FANCONI ANEMIA FOUNDER MUTATION IN MACEDONIAN PATIENTS

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INTRODUCTION

Fanconi anemia (FA) is a rare autosomal recessive disorder (1-5/1000 000 live births) genetically and phenotypically heterogeneous, defined by cellular hypersensitivity to DNA cross-linking agents. Clinically FA is characterized by variable developmental abnormalities, stem cell loss, causing progressive bone marrow failure (BMF) and sterility, and profound predisposition to neoplasia mostly leukemia and solid tumors. FA is the most frequent inherited cause of BMF.

Approximately 65% of all affected individuals have mutation in *FANCA* gene. The mutation spectrum of the *FANCA* gene, located on chromosome 16q24.3, is highly heterogeneous, including point mutations, small insertions or deletions, splicing mutations, and large intragenic deletions. FA-A is usually associated with private *FANCA* mutations in individual families. Thus, the number of different pathogenic variants described for the *FANCA* gene is very high considering the relatively low number of patients.

MATERIAL AND METHODS

Clinical findings of the FA patients are given in Table 1.

The molecular analysis of *FANCA* gene was done by the MLPA Fanconi anemia P-031 kit (MRC Holland, The Netherlands) and direct sequencing using BigDye Terminator Sequencing kit v1.1, performed on ABI 3130 Genetic Analyzer (Life Technologies, USA).

Table 1. Clinical presentation of the Fanconi anemia patients.

Patient Initials	Age/	BMF	Ren arquatus	Renal ectopy	Skin pigment. *(café-au-lait)*	PDA	VUR	Ptosis
J.I.	11/ F	+	+	+	+	+	+	+
F.N.	17/ F	+	+	+	+	-	-	-
P.J.	23/M	+	+	+	+	-	-	-

F=female, M=male, BMF=bone marrow failure, PDA=persistent ductus arteriosus, VUR=vesico-uretral reflux.

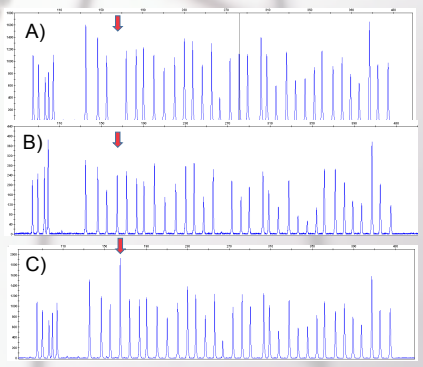

Figure 1. MLPA analysis in a patient homozygous for exon 3 deletion (A), heterozygote for exon 3 deletion (B) and normal sample (C).

RESULTS

The MLPA analysis of *FANCA* gene showed homozygous deletion of exon 3 in all three patients. Familial analysis in patient F.N. proved that the mutation was inherited from her parents. Representative electro-phoreograms of the MLPA analyses are shown in Figure 1.

Sequencing analysis of the flanking regions of exon 3 precisely defined a deletion of 2040 bp and insertion of C (1788_3824insC) (Figure 2) and identified the breakpoints in the *Alu* repetitive sequences of introns 3 and 4 (Figure 3).

These are the first three patients homozygous for deletion of FANCA exon 3 described to date. The patients are not related but they came from the same region from Macedonia.

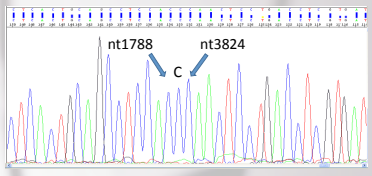

Figure 2. Sequence analysis of the fusion sequence showing the breakpoints of the deletion.

CONCLUSIONS

The homozygous deletion of exon 3 of *FANCA* gene is a founder mutation of Fanconi anemia patients in Macedonia. Our finding has very strong implication in diagnostic and carrier screening strategy for BMF and Fanconi anemia in Macedonian patients and in comprehensive genetic counseling.

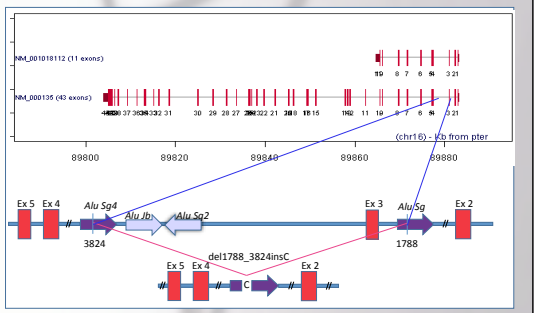

Figure 3. Schematic drawing of *FANCA* gene and breakpoint characterization of the deletion of FANCA ex 3 (1788_3824insC).

ACKNOWLEDGMENTS

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