

## CASE REPORT

### Complex karyotypic abnormalities in a case of acute myeloid leukaemia – M4Eo

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#### Abstract

A 25-year-old man was referred to Hospital UKM with a 2-week history of fever, productive cough and loss of appetite. Physical examination revealed an ill-looking, tachypnoeic young man. No obvious lymphadenopathy or organomegaly was noted. Examination of the respiratory system revealed right pleural effusion. Full blood picture demonstrated leukocytosis with 90% blasts, and bone marrow examination confirmed the diagnosis of acute myeloid leukemia (AML) French-American-British (FAB) classification of M4 with eosinophilia. His chromosome karyotyping showed complex karyotypic abnormalities. Cytological examination of the pleural fluid demonstrated numerous blast cells indicating leukemic infiltration of the lungs, which is a rare presentation in AML. He was then started on induction chemotherapy with intravenous daunorubicin and cytarabine. In the midst of treatment, he developed an episode of seizure and cerebro-spinal fluid cytology confirmed central nervous system (CNS) leukaemic infiltration. Additional intrathecal methotrexate was given. Repeat bone marrow examination done on day 15 of chemotherapy showed persistence of excess blasts indicating refractory AML. He was then reinduced with high dose cytarabine but to no avail. The disease progressed and he succumbed about 8 weeks after the initial diagnosis was made.

We highlight here a case of AML-M4Eo with complex karyotypic abnormalities presenting with leukaemic infiltration of the lungs and CNS which had imparted a bad prognosis for this subtype of AML, AML-M4Eo.

**Keywords:** AML-M4Eo, complex karyotypic abnormalities, lung infiltration, CNS infiltration.

#### INTRODUCTION

Acute myeloid leukemia FAB classification M4 with eosinophilia (AML-M4Eo) is a subtype of M4 characterized by increased in bone marrow eosinophils.<sup>1</sup> It accounts for about 30% of M4 AMLs and appears to be distributed equally between children and adults. The eosinophils typically have large eosinophilic granules, some showing nuclear hyposegmentation or hypersegmentation, and occasional immature forms. No precise percentage of eosinophils and precursors was required for the diagnosis of M4Eo.<sup>2</sup> Most often, M4 with cytologically atypical eosinophils of > 5% is usually categorized as M4Eo.

Chromosomal 16 abnormalities particularly inv(16) was documented to occur in the majority of AML of M4Eo subtype. AML-M4Eo with the

associated inv(16) is well known to be sensitive to chemotherapy and carries a favourable prognosis.<sup>3,4</sup> Contrary to the inv(16), we report here a case of AML-M4Eo with a complex karyotype which was resistant to intensive chemotherapy and had a fatal outcome.

#### CASE REPORT

A 25-year-old Chinese man was referred from a private hospital with a history of fever for 2 weeks, associated with productive cough and loss of appetite. He was previously well with no significant past medical or surgical history. On examination, he was conscious and alert, with a temperature of 38.4°C. Blood pressure was 160/100mmHg and pulse rate 92/min. He was pale but not jaundiced. There was no obvious lymphadenopathy or organomegaly. Respiratory

system examination revealed right basal coarse crepitations. Examination of other systems was unremarkable.

Investigations revealed low haemoglobin (107g/L), leukocytosis ( $62.1 \times 10^9/L$ ) and thrombocytopenia ( $63 \times 10^9/L$ ) with the presence of 90% blasts in the peripheral blood film (Fig 1). Bone marrow aspiration (Fig 2a) and trephine biopsy (Fig 2b) showed that the bone marrow had been replaced by blast cells. These blast cells were composed of a mixture of myeloblasts and monoblasts, with prominence of eosinophilic precursors and abnormal eosinophils. Findings were consistent with acute myelomonocytic leukemia with eosinophilia, FAB subtype AML-M4Eo. The blast cells expressed CD34, CD33, HLA-DR, CD11c and intracellular myeloperoxidase. Cytogenetic analysis of the bone marrow sample showed two clones of abnormal cells. 11 of 13 cells analyzed showed 45 chromosomes with monosomy chromosome X, chromosome 19, derivative of chromosomes 11 and 17 with the presence of a marker chromosome (Fig 3a). The other 2 of 13 cells showed a different clone with 44 chromosomes with monosomy chromosome X, chromosome 16 and chromosome 19, derivative of chromosomes 11 and 17 with the presence of a marker chromosome (Fig 3b). Liver profile

showed mild hypoproteinemia and hypoalbuminemia (total protein 56g/l, albumin 36g/l). Lactate dehydrogenase was elevated (2184mmol/l). Renal profile, random blood sugar and uric acid were normal. Chest radiography showed pneumonic changes at the lower right basal region. Blood, urine and sputum cultures were all negative.

In view of his high spiking temperature and pneumonia findings, chemotherapy was deferred while he was started on intravenous Netilmycin and Piperacillin. Despite 3 days on these antibiotics, his symptoms worsened. His temperature continued to spike above 39°C and he became more dyspnoeic. Physical examination demonstrated signs indicative of right pleural effusion. Repeat chest radiography confirmed the diagnosis of pleural effusion. Intravenous antibiotics were changed to Meropenam. A pleural tap was performed and 50cc of blood-stained pleural fluid was aspirated. Pleural fluid analysis showed occasional pus cells and was negative for acid-fast bacilli. Cytology examination revealed presence of numerous blast cells (Fig 4). About one week into the antibiotic treatment, he remained febrile with a slight improvement of the respiratory symptoms after the pleural tap. Intravenous Amikacin and Fluconazole (antifungal) were

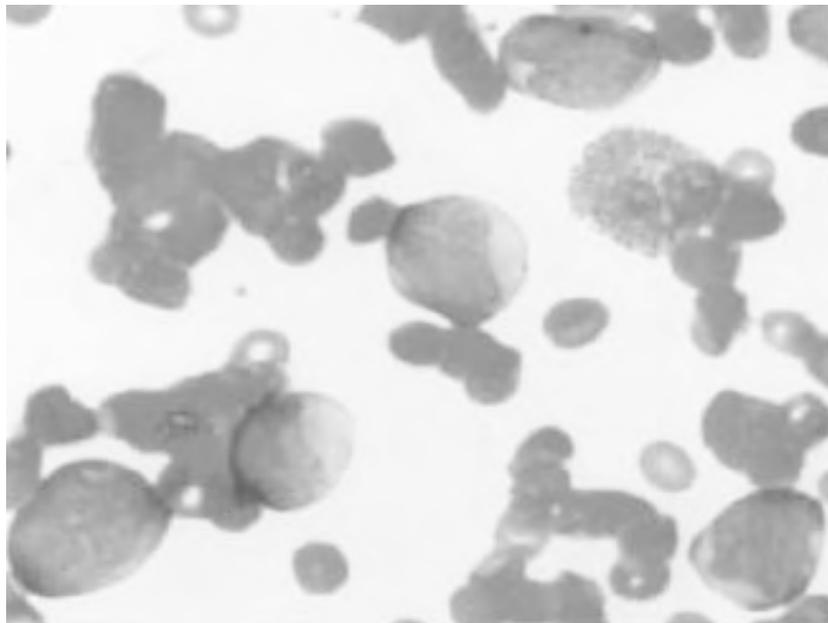


Fig. 1: Peripheral blood film showing numerous blast cells which are large, with prominent nucleoli and presence of eosinophils. (Wright's stain, X 600)

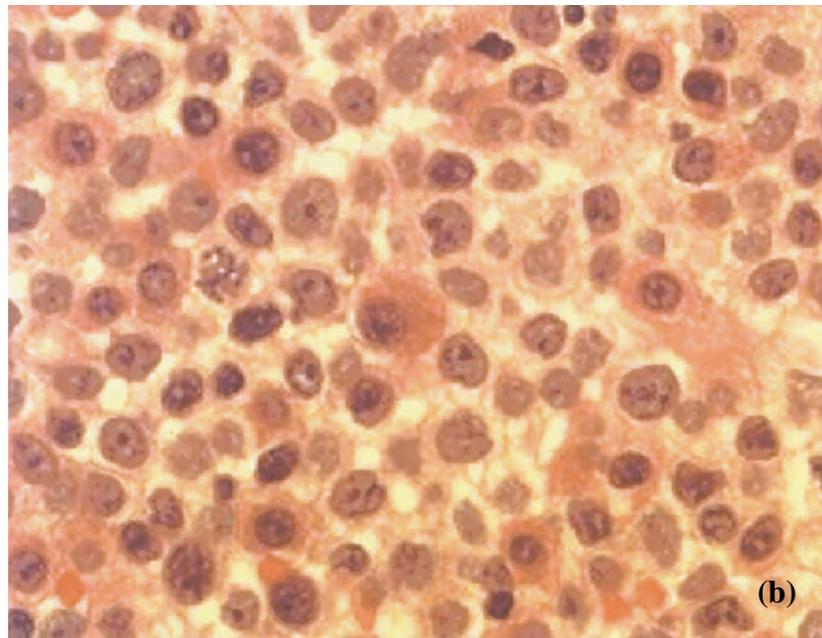
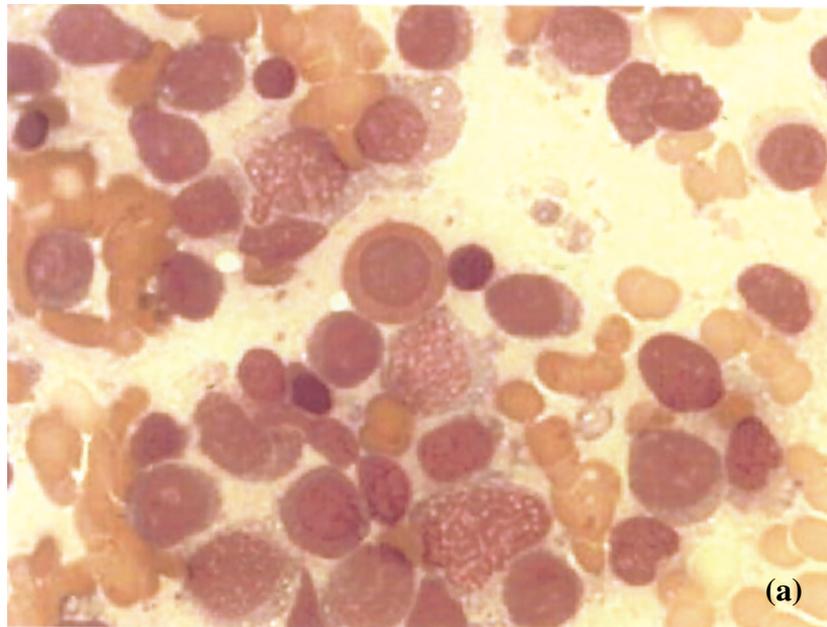


Fig. 2: (a) Bone marrow aspirate showing numerous blast cells with abnormal mononuclear eosinophils consistent with the diagnosis of AML-M4 with eosinophilia. (MGG stain, X 400). (b) Trephine biopsy showing numerous blast cells with abnormal eosinophils. (H & E stain, X 400)

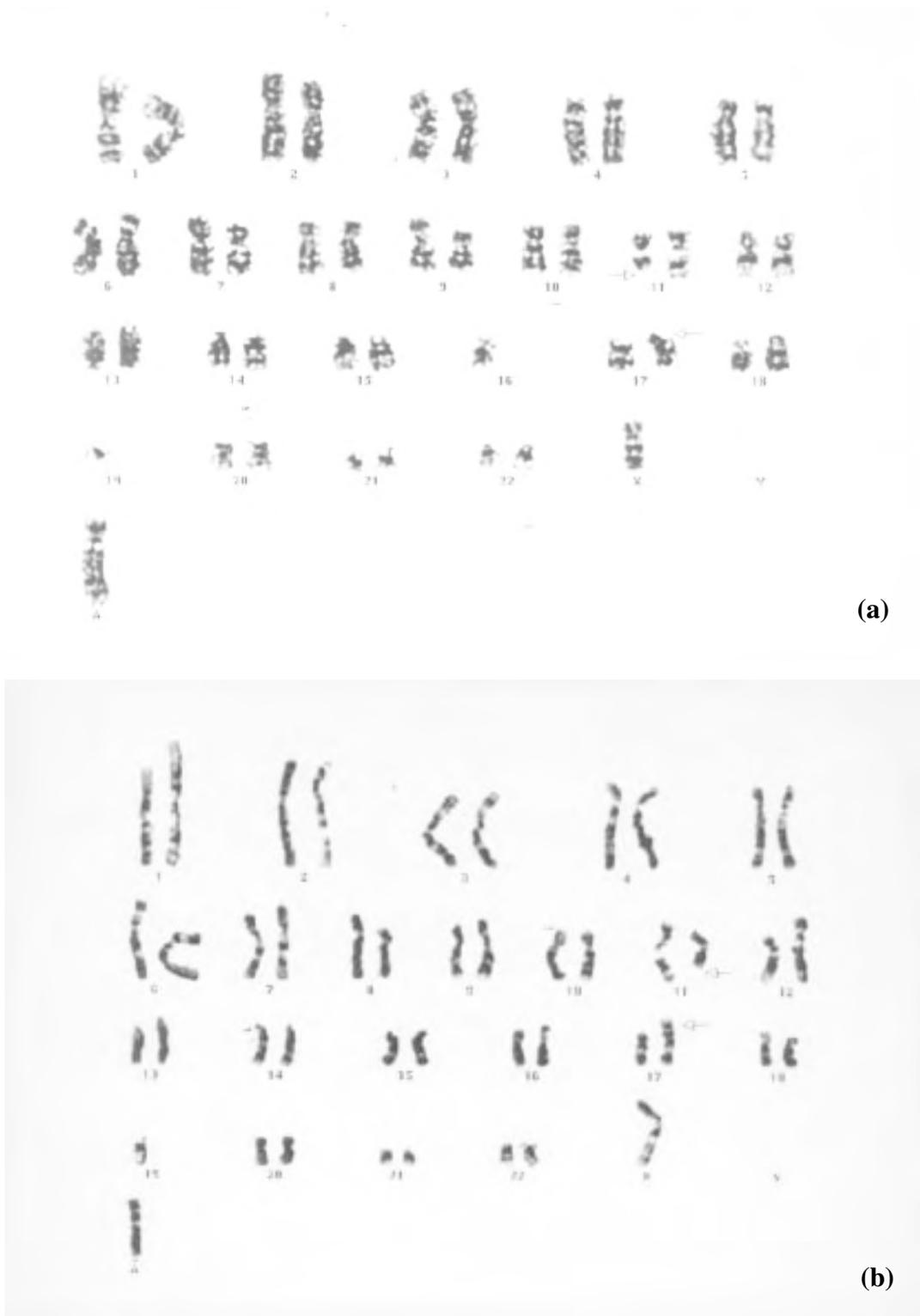


Fig. 3: Chromosomal analysis showing 2 clones of abnormal cells. (a) 44 chromosomes with monosomy chromosome 16 and chromosome 19, derivative of chromosomes 11 and 17 (arrow) with presence of a marker chromosome A. (b) 45 chromosomes with monosomy chromosome 19, derivative of chromosomes 11 and 17 (arrow) with presence of a marker chromosome A.

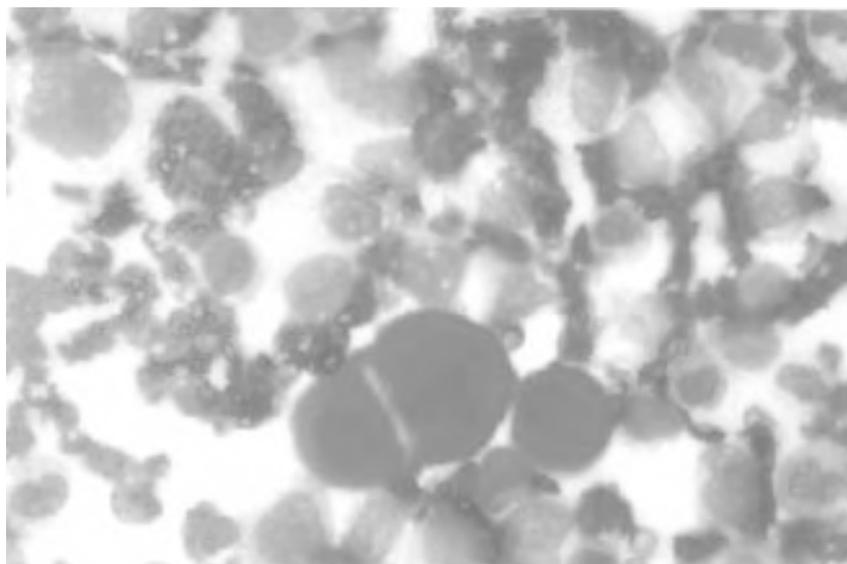


Fig. 4: Pleural fluid cytology showing numerous blast cells. (PAP stain, X 400)

added. However, despite being on multiple antibiotics and antifungal treatment, his fever persisted above 39°C and he remained dyspnoeic. On the 13<sup>th</sup> day of admission, he was noted to have developed macular-papular rashes starting from the lower back, gradually spreading to the upper thighs and lower abdomen. The dermatologist thought that these could be leukemic infiltrations of the skin. However, the patient refused skin biopsy. Meanwhile, his white cell count had decreased dramatically from  $62.1 \times 10^9/L$  (with eosinophilia) on admission to  $8.1 \times 10^9/L$  (with normal eosinophil count) without any chemotherapy.

Following the commencement of the chemotherapy (Daunorubicin and Cytarabine), his fever subsided within 2 days. His respiratory symptoms improved dramatically and the skin rashes resolved. At 15 days post-chemotherapy, a repeat bone marrow examination revealed persistence of excess blast cells. He then developed an episode of seizure in the ward. Lumbar puncture was performed and cerebrospinal fluid (CSF) cytology showed numerous blast cells. He was then treated with intrathecal methotrexate and high dose intravenous Cytarabine (HIDAC). Full blood picture on day 14 after commencement of HIDAC showed severe neutropenia with presence of 30% blast cells. On day 21 post HIDAC, the white cell count had increased to  $35.0 \times 10^9/L$  with > 90% blast cells indicating resistance of blast cells to the intensive chemotherapy. The patient succumbed to the disease 8 weeks after the initial diagnosis.

## DISCUSSION

The different categories of AML have many clinical features in common but some differences. Primary manifestations include mucosal pallor secondary to anemia, gingival bleeding and petechiae secondary to thrombocytopenia and fever in most cases of leukemia. Some degree of hepatosplenomegaly and lymphadenopathy is also common particularly in those categories with a prominent monocytic component. However, infiltration of the skin, gums, tonsils and pleura or pericardial effusion is rare and if it occurs, it is usually found in AML with a monocytic component. AML-M4Eo was documented to have a higher incidence of central nervous system involvement especially at relapse.<sup>5</sup> But Campell *et al*<sup>4</sup> in 1991 documented that CNS relapse was only observed in 8% of his patients indicating no increased incidence of this complication in AML-M4Eo. In our patient, we have documented leukemia infiltration in the lungs and CNS.

Multiple parameters have been documented to contribute to the treatment outcome as well as prognosis for acute myeloid leukemia. Patient associated parameters such as sex and age, leukemia-associated parameters such as leukocyte count, FAB subtype, and lactate dehydrogenase are some of the parameters. Recently, cytogenetics is unequivocally considered to be of major importance in delineation of prognostically distinct categories of AML. The majority of the newly diagnosed leukaemias show chromosomal abnormalities.

Up to 70% of patients with acute myeloid leukemia (AML) have abnormal karyotypes.<sup>6</sup> Inv(16)(p13q22) is one of the commonest chromosomal abnormalities found in acute myeloid leukemia, representing approximately 16% of documented karyotypic abnormalities.<sup>7</sup>

Chromosome 16 abnormalities are nearly pathognomonic of AML-M4Eo with inv(16) being the commonest. AML-M4Eo with inv(16) is known to be sensitive to chemotherapy and carries a favorable prognosis.<sup>3,4</sup> It was documented by Shurtleff *et al*<sup>7</sup> in 1995 and Betts *et al*<sup>8</sup> in 1992 that 61% and 86% respectively of AML cases with inv(16) were AML-M4Eo, whereas about 20% of AML-M4 will have inv(16).<sup>9</sup> However, some other chromosome 16 abnormalities such as t(16,16)(p13q22), del(16)(q22) and complex variant forms have also been reported in the literature to be associated with AML-M4Eo, though not common. Some of these cases may not have the typical AML-M4Eo morphology.

Complex karyotypic abnormalities are defined as three or more numerical and/or structural chromosome aberrations.<sup>10</sup> Previously, additional cytogenetic abnormalities present at diagnosis or variants of chromosome 16 abnormalities were not believed to alter the prognosis of the disease.<sup>6</sup> However, Braess *et al*<sup>11</sup> in 2001 have shown that the leukemic blasts with complex aberrant karyotypes have a low proliferative activity which by itself is associated with a poor response to induction therapy. Wheatley *et al*<sup>12</sup> in 1999 reported that the response status after one course of chemotherapy and cytogenetic abnormalities were the two most important independent prognostic factors. Complex karyotype was one of the adverse karyotypes with a survival rate of 17% and a relapse rate of 75% as compared to the favourable karyotype (survival rate 73% and relapse rate 34%) and those patients with adverse karyotypes were more likely to have resistant disease. Similar findings were reported by Claudia Schoch *et al*<sup>10</sup> in 2001. They shown that 10% of patients with *de novo* AML with complex aberrant karyotypes had poor outcome even when treated with intensive therapy at both younger and older ages.

Our patient presented with multiorgan involvement (pulmonary, central nervous system) and complex karyotypic abnormalities, without the characteristic inv(16) abnormality. He did not respond to chemotherapy and had a fatal outcome. It appears that complex karyotypic

abnormalities do impart a bad prognosis for AML-M4Eo.

## REFERENCES

1. Bain BJ. Bone Marrow Pathology: Acute Myeloid Leukaemia. 2<sup>nd</sup> Edition. Blackwell Science Ltd, 2000, pp 94-97.
2. Bain BJ. Eosinophilic leukaemias and the idiopathic hypereosinophilic syndrome. Br J Haematol 1995; 1: 2-9.
3. Larson RA, Williams SF, LeBeau MM, Bitter MA, Vardiman JW, Rowley JD. Acute myelomonocytic leukemia with abnormal eosinophils and inv(16) or t(16,16) has a favourable prognosis. Blood 1986; 6: 1242-9.
4. Campell LJ, Challis J, Fok T, Garson OM. Chromosome 16 abnormalities associated with myeloid malignancies. Genes Chromosomes Cancer 1991; 3: 55-61.
5. Ohyashiki K, Ohyashiki JH, Iwabushi A, Ito H, Toyama K. Central nervous system involvement in a acute nonlymphocytic leukaemia with inv(16)(p13q22). Leukemia 1988; 2: 398-9.
6. Harrison, Christine J. The Management of patients with leukaemia: The role of cytogenetics in this molecular era (review). Br J Haematol 2000; 108: 19-33.
7. Shurtleff SA, Meyers S, Hiebert SW *et al*. Heterogeneity in CBF beta/MYH11 fusion messages encoded by the inv(16)(p13q22) and the t(16,16)(p13q22) in acute myelogenous leukemia. Blood 1995; 85: 3695-703.
8. Betts DR, Rohatiner AZ, Evans ML, Rassam SM, Lister TA, Gibbons B. Abnormalities of chromosome 16q in myeloid malignancy: 14 new cases and a review of the literature. Leukemia 1992; 6: 1250-6.
9. Tien HF, Wang CH, Lin MT *et al*. Correlation of cytogenetic results with immunophenotype, genotype, clinical features, and ras mutation in acute myeloid leukemia. A study of 235 Chinese patients in Taiwan. Cancer Genet Cytogenet 1995; 84: 60-8.
10. Schoch C, Haferlach T, Haase D *et al*. Patients with *de novo* acute myeloid leukemia and complex karyotype aberrations show a poor prognosis despite intensive treatment: a study of 90 patients. Br J Haematol 2001; 112: 118-26.
11. Braess J, Jahns-Streubel G, Schoch C *et al*. Proliferative activity of leukaemia blasts and cytosine arabinoside pharmacodynamics are associated with cytogenetically defined prognostic subgroups in acute myeloid leukaemia. Br J Haematol 2001; 113: 975-82.
12. Wheatley K, Burnett AK, Goldstone AH *et al*. A simple, robust, validated and highly predictive index for the determination of risk-directed therapy in acute myeloid leukaemia derived from the MRC AML 10 trial. Br J Haematol 1999; 107: 69-79.