

Azathioprine-induced severe aplastic anaemia in thiopurine S-methyltransferase deficient patient with Crohn's disease

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Abstract

*TPMT is an enzyme that catalyses S-methylation of thiopurine drugs, which activity is genetically determined. TPMT-deficient patients are at risk of toxicity after standard doses of thiopurine drugs. We describe a case of Crohn's disease diagnosed patient inheriting genetic TPMT deficiency (TPMT*3A/*3A genotype), who developed severe aplastic anaemia, confirmed by biopsy, after 55 days of treatment with AZA at a standard daily dose of 2.5 mg/kg. This case demonstrates the importance of TPMT polymorphism testing in order to prevent myelotoxicity during treatment with thiopurine drugs.*

Key words: Thiopurine S-methyltransferase, azathioprine, Crohn's disease, aplastic anaemia

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Introduction

Thiopurine S-methyltransferase (TPMT) is a cytoplasmic transmethylase that catalyses methylation of sulfur atoms in aromatic and heterocyclic compounds, among them thiopurine drugs, e.g. azathioprine (AZA) [1]. AZA is a drug initially converted in the liver to mercaptopurine, which is subsequently metabolized by two enzymes: xanthine oxidase, an intracellular enzyme present in the liver and the gastrointestinal tract, but not in hematopoietic tissue, or alternatively, by TPMT, an enzyme present in most tissues, including hematopoietic tissues and red blood cells (RBCs) [2]. There is a large interindividual variability of TPMT activity, mainly due to genetic polymorphism. The distribution of enzyme activity is trimodal. In Caucasian population over 90% of subjects shows high TPMT activity (13.7-20.0 U/mL of packed RBCs), corresponding to the homozygous wild-type genotype. Some 10% is heterozygous for TPMT gene locus and characterized by intermediate activity (5.0-13.7 U/mL of packed RBCs), and homozygotes, with two TPMT mutant alleles, have deficient TPMT activity (< 5 U/mL of packed RBCs) and account for only 0.3% of the population [3]. It was demonstrated that TPMT-deficient

patients accumulate very high concentrations of toxic thioguanine nucleotides (TGNs) in hematopoietic tissues and suffer from hematopoietic toxicity, including neutropenia, anemia, and thrombocytopenia [4]. The three variant alleles: TPMT*2, *3A and *3C are responsible for over 95% cases of lower enzyme activity [5]. This report demonstrates how life-threatening myelotoxicity was induced in a TPMT-deficient genotype patient diagnosed with Crohn's disease after nearly 2-months AZA-exposure, pointing out the importance of TPMT genotype evaluation in the treatment with thiopurine drugs.

Case report

A 49 year-old male patient diagnosed with Crohn's disease at the age of 45 medicated with mesalazine (3 g/daily) and prednisone (20 mg/daily) was admitted to hospital after flare up. He was started azathioprine (AZA) at a daily dose of 2.5 mg/kg. After a 55 days of treatment he developed severe aplastic anaemia (SAA), confirmed by bone marrow biopsy (Fig. 1) and was hospitalized. The following blood parameters at the admission were recorded: RBC - 2.96 T/L, HGB - 8.5 g%, Ht - 0.238 l/l, WBC - 1,4 x G/L (neutrophils

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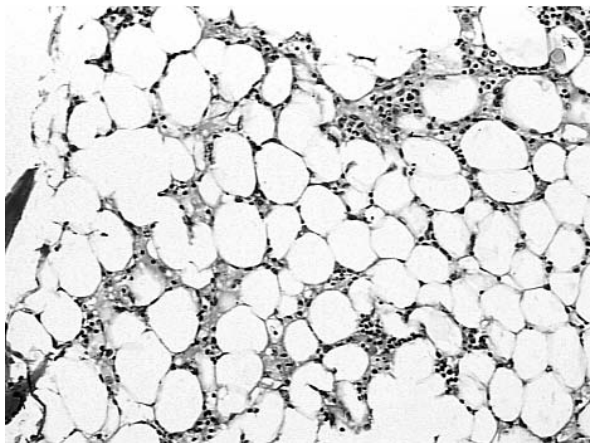


Fig. 1. Bone marrow trephine biopsy, confirming severe aplastic anemia (SAA). H & E x 400

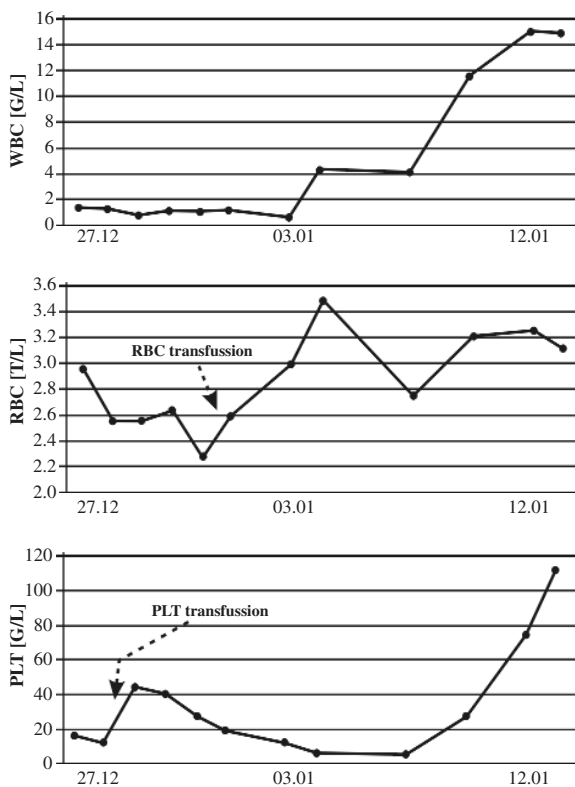


Fig. 2. Patient's blood morphology in the course of treatment

count 0,3 G/L), PLT - 17.0 G/L, and AZA along with mesalazine were withdrawn from medication. Genotyping for TPMT polymorphism (*1, *2, *3A, *3B and *3C) was performed by PCR-based methods, described by us elsewhere [6], and revealed variant homozygous genotype (*TPMT*3A/*3A*), determining deficiency of TPMT activity. After AZA withdrawal from treatment the patient's blood

cell count's started to normalize. He was prescribed methylprednisolone (1000 mg iv on days 1-3 and 500 mg iv on days 4-15), cefepime, red cell and platelet transfusions and finally disposed home 17 days afterwards AZA withdrawal, with the following blood parameters: RBC - 2.99 T/L, HGB 8,9 g%, WBC - 14.9 G/L (neutrophil count 12,07 G/L), PLT - 111.0 G/L. Figure 2 presents patient's blood morphology on course of treatment.

Discussion

We describe a case of a patient inheriting genetic TPMT deficiency, treated with standard doses of AZA who developed severe myelosuppression after 55 days of treatment with AZA at a daily dose of 2.5 mg/kg. This observation is in concordance with the previous reports, indicating that myelosuppression in TPMT-deficient patients treated with a standard dosage of thiopurines occurs on average 1 month after initiation of treatment [7, 8]. Available data shows, that TPMT-deficient Crohn's disease patients can be efficiently and safely treated with 5-15% of standard AZA dose (0.16-0.29 mg/kg) or AZA should be replaced by other medication, otherwise it involves severe myelosuppression [7, 8, 9]. In the present report a clear coincidence of AZA administration and severe myelosuppression is documented. As it is known that patients carrying *TPMT*3A/*3A* genotype exhibit very low or no enzyme activity, it can be concluded that accumulation of toxic thioguanine nucleotides in a TPMT deficient patient was responsible for the observed SAA. On the other hand, it had been presented that benzoic acid derivatives such as sulfasalazine, olsalazine and mesalazine (5-aminosalicylic acid), are moderately potent inhibitors of TPMT, and co-administration of these drugs with AZA may lead to myelosuppression, even in patients having TPMT activity within normal range [10, 11]. In the case of patient described above, in the lack of a genetic test, TPMT deficiency could have been easily mistaken with the result of AZA/mesalazine drug interaction. TPMT - genotyping allows to establish etiology of complications and also prevent the patient from AZA standard dosage administration in the future. Even though individuals carrying two variant TPMT alleles are relatively rare in general population (0.3%), it was revealed that they might be overrepresented in a group of patients experiencing leukopenia during AZA therapy - up to 10% in a study of Colombel et al. [7]. Heterozygous Crohn's disease patients with one TPMT variant allele, standing for about 10% of white population [3, 12], are also at greater risk of early bone marrow toxicity than wild-type homozygotes [13]. Identification of heterozygotes would also indicate those patients who could be safely managed on AZA dosages lower than 2.0 mg/kg [13, 14]. Even if some studies did not find measurement of TPMT activity cost-effective [15], it still seems that TPMT phenotyping or genotyping is the only way to avoid severe, potentially lethal toxic effects of AZA in patients carrying two variant TPMT alleles. For the reasons

given above, evaluation of TPMT polymorphism in patients treated with thiopurine drugs should be mandatory in order to optimize therapy.

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