostal space, 1 cm lateral to the mid-clavicular line.

Full blood count showed severe anaemia with haemoglobin of 2.6 g/dL, leukocyte count of 19.9 x 10^9/L, and platelet count of 23 x 10^9/L. The full blood picture revealed severe anaemia, ovalostomatocytosis, thrombocytopenia with leukoerythroblastic picture and presence of blasts (Figure 1, 2). The bone marrow aspirate showed presence of 60% blasts which are heterogenous in size. The blasts have high nucleo-cytoplasmic ratio, immature nuclei, inconspicuous nucleoli, and abundant basophilic cytoplasm. Some of the blasts have cytoplasmic blebs. No Auer rods or granules are noted within the cytoplasm (Figure 3). The peroxidase stain was negative (Figure 4).

Immunophenotyping of the bone marrow aspirate performed

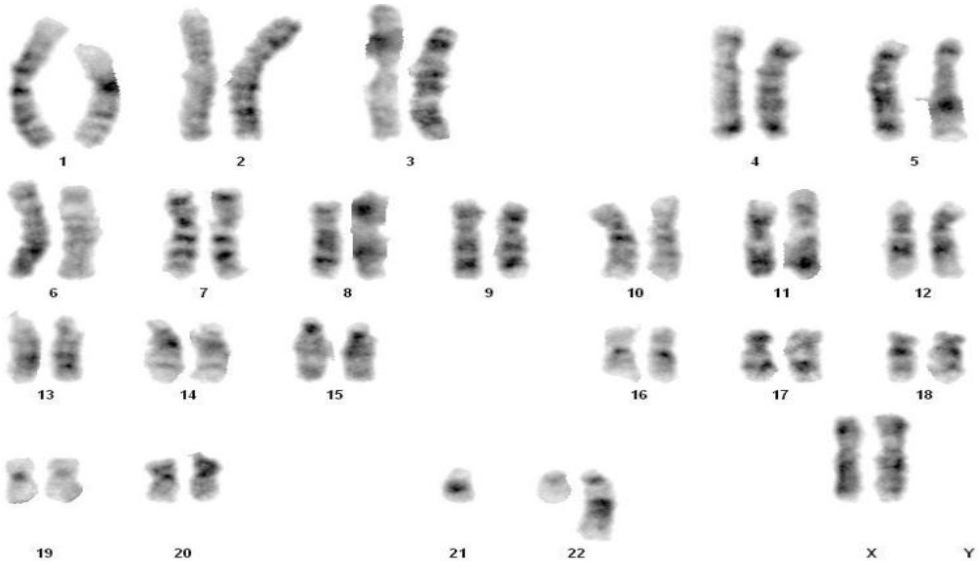

Figure 5: High resolution karyotyping result showing the t(1;22)(p13;q13).

demonstrated an abnormal population of blasts expressing CD117, CD13, CD33(dim) and CD61 and negative for other B and T cell markers consistent with acute megakaryoblastic leukaemia. The trephine biopsy was inconclusive as the sample was unsatisfactory. Interestingly, the cytogenetic study showed translocation between chromosome 1 and chromosome 22 (t(1;22)(p13;q13)) in sixteen metaphase spreads (Figure 5). Flourescent in situ hybridisation (FISH) analysis showed no visible BCR/ABL, TEL/AML1, or MLL gene rearrangement. The serum lactate dehydrogenase (LDH) was markedly elevated (7916 U/L).

She received multiple transfusion of packed cells, platelets and was started on the ADE chemotherapy protocol which consist of cytosine arabinoside (Ara-C), daunorubicin and etoposide. She developed haematuria following platelet transfusion which after ultrasound of the kidney revealed mild to moderate hydronephrosis with hydroureter.

During the course of chemotherapy, she developed neutropenic sepsis with bronchopneumonia complicated with acute respiratory distress and needed intensive care monitoring, continuous positive airway pressure (CPAP) and inotrope support. She was ventilated for 12 days and was given broad spectrum antibiotics Meropenem and Amphocil, as well as granulocyte colony-stimulating factor (G-CSF) to increase the neutrophil counts. Her condition improved and was allowed for discharge after one week recovery. The parents were informed to bring her for follow up within a week for further management. Unfortunately, her parents defaulted the follow-up. A few months later the child was brought in with relapsed acute myeloid leukaemia. She was re-started on the ADE protocol

and completed the cycles. Currently she is well and is on maintenance chemotherapy with oral thioguanine and sub-cutaneous Ara-C.

DISCUSSION

Acute megakaryoblastic leukaemia (AML M7) is an uncommon subtype of AML, comprising approximately 10% of paediatric and 5% of adult AML (Bernstein et al. 2000; Carroll et al. 1991). It has a bimodal age distribution that peaks in early childhood, normally in children less than 3 years of age and adulthood (Gewirtz et al. 1989; Ribeiro et al. 1993). In a large series of childhood AML from a single institution, 14.6% of all cases of AML were identified as AML M7 (Athale et al. 2001). The incidence of AML M7 is much higher than what was previously reported. This could be attributed to the complexity of its clinical presentation and difficulty in obtaining and interpretating the bone marrow samples for diagnostic testing (Ravindranath et al. 1992). Thus, many of these AML M7 cases may have been diagnosed as undifferentiated leukaemia, myelodysplastic syndrome, myelofibrosis or other disease (Athale et al. 2001). Currently, the improvement in knowledge on the morphology, cytochemistry, and immunophenotyping of the megakaryoblasts, together with the advancement in cytogenetic testing have allowed a more accurate and reliable diagnosis of AML M7 as well as determination of the character and prognosis of this disease.

In children, the incidence of AML M7 is highest among infants with Down syndrome with trisomy 21 and also in children with mediastinal germ cell tumors (Swerdlow et al. 2008).

In contrast, AML M7 in infants without Down syndrome is found to be exclusively associated with t(1;22)/ RBM15-MKL1 resulting in fusion of the OTT(RBM15) gene on 1p13 to the MAL(MLK1) gene on chromosome 22, leading to the OTT-MAL fusion gene on the derivative 22 (Swerdlow et al. 2008; Carroll et al. 1991; Ma et al. 2001; Mercher et al. 2009). It is an uncommon subtype in AML, constituting less than 1% of all AML cases and found most commonly among non-Down syndrome infants, predominantly in females. Other cytogenetic abnormalities that may be present in these non-Down syndrome infants with acute megakaryoblastic leukaemia includes inv(3)(q21;q26), which is not specific to AML-M7 and may be seen in other types of AML (Edmond et al. 2001). The insertion(12p) can also be found among patients with AML-M7 but it is more commonly associated with mediastinal germ cell tumours (Edmond et al. 2001).

AML with t(1;22) is a de novo AML, restricted to infants and young children, with most cases occuring in the first six months of life. It is suggested that prenatal genetic factors are involved in leukaemogenesis in these AML M7 infants with t(1;22) (Carroll et al. 1991). These infants usually present with huge hepatosplenomegaly, anaemia, thrombocytopenia and moderate leucocytosis. Cytopenias, particularly thrombocytopenia, are common at presentation but the platelet count can be normal or even increased in some cases. Characteristically, there is no

preceeding myelodysplasia, or history of transient leukaemoid reaction in these infants (Verschuur et al. 2004). These features were well illustrated as reported in our case.

The morphological features of the blasts presented in our patient were similar to that described for AML M7 with or without t(1;22). In addition, the stromal pattern of bone marrow infiltration by these megakaryoblasts may mimic a metastatic tumour. The blasts are usually myeloperoxidase negative, but are positive for acid phosphatase and alpha-naphthyl acetate esterase (sodium fluoride resistant).

Immunophenotypically, the megakaryoblasts express platelet glycoproteins, namely CD41 (glycoprotein IIb/IIIa), and/or CD61 (glycoprotein IIIa), together with the myeloid markers of CD13 and CD33. CD34, CD45, MPO and HLA-DR are usually negative (Swerdlow et. al 2008). The immunophenotyping from our patient showed expression of CD117, CD13, CD61 and CD33(dim) which are consistent with the diagnosis of AML M7.

Based on the general principle of leukaemia therapy, there are multiple chemotherapy protocols available from different centres conducting these trial studies. Among them is the AML 15 trial study conducted in Birmingham in which our patient's treatments were based upon. The ADE 10+3+5 protocol from the AML 15 trial were used. Each course of chemotherapy results in a temporary bone marrow suppression, leading to prolonged anaemia, leukopenia, neutopenia and thrombocytopenia. The prolonged myelosuppression often render the patients susceptible to opportunistic bacterial, fungal or viral infection, which can be life-threatening. Moreover, the chemotherapy courses can also cause varying degree of mucositis, which is caused by the cytotoxic effect of the chemotherapy agents on the epithelium of the intestinal tract, thus requiring various supportive measures.

The prognosis in both children and adults with this form of leukemia is poor, primarily attributing to its resistant disease (Bain' 2007). Approximately 50% of patients will obtain an initial remission with cytarabine and anthracycline but the median survival is only 10.5 months (Carroll et al. 1991). AML M7 has a better prognosis among Down's syndrome infants, with a 4 year event free survival of 73% and bone marrow transplantation is indicated in cases of AML M7 with t(1;22) (Lion et al. 1992). However, long term survivors following intensive therapies have been reported (Reinhardt et al. 2005).

In conclusion, AML M7 is a heterogenous disease affecting the two extreme ages and carries a poor prognosis, contributed by the resistant nature of the disease. AML M7 commonly affect infants with Down's syndrome or associated with t(1;22) in those with non-Down syndrome. Early recognition and diagnosis of AML M7 especially to identify the subtype with t(1;22) is very crucial to stratify these patients to the more intensive management in order to achieve good response to treatment and improving their overall survival. We highlighted a case of an infant with a rare subtype of AML M7 with t(1;22) who had

successfully undergone an intensive series of chemotherapy and currently well on maintenance treatment.

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