British Oncology Pharmacy Association

bopa

Guideline for 6-mercaptopurine dosing in adult acute lymphoblastic leukaemia based on *TPMT* and *NUDT15* genotypes

Version 1.0, 15 March 2023

Document Control

Version	Date	Summary of changes
1.0	15/03/2023	New guideline

Document name	Guideline for 6-mercaptopurine dosing in adult acute lymphoblastic leukaemia based on *TPMT* and *NUDT15* genotypes
Version	1.0
Primary author(s)	Dr Deborah Yallop, Consultant Haematologist, King's College Hospital (deborah.yallop@nhs.net)
Primary author(s) supported by	Dharmisha Chauhan, Pharmacy Lead, NHS North Thames Genomic Medicine Service Alliance (dharmisha.chauhan1@nhs.net) Rachel Palmer, Pharmacy Lead, NHS South West Genomic Medicine Service Alliance (rachel.palmer6@nhs.net)
Acknowledgements	National Cancer Research Institute Acute Lymphoblastic Leukaemia subgroup (reviewed and provided comments on the guidance)
Ratified and endorsed by	British Oncology Pharmacy Association
Date of ratification and endorsement	15/03/2023

Conflicts of Interest

No conflicts to be declared.

Any questions on the guidelines please contact the authors listed above.

Disclaimer

The information contained in this guideline is a consensus view of the development group and consultation groups' views on current treatment and are intended only to assist clinicians in decision making. It should be used in conjunction with any local policies/procedures/guidelines and should be approved for use according to individual organisational clinical governance processes. Guidelines do not account for all individual variation, needs, preferences and values of patients and cannot be considered inclusive of all appropriate methods of care or exclusive of other treatments. Care has been taken in the preparation of the information contained in this guideline. Nevertheless, any person seeking to consult this guideline, apply its recommendations or use its content is expected to use independent, personal, medical and clinical judgement in the context of the clinical circumstances of an individual patient, or to seek out the supervision of a qualified clinician. It remains the responsibility of individual organisations to determine the best course of treatment for the patient. It is not mandatory to adhere to this guideline, with the ultimate determination regarding its application to be made by the clinician and the patient. The development group, and any endorsing group, make no representation or guarantee of any kind whatsoever regarding the guideline content or its use or application and disclaim any responsibility for its use or application in any way.

Contents

Purpose 5
Scope 5
Background 5
Recommendations 7
Ordering a Test 8
Clinical Interpretation of the Result 8
Definitions 12
References 13

Purpose

The aim of this guideline is to provide recommendations on 6-mercaptopurine (6-MP) dosing in adult acute lymphoblastic leukaemia (ALL) based on *TPMT* and *NUDT15* genotypes.

Scope

This guideline provides recommendations on the starting doses for 6-MP in adult patients aged 25 years and older who have a confirmed diagnosis of ALL and treatment with 6-MP is indicated. This guideline is relevant to all clinical staff involved in the management of adult patients aged 25 years and older diagnosed with ALL. The recommendations in this guideline are not intended to guide the treatment of patients under the age of 25 years or patients enrolled in a research study. In such circumstances, clinical staff should refer to the locally agreed treatment protocol or the research study treatment protocol.

Background

6-MP is a prodrug that is metabolised to active thioguanine nucleotides which are incorporated into DNA. Thiopurine S-methyltransferase (TPMT) and nudix hydrolase 15 (NUDT15) are key enzymes in the metabolism and inactivation of 6-MP and its active metabolites. Individuals with lower enzymatic activity are more likely to experience toxicity such as severe bone marrow suppression [1, 2]. TPMT and NUDT15 enzyme activity is regulated by common genetic polymorphisms which are inherited in an autosomal co-dominant manner and associated with lower enzymatic activity [3]. These single nucleotide polymorphisms (SNPs) are seen at variable frequencies across ethnicities.

TPMT

Over 90% of low TPMT enzyme activity phenotypes are accounted for by three SNPs: c.238G>C, c.460G>A and c.719A>G. [4] Between 30-60% of individuals who are heterozygous for *TPMT* variants (one non-functional *TPMT* allele) cannot tolerate full dose 6-MP [4]. Approximately 0.3% of individuals of European ancestry are homozygous for *TPMT* variants (two non-functional *TPMT* alleles) [1]. Individuals who are homozygous or compound heterozygous for *TPMT* variants are poor metabolisers of 6-MP and are at very high risk of experiencing myelosuppression when treated with standard doses of 6-MP.

NUDT15

Variants of *NUDT15* are less well described but the commonest variant correlated with low enzyme activity is c.415C>T [5], with near complete loss of enzyme activity in vitro [4]. Individuals who are homozygous for this *NUDT15* variant are at very high risk of myelosuppression with studies of childhood ALL demonstrating tolerance of only 8% of 6-MP planned dose in homozygous individuals (two non-functional *NUDT15* alleles) and 63% of 6-MP planned dose in heterozygous individuals (one non-functional *NUDT15* allele) [6].

Standard dosing of 6-MP in adult ALL protocols

Doses of 6-MP in adult ALL protocols are typically 60mg/m^2/day during induction and consolidation and starting doses in maintenance are up to 75mg/m^2/day. Starting doses in adults over 60 years are typically lower as per local protocols. During maintenance, doses of 6-MP are adjusted upwards in 25% dose increments (alternating with titration of oral methotrexate) at 4 weekly intervals to maintain the neutrophil count between 0.75 and 1.5 x 10^9/l and platelet count between 75 and 150 x 10^9/l. There is no maximum dose of 6-MP. If neutrophils fall below 0.75 x 10^9/l, 6-MP and methotrexate should be reduced by 50%, and if neutrophils fall below 0.5 x 10^9/l, both drugs should be stopped and restarted at 100% when neutrophils > 0.75 x 10^9/l. Similar adjustments are made to maintain the platelet count between 75 and 150 x 10^9/l.

Recommendations

Adult patients with a confirmed diagnosis of ALL and proposed treatment with 6-MP should be genotyped for the following *TPMT* and *NUDT15* no function variants prior to commencing 6-MP:

TPMT variants to be tested

Star (*) Allele	HGVS cDNA Nomenclature	HGVS Protein Nomenclature	rsID
*TPMT**2	NM_000367.5: c.238G>C	NP_000358.1: p.Ala80Pro	rs1800462
*TPMT**3A	NM_000367.5: c.460G>A, NM_000367.5: c.719A>G	NP_000358.1: p.Ala154Thr, NP_000358.1: p.Tyr240Cys	rs1800460, rs1142345
*TPMT**3B	NM_000367.5: c.460G>A	NP_000358.1: p.Ala154Thr	rs1800460
*TPMT**3C	NM_000367.5: c.719A>G	NP_000358.1: p.Tyr240Cys	rs1142345

Table 1: *TPMT* recommended variant alleles (HGVS, Human Genome Variation Society; rsID, reference single nucleotide polymorphism identifier). Note: *TPMT**1 is the normal function allele.

NUDT15 variants to be tested

Star (*) Allele	HGVS cDNA Nomenclature	HGVS Protein Nomenclature	rsID
*NUDT15**3	NM_018283.4: c.415C>T	NP_060753.1: p.Arg139Cys	rs116855232

Table 2: *NUDT15* recommended variant alleles (HGVS, Human Genome Variation Society; rsID, reference single nucleotide polymorphism identifier). Note: *NUDT15**1 is the normal function allele.

It must be noted that pharmacogenetic testing for the recommended *TPMT* and *NUDT15* variants will not exclude other *TPMT* and *NUDT15* variants that may cause individuals to be intermediate or poor metabolisers of 6-MP. Genomic laboratory reports should specify the variants tested and state that the test result does not exclude the presence of other *TPMT* and *NUDT15* variants.

Ordering a Test

Individual organisations should ensure there is a clear procedure for requesting *TPMT* and *NUDT15* genotyping and work with the appropriate testing laboratory to aim for a clinically appropriate turnaround time.

Clinical Interpretation of the Result

Patients who are identified as having one or more no function alleles should be considered for dose modification of 6-MP as suggested in Table 3, Table 4 and Table 5. If a patient tolerates the suggested dose modification based on their *TPMT* and *NUDT15* genotypes, doses can be carefully increased as determined by patient response and monitoring.

Where a patient has experienced toxicities but *TPMT* and *NUDT15* genotyping has shown none of the recommended variants to be present, consideration should be given to reducing the dose of 6-MP (and methotrexate) or stopping treatment of 6-MP (and methotrexate) and restarting in line with standard ALL protocols based on the clinical circumstances of the individual patient.

It is important to note that many factors can contribute to the variability in response to medicines. Additional factors that can affect the prediction of phenotype from genotype are environmental as well as endogenous in nature e.g. co-morbidities, co-medication, renal function, hepatic function. Non-genetic factors must be considered in the context of the clinical circumstances of an individual patient when applying the suggested dose modifications.

TPMT

Genotype Description	Variant Allele(s)	Diplotype	Likely Phenotype	Therapeutic Recommendation for 6-MP Dosing
Wild type	Not applicable	*TPMT* *1/*1	Normal metaboliser	Standard doses of 6-MP recommended.
Heterozygous	c.238G>C	*TPMT* *1/*2	Intermediate metaboliser	Patients generally tolerate a dose of 6-MP 50-60 mg/m^2 a day. Starting doses not exceeding 60mg/m^2/day are recommended and if tolerated the dose can then carefully be increased in 25% increments after at least a 2 week interval.
	c.460G>A, c.719A>G	*TPMT* *1/*3A		
	c.460G>A	*TPMT* *1/*3B		
	c.719A>G	*TPMT* *1/*3C		
Homozygous	c.238G>C	*TPMT* *2/*2	Poor metaboliser	Patient is at high risk of myelosuppression. It is recommended that 6-MP be commenced at 5mg/m^2 a day and if tolerated the dose can then carefully be increased in 10% increments at no more frequently than 4 weekly intervals.
	c.460G>A, c.719A>G	*TPMT* *3A/*3A		
	c.460G>A	*TPMT* *3B/*3B		
	c.719A>G	*TPMT* *3C/*3C		
Compound Heterozygous	c.238G>C; c.460G>A, c.719A>G	*TPMT* *2/*3A	Poor metaboliser	Patient is at high risk of myelosuppression. It is recommended that 6-MP be commenced at 5mg/m^2 a day and if tolerated the dose can then carefully be increased in 10% increments at no more frequently than 4 weekly intervals.
	c.238G>C; c.460G>A	*TPMT* *2/*3B		
	c.238G>C; c.719A>G	*TPMT* *2/*3C		
	c.460G>A, c.719A>G; c.460G>A	*TPMT* *3A/*3B		
	c.460G>A, c.719A>G; c.719A>G	*TPMT* *3A/*3C		
	c.460G>A; c.719A>G	*TPMT* *3B/*3C*		

Table 3: Therapeutic recommendation for 6-MP based on *TPMT* genotype and likely phenotype (assuming likely phenotype for *NUDT15* is normal metaboliser). Note: In extremely rare cases results reported as *TPMT* *1/*3A heterozygous genotype (intermediate metaboliser) may represent a *TPMT* *3B/*3C compound heterozygous genotype (poor metaboliser).

It is important to note that many factors can contribute to the variability in response to medicines. Non-genetic factors must be considered in the context of the clinical circumstances of an individual patient when applying the suggested dose modifications.

NUDT15

Genotype Description	Variant Allele(s)	Diplotype	Likely Phenotype	Therapeutic Recommendation for 6-MP Dosing
Wild type	Not applicable	*NUDT15* *1/*1	Normal metaboliser	Standard doses of 6-MP recommended.
Heterozygous	c.415C>T	*NUDT15* *1/*3	Intermediate metaboliser	Patients generally tolerate a dose of 6-MP 50-60 mg/m^2 a day. Starting doses not exceeding 60mg/m^2/day are recommended and if tolerated the dose can then carefully be increased in 25% increments after at least a 2 week interval.
Homozygous	c.415C>T	*NUDT15* *3/*3	Poor metaboliser	Patient is at high risk of myelosuppression. It is recommended that 6-MP be commenced at 5mg/m^2 a day and if tolerated the dose can then carefully be increased in 10% increments at no more frequently than 4 weekly intervals.

Table 4: Therapeutic recommendation for 6-MP based on *NUDT15* genotype and likely phenotype (assuming likely phenotype for *TPMT* is normal metaboliser).

It is important to note that many factors can contribute to the variability in response to medicines. Non-genetic factors must be considered in the context of the clinical circumstances of an individual patient when applying the suggested dose modifications.

TPMT & *NUDT15* Combination Phenotypes

***TPMT* Likely Phenotype**	***NUDT15* Likely Phenotype**	**Therapeutic Recommendation for 6-MP Dosing**
Intermediate metaboliser	Intermediate metaboliser	Patient is at high risk of myelosuppression. It is recommended that 6-MP be commenced at 5mg/m^2 a day and if tolerated the dose can then carefully be increased in 10% increments at no more frequently than 4 weekly intervals.
Intermediate metaboliser	Poor metaboliser	Patient is at high risk of myelosuppression. It is recommended that 6-MP be commenced at 5mg/m^2 a day and if tolerated the dose can then carefully be increased in 10% increments at no more frequently than 4 weekly intervals.
Poor metaboliser	Intermediate metaboliser	Patient is at high risk of myelosuppression. It is recommended that 6-MP be commenced at 5mg/m^2 a day and if tolerated the dose can then carefully be increased in 10% increments at no more frequently than 4 weekly intervals.
Poor metaboliser	Poor metaboliser	Patient is at high risk of myelosuppression. It is recommended that 6-MP be commenced at 5mg/m^2 a day and if tolerated the dose can then carefully be increased in 10% increments at no more frequently than 4 weekly intervals.

Table 5: Therapeutic recommendation for 6-MP based on *TPMT* and *NUDT15* likely phenotypes.

It is important to note that many factors can contribute to the variability in response to medicines. Non-genetic factors must be considered in the context of the clinical circumstances of an individual patient when applying the suggested dose modifications.

Definitions

Allele	Alternative forms of a gene at the same position on a chromosome
Autosomal co-dominant inheritance	A type of inheritance where two different alleles of the same gene within an individual's genome are expressed separately leading to different phenotypes in an individual.
Compound heterozygous	The presence of two different variant alleles of the same gene within an individual's genome
Diplotype	A combination of alleles found on different chromosomes within an individual's genome
Genotype	The DNA sequence of an individual, which determines (along with environmental influences) the specific characteristics (phenotype) of that individual.
Heterozygous	The presence of two different alleles of the same gene within an individual's genome
Homozygous	The presence of two identical alleles of the same gene within an individual's genome
Phenotype	An individual's observable physical and biochemical characteristics directly influenced by the genotype and/or environment.
Polymorphism	Genetic variants found in a significant proportion of the population (usually 1% of people) that may or may not be associated with disease. An association, however, does not necessarily mean that it is the cause of disease.
Variant	Any difference between the sequence of two individuals' genomes or an individuals' genomes to a reference genome
Wild type	A phenotype, genotype, or gene that predominates in a natural population

Definitions adapted from Health Education England Genomics Education Programme Glossary where available. [8]

References

1. Dean L, Kane M. Mercaptopurine Therapy and TPMT and NUDT15 Genotype. 2012 Sep 20 [updated 2020 Oct 26]. In: Pratt VM, Scott SA, Pirmohamed M, Esquivel B, Kane MS, Kattman BL, Malheiro AJ, editors. Medical Genetics Summaries [Internet]. Bethesda (MD): National Center for Biotechnology Information (US); 2012–. PMID: 28520348.

2. Dean L. Thioguanine Therapy and TPMT and NUDT15 Genotype. 2012 Sep 20 [updated 2020 Aug 7]. In: Pratt VM, Scott SA, Pirmohamed M, Esquivel B, Kane MS, Kattman BL, Malheiro AJ, editors. Medical Genetics Summaries [Internet]. Bethesda (MD): National Center for Biotechnology Information (US); 2012–. PMID: 28520351.

3. Lennard L. Implementation of TPMT testing. Br J Clin Pharmacol. 2014 Apr;77(4):704-14. doi: 10.1111/bcp.12226. PMID: 23962279; PMCID: PMC3971986.

4. Relling MV, Schwab M, Whirl-Carrillo M, Suarez-Kurtz G, Pui CH, Stein CM, Moyer AM, Evans WE, Klein TE, Antillon-Klussmann FG, Caudle KE, Kato M, Yeoh AEJ, Schmiegelow K, Yang JJ. Clinical Pharmacogenetics Implementation Consortium Guideline for Thiopurine Dosing Based on TPMT and NUDT15 Genotypes: 2018 Update. Clin Pharmacol Ther. 2019 May;105(5):1095-1105. doi: 10.1002/cpt.1304. Epub 2019 Jan 20. PMID: 30447069; PMCID: PMC6576267.

5. Moriyama, T., et al., NUDT15 polymorphisms alter thiopurine metabolism and hematopoietic toxicity. Nat Genet, 2016. 48(4): p. 367-73.

6. Yang, J.J. et al. Inherited NUDT15 variant is a genetic determinant of mercaptopurine intolerance in children with acute lymphoblastic leukemia. J. Clin. Oncol. 33, 1235–1242 (2015)

7. Royal Dutch Pharmacists Association (KNMP). Dutch Pharmacogenetics Working Group (DPWG). Pharmacogenetic Guidelines [Internet]. Netherlands. Azathioprine – TPMT and NUDT15 [Cited Dec 2019]. Available from: https://www.knmp.nl/media/1058

8. Health Education England Genomics Education Programme. Glossary [internet]. 2019 [cited 2022 September 16]. Available from; https://www.genomicseducation.hee.nhs.uk/glossary/