

Case Report

Agranulocytosis Induced by Overdosage of Mercaptopurine: A Case Report

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Abstract

Drug-induced agranulocytosis (DIAG) is a rare but potentially fatal hematological complication. Thiopurines, such as mercaptopurine (6-MP), are widely used in the treatment of chronic inflammatory bowel diseases (IBD), but can cause myelotoxicity due to the accumulation of active metabolites. We report the case of a 48-year-old woman with ulcerative colitis who developed severe agranulocytosis following an accidental overdose of 6-MP. The patient presented with febrile pancytopenia with a neutrophil count of 0.04 G/L. Bone marrow examination revealed normal cellularity with an inversion of the maturation pyramid of the granulocyte lineage. A treatment combining growth factors (filgrastim) and antibiotics (tazobactam) was initiated, leading to progressive improvement over 5 weeks. The toxicity of 6-MP is mainly due to the accumulation of 6-thioguanine nucleotides (6-TGN). The polymorphism of the gene encoding the enzyme thiopurine S-methyltransferase (TPMT) can influence the risk of myelotoxicity. Two strategies are proposed to minimize this risk: evaluation of TPMT activity with dosage adjustment, or regular monitoring of blood counts with gradual dose increase. This case highlights the importance of rigorous biological monitoring when initiating 6-MP treatment, regardless of the TPMT test. Increased vigilance is necessary when using immunosuppressive drugs in the treatment of IBD to optimize efficacy and minimize toxicity.

Keywords

Inflammatory Bowel Disease, Myelotoxicity, Immunosuppressant, Mercaptopurine, Agranulocytosis

1. Introduction

Drug-induced agranulocytosis (DIAG) is a rare but serious hematologic condition characterized by a significant decrease in the number of neutrophils in the peripheral blood. Although the definition has not yet been universally established, particularly regarding the depth of neutropenia [1, 2], in practice, it is defined by neutropenia less than 500/mm³ [1], meeting strict causality criteria reported by Bénichou et al. [3], em-

phasizing the importance of establishing a causal relationship between drug administration and the occurrence of neutropenia. These agranulocytoses sometimes require hospitalization and specialized management. In this paper, we report a case of agranulocytosis induced by mercaptopurine (6-MP). Thiopurines, such as azathioprine and mercaptopurine, are widely used immunosuppressive drugs in the treatment of

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chronic inflammatory bowel diseases (IBD), including Crohn's disease and ulcerative colitis (UC) [4, 5]. They play a crucial role in maintaining remission and managing symptoms in many patients. However, the use of these drugs is associated with an increased risk of severe myelosuppression, especially in patients with reduced activity of the enzyme thiopurine S-methyltransferase (TPMT) [6, 7].

2. Case Presentation

We report the case of a 48-year-old female patient. Her medical history includes treated and resolved pulmonary tuberculosis 20 years ago, with no other notable pathological history (no active or passive smoking, no family history of IBD).

Initial Symptoms and Diagnosis: The patient presented with symptoms starting two years ago, marked by the onset of bloody mucus diarrhea at a frequency of 1 to 2 stools per day. This was accompanied by 6 to 7 mucous-bloody, non-fecal discharges and atypical, moderate-intensity peri-umbilical abdominal pain, without radiation, aggravating or relieving

factors, or other associated digestive or extra-digestive signs. The condition evolved in a febrile context with general health deterioration (asthenia, anorexia, and unspecified weight loss). The diagnosis of UC of undetermined extent was made. The initial flare was considered moderate, and treatment was initiated with Mesalazine 2g/day as maintenance therapy, resulting in good clinical and biological improvement.

Subsequent Severe Flare and Hospitalization: The patient experienced a second, severe flare and was admitted to the Hepato-Gastro-Enterology department of CHU Mohammed 6 in Marrakech. During hospitalization, colonoscopy confirmed active UC (Mayo score 3) with endoscopic severity signs. Intravenous corticosteroid therapy was administered for 7 days without significant improvement, and treatment was switched to Ciclosporine 5mg/kg/day for 3 months, yielding a good clinico-biological response. Subsequently, Azathioprine (2.5 mg/kg/day) was initiated in March 2022 as maintenance therapy but was discontinued in January 2023 due to bicytopenia (Hemoglobin: 10.3 g/dl, White Blood Cells: 2.09 G/L), despite a normal TPMT level (9.5).

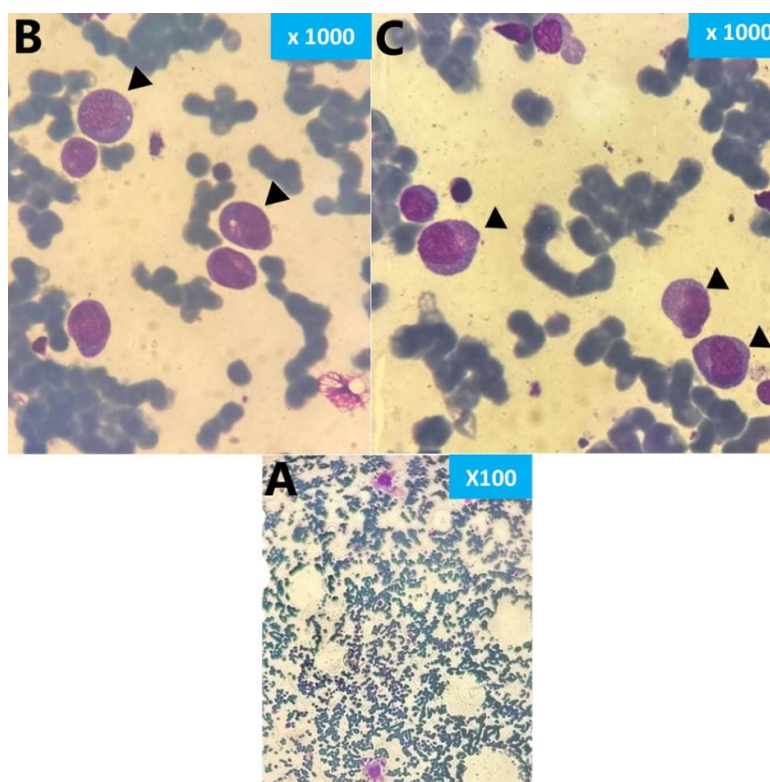


Figure 1. Appearance of promyelocytic blockage in the bone marrow (MGG stain): A: Low magnification, Bone marrow of normal cellularity, with megakaryocytes present. B and C: High magnifications, Blockage of granular differentiation with an excess of normal-appearing promyelocytes (black arrows): large cells, highly granular basophilic cytoplasm, and well-visible archoplasm.

Complications and Management: The patient was then started on 6-mercaptopurine at 1.5 mg/kg/day in February 2023, with an incident of overdosage (3g/day for 3 weeks). In March 2023, she developed febrile pancytopenia, with bio-

logical exams revealing agranulocytosis (neutrophils: 0.04 G/L), anemia (Hemoglobin: 5.1 g/dl), and thrombocytopenia (Platelets: 37 G/L) without other abnormalities on the blood smear. Precautionary isolation and cessation of treatment

were implemented. Bone marrow aspiration revealed a regenerative marrow with normal cellularity and an inverted maturation pyramid of the granulocyte lineage, with no notable morphological abnormalities or abnormal cells, compatible with peripheral pancytopenia (Figure 1). A treatment combining growth factors and antibiotics (Filgrastim 30 MUI/day for 5 days and Tazobactam 4g 4 times/day for 7 days) was initiated, leading to good biological improvement. A progressive correction of agranulocytosis was observed over 5 weeks (neutrophils: 0.04 G/L on day 1, 0.08 G/L on day 6, 0.55 G/L on day 12, 1.20 G/L on day 32).

3. Discussion

Many drug classes [8] have the potential to induce agranulocytosis. For most, the risk is very low, except for certain

contemporary drugs such as clozapine, carbimazole, ticlopidine, sulfasalazine, trimethoprim-sulfamethoxazole, penicillamine, and phenylbutazone. (Table 1) lists the medications most commonly associated with agranulocytosis. According to the experience of the University Hospitals of Strasbourg [1], antibiotics are the most frequent cause of agranulocytosis (25%), particularly beta-lactams and trimethoprim-sulfamethoxazole, followed by antithyroid drugs (23%) and antiplatelet agents (16%). These findings align with recent studies by van der Klauw et al [9] and Shapiro et al [10]. In clinical practice, it is often extremely challenging to establish a causal relationship between the hematologic incident and a specific drug, especially when patients are on multiple medications simultaneously. The duration of exposure to the offending drug is typically around one month.

Table 1. Drugs most frequently associated with DIAG [2-10].

DRUG CLASSES	DRUGS
ANTIBIOTIQUES	Cephalosporins, Chloramphenicol, Ciprofloxacin, Clindamycin, Cotrimoxazole, Tetracyclines, Ethambutol, Gentamicin, Isoniazid, Lincomycin, Metronidazole, Nitrofurantoin, Novobiocin, Penicillins, Rifampicin, Sulfamethoxazole, Sulfonamides, Streptomycin, Thiacetazone, Tinidazole, Vancomycin, Chloroquine, Flucytosine, Dapsone, Hydroxychloroquine, Levamisole, Mebendazole, Pyrimethamine, Quinine, Quinacrine, Acyclovir, Zidovudine, Terbinafine
ANALGESICS AND NSAIDS	Acetylsalicylic acid, Diclofenac, Diflunisal, Tenoprofen, Flurbiprofen, Indomethacin, Ibuprofen, Noramidopyrine, Phenylbutazone, Piroxicam, Sulindac, Tenoxicam, Tolmetin
ANTIPSYCHOTICS, SEDATIVES AND ANTIDEPRESSANTS	Amoxapine, Chlordiazepoxide, Clozapine, Diazepam, Haloperidol, Tricyclic antidepressants, Meprobamate, Mianserin, Phenothiazines, Risperidone, Tiapride
ANTIEPILEPTIC	Carbamazepine, Ethosuximide, Phenytoins, Trimethadione, Valproic acid
ANTITHYROID DRUGS	Carbimazole, Methimazole, Potassium perchlorate, Thiocyanate, Thiouracils
CARDIAC MEDICATIONS	Acetylsalicylic acid, Aprindine, Captopril, Furosemide, Hydralazine, Lisinopril, Methyl dopa, Nifedipine, Phenindione, Procainamide, Propafenone, Propranolol, Quinidine, Spironolactone, Thiazide Diuretics, Ticlopidine
ANTI-HISTAMINES	Brompheniramine, Chlorpheniramine, Cimetidine, Ranitidine, Tripeleminamine
HEAVY METALS	Arsenic derivatives, gold-containing compounds, and even mercury
VARIOUS	Acetazolamide, Allopurinol, Aminoglutethimide, Bezafibrate, Colchicine, Dapsone, Deferiprone, Famotidine, Fludione, Flutamide

Drug-induced agranulocytosis (DIAG) can result from immunoallergic phenomena and/or toxic mechanisms [10]. In cases of so-called immunoallergic DIAG, there is an indirect destruction mediated by antibodies present on the cell surface. These antibodies recognize the drug as a foreign antigen, leading to the destruction of the target cell through complement system activation or antibody-dependent cellular cyto-

toxicity (ADCC). On the other hand, toxic-origin agranulocytosis results from intermediate products arising from the metabolism of the implicated drug. These metabolites exert their toxicity directly on myeloid precursors or indirectly on the bone marrow microenvironment, thereby causing a dysregulation of normal granulopoiesis [11] (Table 2).

Table 2. Mechanisms of agranulocytosis [11].

TOXIC	IMMUNO-ALLERGIC
Direct cytotoxic effect of the drug on granulopoiesis	Indirect cytotoxicity through drug (antigen) binding to surface cell antibodies
Dose-dependent effect	Non-dose dependent
No prior sensitization effect	History of prior sensitization for more than 8 days
Progressive agranulocytosis	Abrupt agranulocytosis
Inhibition of granulopoiesis in vitro by the drug	Inhibition of granulopoiesis in vitro by the patient's serum
Example: Cytotoxic chemotherapy, Carbamazepine, Quinine, Phenothiazines	Example: Synthetic antithyroid drugs, Antimalarials, Penicillins

The clinical presentation modes of patients with DIAG vary widely. The classic ulceronecrotic sore throats, a major criterion for DIAG as described by Schultz et al. [12], and extensive cellulitis are becoming unusual 4 of 7 modes of presentation [13]. Discovery may be incidental on a blood test in an asymptomatic individual or revealed by signs of infection

such as 'bare' fever, sore throat, pneumonia, skin infection, cholecystitis, pyelonephritis... or bacteremia and septic shock [2-13]. According to a Strasbourg series of DIAG cases, isolated fever is reported in 40% of cases, septicemia and/or septic shock in 34% of cases, and localized infections in 26% of cases, with 10% being pneumonia [1] (Figure 2).

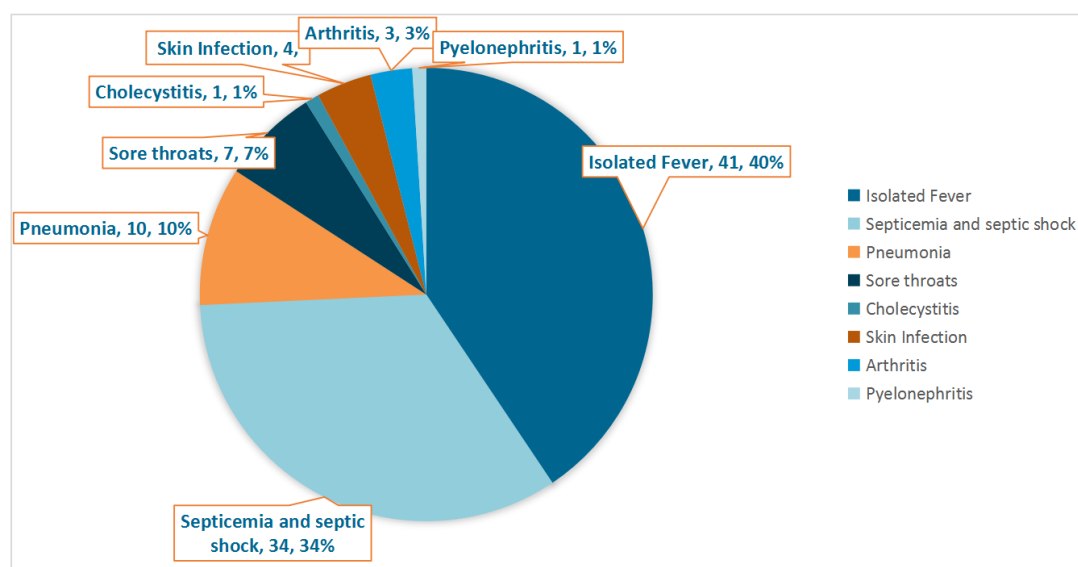


Figure 2. Main clinical manifestations reported in the series of drug-induced agranulocytosis at the University Hospitals of Strasbourg (n = 91) [1].

The mortality associated with DIAG is estimated to be around 10 to 16% in European studies dating back about twenty years [14]. Currently, it has decreased to approximately 5%, reflecting advancements in the management of DIAG [14, 13]. The main predictors of poor prognosis include age, neutrophil count at diagnosis lower than 100 cells/mm³, severe infectious state (septicemia and septic shock), and the presence of comorbidities, particularly renal insufficiency. Regarding the diagnostic approach to DIAG, it is crucial to specify the following elements: whether neutropenia is isolated or not. When neutropenia is the only cytopenia present,

it suggests a peripheral origin, while the coexistence of one or more other abnormalities suggests a central origin and requires a bone marrow examination. The acute or chronic mode of evolution. The commonly accepted delay to consider chronic neutropenia is 3 months [4]. The severity of neutropenia determines management (precautionary isolation/antibiotic therapy). The recent appearance of neutropenia is often associated with infectious (viral, bacterial, or parasitic), drug-induced, or autoimmune etiologies, or even a malignant hematologic disorder, while chronic evolution is more likely due to vitamin deficiencies, autoimmune etiology,

endocrine causes, or a hematologic disorder.

Systematic performance of a bone marrow examination in the case of DIAG in young individuals is debatable. However, in elderly individuals, the systematic performance of a bone marrow examination seems reasonable, especially to rule out myelodysplastic syndrome or another hematologic disorder, particularly considering that associated abnormalities in the blood count are frequently present: anemia (Hemoglobin > 12 g/dl) in 30% of cases and thrombocytopenia (less than 150 G/L) in 10% of cases. When a bone marrow examination is conducted, it typically shows normal or slightly decreased cellularity contrasting with the absence of myeloid cellular precursors [14, 13]. In the case of our patient, the medication inquiry led to the conclusion that the hematologic event was attributable to 6-MP, based on chronological criteria (compatible onset delay and regression upon treatment discontinuation) and biological findings (isolated neutropenia, absence of other causes, suggestive aspect on bone marrow examination). Literature review also supported the known effect of the molecule, and the absence of other suspicious treatments further strengthened this conclusion. Moreover, given the association of neutropenia with other hematologic abnormalities on the blood count, anemia (5.1 g/dl) and thrombocytopenia (37 G/L), a bone marrow examination was indicated. It revealed normal marrow cellularity with selective involvement of the granulocytic lineage estimated at 40%, marked by the predominance of early immature elements (myeloblast-promyelocyte) without notable morphological abnormalities and the absence of abnormal cells. A report to pharmacovigilance was made, and a list of contraindicated medications was provided to the patient.

The most common toxicity associated with 6-MP is agranulocytosis. This can manifest as anemia, leukopenia, thrombocytopenia, or a combination of these reactions. 6-MP is primarily metabolized by thiopurine S-methyltransferase (TPMT), an enzyme that plays a crucial role in the availability of pharmacologically active final metabolites: 6-thioguanine nucleotides (6-TGN) and 6-methylmercaptopurine (6-MMP) [15]. Myelotoxicity is mainly attributed to high concentrations of 6-TGN, leading to apoptosis and direct cytotoxicity due to DNA strand breakage [16]. However, when concentrations of 6-MMP are (extremely) high, as is the case with high-dose thiopurine therapy in oncology patients or in patients with IBD, 6-MMP can inhibit de novo purine synthesis, thereby causing subsequent myelotoxicity [17]. TPMT activity can be highly variable due to the gene polymorphism that codes for it. Approximately 0.3% of Caucasians and African Americans have a partial or complete deficiency in TPMT, as both alleles of the gene coding for this enzyme are non-functional in these individuals (homozygous deficiency). In 10% of patients, only one allele is functional, resulting in intermediate TPMT activity. However, for the remaining 90%, both alleles are functional, leading to normal TPMT activity. Patients with low or intermediate TPMT activity accumulate more toxic metabolites from the breakdown of 6-MP. Alt-

hough some phenotypic or genotypic tests can determine if an individual has a TPMT deficiency, these tests are not routinely performed [18]. The management of iatrogenic agranulocytosis relies on both protective isolation measures and antibiotic prophylaxis for associated infections, as well as on a thorough drug investigation to eliminate other toxic exposures and discontinue the implicated molecule, which will be definitively contraindicated. Recovery is generally complete. The use of growth factors may be considered based on prognostic factors and disease evolution, but their effectiveness remains controversial due to their short duration of action. Their utility may be more pronounced in older individuals. If their use is chosen, it is recommended to opt for the minimum effective dose [16]. Additionally, close hemodynamic and clinical monitoring (every 4 hours) should be implemented [19].

4. Conclusions

The myelotoxicity induced by 6-MP has been extensively elucidated over the years. When initiating 6-MP treatment, two strategies have been proposed to minimize this risk. The first approach involves assessing TPMT activity and adjusting the dosage in patients with complete or partial deficiency. The second approach involves regular monitoring of blood cell counts (CBC) and a gradual dose escalation until reaching the target dose to quickly detect any toxicity, thereby reducing the dose before severe complications occur. We emphasize the importance of regular CBC monitoring, starting in the first weeks after initiating treatment, regardless of whether TPMT testing is performed, to optimize efficacy and minimize toxicity.

Abbreviations

DIAG	Drug-Induced Agranulocytosis
6-MP	Mercaptopurine
IBD	Chronic Inflammatory Bowel Diseases
6-TGN	6-thioguanine Nucleotides
TPMT	S-methyltransferase
UC	Ulcerative Colitis
ADCC	Antibody-dependent Cellular Cytotoxicity
6-MMP	6-Methylmercaptopurine
CBC	Blood Cell Counts

Author Contributions

Touria El Bardi: Conceptualization, Data curation, Investigation, Writing – original draft, Writing – review & editing

Assia El Ouarradi: Funding acquisition, Investigation, Resources

Salma Rouhi: Formal Analysis, Funding acquisition, Methodology, Project administration, Resources

Wafae Quidi: Methodology, Supervision, Validation,

Visualization

Sanae Sayagh: Methodology, Supervision, Validation, Visualization

Conflicts of Interest

The authors declare no conflicts of interest.

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