diagnostic regions. In bacteria, three genes make up the rRNA functionality (i.e., 5S, 16S and 23S rRNA). Historically, the 16S rRNA gene has been employed most commonly and hence has the most comprehensive database for comparison during searches of unknown sequences. Use of such rRNA-based techniques has gained increasing popularity as a means of identifying organisms that are troublesome and phenotypically difficult to identify. Even with improvements to conventional phenotypic identification techniques, certain genera and species continue to cause diagnostic dilemmas for the routine clinical microbiology laboratory. Given that most clinically significant organisms are relatively easy to identify by routine phenotypic laboratory methods (e.g., API identification scheme), an identification dilemma may still exist with the correct identification of certain genera and species of unusual but clinically significant bacteria.

What does the adoption of such techniques mean for the routine clinical microbiology laboratory? The authors have found these techniques to be particularly useful in respiratory specimens (atypical TB and CF) and in a small proportion of blood cultures, including those from haematology patients. As such, molecular techniques have become commonplace in many specialist and reference laboratories, with the value of associated reports dependent on how these methods perform in the routine setting. Continued use of such techniques will permit the description of emerging bacterial pathogens, which to date have not been described, as illustrated by the publication of several case reports using this technology.[3,4]

For the medical microbiologist, such techniques should be regarded as simply another technology to obtain quality data on the identification of isolates, where interpretive criteria on the clinical significance of the organism is no different to those isolates identified conventionally.[5]

Judging by the authors' experience, specimens for molecular analysis originate from a relatively narrow source (respiratory; CF and atypical mycobacteria) and blood culture, even in a large teaching hospital, indicating that adoption of such technology in all clinical microbiology laboratories is unwarranted and not practical. Hence, such techniques should be restricted to specialist/reference laboratories.

The inability to report accurately the identification of an organism to clinical users of the service may compromise the clinical management of the patient (e.g., if the unidentified organism is important in terms of infection control). When such molecular techniques are adopted, the laboratory is able to provide reliable identification of organisms, which is a fundamental objective of any microbiology service.

In conclusion, when use of molecular identification methods is justified, employment of partial 16S rDNA PCR and sequencing provides a valuable and reliable method of identification for bacteria that prove difficult to identify by conventional phenotypic techniques.

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An unbalanced translocation, der(17)t(1;17)(p13;p11.2), leads to heterozygous loss of *TP53* and is associated with clinical evolution in myelodysplastic syndrome

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Cytogenetic abnormalities are found in half of all myelodysplastic syndrome (MDS) cases.[1,2] Gross karyotypic changes are frequent, including chromosomal gain (most commonly trisomy 8) and chromosomal deletion and loss (most notably monosomy 5 and 7, loss of Y, deletion of 5q and 7q).[3] These changes are usually found in the setting of a complex karyotype.

In comparison to the situation in acute myeloid leukaemia (AML), isolated reciprocal translocations are uncommon in MDS. This has hampered the identification of specific genetic defects, although the prognostic importance of chromosomal abnormalities is well recognised in MDS.[4]

This study reports a der(17)t(1;17) (p13;p11.2) unbalanced translocation as the sole abnormality in a case of MDS showing disease progression.

A 71-year-old woman presented with the symptoms of anaemia. Peripheral blood count showed the following values: haemoglobin: 7.8 g/dL, platelets: 372 x 10^9/L and leucocytes: 3.4 x 10^9/L (66% neutrophils, 20% lymphocytes, 10% monocytes, 2% basophils and 2% myelocytes). Bone marrow examination revealed hypercellular particles with 3% blasts and dysplastic granulopoiesis and megakaryopoiesis. Marked erythroid hypoplasia was noted. Iron staining revealed no ringed sideroblasts.

A diagnosis of refractory cytopenia with multilineage dysplasia (RCMD) was made, in accordance with the World Health Organization (WHO) classification.[5] Cytogenetic analysis performed by overnight fluorodeoxyuridine-

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synchronised culture of marrow mononuclear cells showed a normal female karyotype (46, XX). The patient received supportive treatment, including antibiotics for infection, and transfusions. Chemotherapy was not given.

One year later, the patient developed thrombocytopenia and leucocytosis. Her blood count at the time showed the following values: haemoglobin: 8.8 g/dL, platelets: 28 x 10^9/L and leucocytes: 25.7 x 10^9/L (71% neutrophils, 8% lymphocytes, 1% monocytes, 1% eosinophils, 6% blasts, 6% myelocytes and 10% metamyelocytes). Blood film examination revealed frankly dysplastic neutrophils.

Bone marrow study showed extremely active, left-shifted and dysplastic granulopoiesis with a blast count of 10%. Once again, dysmegakaryopoiesis and erythroid hypoplasia were evident. No ringed sideroblasts were seen. Disease progression to refractory anaemia with excess blasts was diagnosed.[5]

Repeated cytogenetics analysis showed a clonal abnormality, 46,XX,der(17)t(1;17) (p13;p11.2)[18]/46,XX[1] (Fig. 1A). Fluorescence *in situ* hybridisation (FISH) using a SpectrumOrange probe for *TP53* on 17p13.1 (Abbott Molecular/Vysis, Des Plaines, IL) was performed on the nucleated marrow cells. A 200-cell count revealed a single *TP53* deletion in 86% of the cells (Fig. 1B).

Chromosomal translocations are uncommon in MDS and are usually accompanied by other complex changes (most notably -5, -7 and +8).[6] Unbalanced translocations that result in partial trisomy and monosomy provide a pathogenic scenario for the development of MDS. The unbalanced form der(17)t(1;17)(p13;p11) in this case report has been found in only four other patients to date (one B-lineage and one T-lineage precursor lymphoblastic leukaemia, one adenocarcinoma and one osteosarcoma).[6] All four cases showed a complex karyotype. The present case is the first in which this unbalanced translocation has been found in isolation and in a myeloid neoplasm.

p53 is an important tumour suppressor through its regulation of the cell cycle and apoptosis following DNA damage.[7] Inactivating *TP53* mutations at 17p13.1 have been detected in 50% of solid tumours. In AML/MDS, however, deletion and mutation of *TP53* has been found in only 17% and 10% of cases, respectively.[8]

In the case reported here, there is evidence from FISH studies of a loss of *TP53* heterozygosity through deletion of 17p from the unbalanced translocation. A search of the Mitelman database[6] showed that most of the reported cases of MDS with an unbalanced translocation involving 17p13.1 are associated with complex karyotypic changes. Thus, the contribution that loss of *TP53* has to pathogenesis and progression in these cases is unclear.

In contrast, the detection of der(17)t(1;17)(p13;p11.2) as a sole chromosomal abnormality at disease evolution in the present case, with further evidence for *TP53* deletion from FISH study, suggests a causal role for the p53 pathway in the progression of MDS. This is consistent with the reported association of p53 inactivation and myeloblastic transformation in chronic myeloid leukaemia in clinical[8] and animal studies.[9,10]

All seven previously reported MDS cases with der(17)t(1;17) showed breakpoints in 1q, and thus partial trisomy 1q. Five had a breakpoint at 1q21, where several candidate genes (including *MLLT11* and *MCL1*) implicated in myeloid malignancies reside. This suggests that changes in the partner chromosome of der(17)t(17;v) may have a role in the pathogenesis of MDS, either in relation to partial trisomy of the partner chromosome or to disruption of genes at the breakpoint.

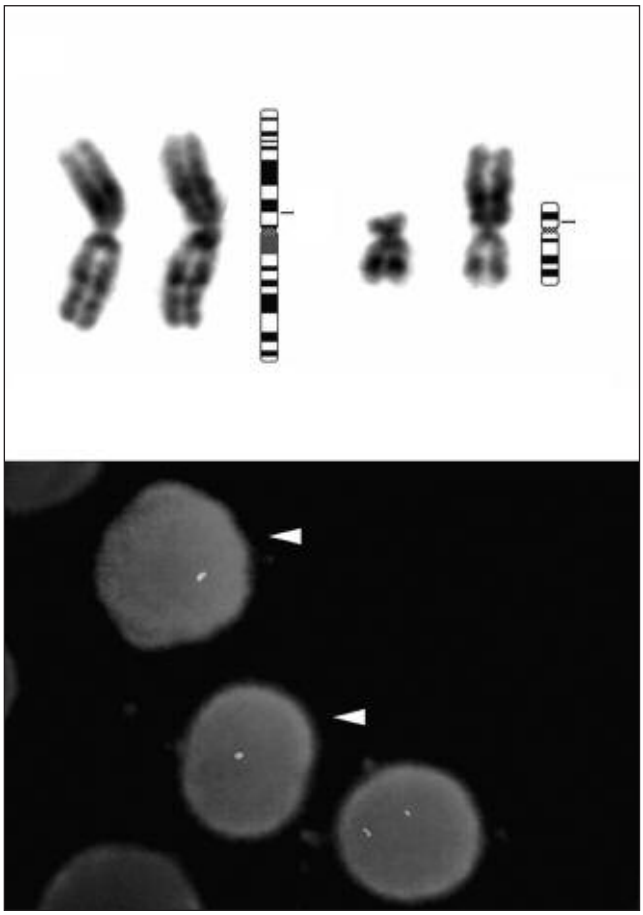

Fig 1. A) Partial karyotype showing der(17)t(1;17)(p13;p11.2). G-banding with trypsin-Giemsa. B) Interphase fluorescence *in situ* hybridisation analysis using the SpectrumOrange probe for *TP53* on 17p13.1. A single signal in two marrow cells (arrowheads) is seen, consistent with heterozygous deletion of *TP53*.

In contrast, the unbalanced translocation in the present case showed partial trisomy of 1p. While loss of heterozygosity on 1p has been reported in MDS,[11] partial trisomy 1p is not a recognised change. Interestingly, a gene of the *RAS* family, *RAP1A* (Ras-proximate 1), is located at 1p13.3, and expression of a mutated homologue of *RAP1A* (*RAP1B-retro*) has been found in MDS and AML.[12,13]

Clearly, further study of the contribution of trisomy 1p and *RAP1A* mutation in the pathogenesis and progression of MDS is warranted.

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