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Clinical Pharmacogenetics Implementation Consortium (CPIC) guideline for thiopurine dosing based on *TPMT* and *NUDT15* genotypes: 2018 update

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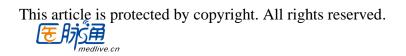
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CONFLICT OF INTEREST

The authors declared no competing interests for this work.

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ABSTRACT

TPMT activity exhibits a monogenic co-dominant inheritance and catabolizes thiopurines. *TPMT* variant alleles are associated with low enzyme activity and pronounced pharmalogic effecs of thiopurines. Loss-of-function alleles in the *NUDT15* gene are common in Asians and Hispanics and reduces the degradation of active thiopurine nucleotide metabolites, also predisposing to myelosuppression. We provide recommendations for adjusting starting dosesof azathioprine, mercaptopurine, and thioguanine based on *TPMT* and *NUDT15* genotypes (updates on www.cpicpgx.org) .

INTRODUCTION

This document is an update to the Clinical Implementation Consorium (CPIC) Guidelines for Thiopurine Methyltransferease Genotype and Thiopurine guideline updated last in April 2013. The guideline text, evidence table and recommendations have been updated to reflect any new evidence. Specifically, this guideline adds a recommendation for *NUDT15* genotype with minor changes to the TPMT recommendation. Although most of the dosing recommendations have been generated from clinical studies in just a few diseases, we have extrapolated recommended doses to all conditions, given the pharmacokinetic nature of the genotype/phenotype associations. CPIC guidelines are published and periodically updated on www.cpicpgx.org. Detailed guidelines for use of phenotypic tests (e.g. TPMT activity and thiopurine metabolite levels), as well as analyses of cost effectiveness, are beyond the scope of this document.

FOCUSED LITERATURE REVIEW

A systematic literature review focused on *TPMT* and *NUDT15* genotypes and thiopurine use was conducted (details in **Supplement**). Definitive reviews (1-4) were relied upon to summarize much of the earlier literature.

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DRUGS: THIOPURINES Background

Three thiopurines are used clinically: azathioprine (a prodrug for mercaptopurine), mercaptopurine, and thioguanine. Although all three medications share many of the same pharmacologic effects, mercaptopurine and azathioprine are generally used for non-malignant immunologic disorders, mercaptopurine for lymphoid malignancies, and thioguanine for myeloid leukemias. Because azathioprine is a prodrug for mercaptopurine, the two drugs can be considered to have identical interactions with TPMT and NUDT15. Recommendations for individuals with variants in one or both of these genes will be addressed in detail in the following sections.

GENES: TPMT AND NUDT15

Background

TPMT. TPMT activity is inherited as a monogenic, autosomal co-dominant trait (**Supplement**, **Figure S1**). Three *TPMT* single nucleotide polymorphisms (SNPs), which result in unstable proteins and enhanced TPMT protein degradation (2, 3), account for over 90% of low activity phenotypes and are the most common inactivating alleles, and so genotyping tests including these three variants have a high likelihood of being informative for TPMT phenotype (5, 6). Complementary phenotype laboratory tests can be helpful adjuncts to genotyping tests (**Supplement**, Other Considerations) (7).

TPMT catabolizes mercaptopurine to an inactive methylmercaptopurine base, leaving less parent drug available for eventual anabolism to active thioguanine nucleotides (TGNs, **Figure 1**). The secondary metabolite of mercaptopurine, thioinosine monophosphate (TIMP), is also a substrate for TPMT, and methylTIMP (and its further phosphorylated metabolites, methylmercaptopurine nucleotides or MeMPN) have pharmacologic activity (mostly immunosuppressive and hepatotoxic), inhibit *de novo* purine synthesis, and may contribute to some of the adverse effects of mercaptopurine, generally hepatotoxicity (2, 8, 9). Individuals who inherit two loss-of-function *TPMT* alleles (homozygous or

compound heterozygous *TPMT* deficient individuals) are at very high risk for life-threatening myelosuppression, due to very high TGNs, if given conventional doses of mercaptopurine (or azathioprine). Despite having higher TGNs than wild-type patients, only about 30-60% of *TPMT* heterozygotes cannot tolerate full doses of mercaptopurine or azathioprine (8, 10, 11). Good thiopurine tolerance in some heterozygotes may be because although they have higher TGNs than homozygous wild-type patients, they have lower concentrations (and thus fewer toxic effects) of the MeMPNs than do normal metabolizers, which may offset the toxic effects of having higher TGNs. Thus, there is less of a consensus over how to dose azathioprine and mercaptopurine in patients who are heterozygous for *TPMT* compared to those that are homozygous, although they are at a higher risk for toxicity compared to patients carrying two normal function alleles (12).

Although there is lower affinity between thioguanine and TPMT than between mercaptopurine and TPMT, TPMT significantly affects thioguanine pharmacokinetics and its cytotoxic effects (12-16). Thioguanine is directly metabolized by TPMT to inactive methylthioguanine base, leaving less drug available for anabolism by HPRT and other enzymes to active TGN metabolites. There is not an analogous secondary metabolite of thioguanine to undergo activation via TPMT (i.e. there are no methylTIMP or methylmercaptopurine nucleotides). As a result, patients receiving thioguanine are able to tolerate substantially higher TGN concentrations than do those receiving mercaptopurine or azathioprine (15). Within each TPMT phenotypic group, the initial recommended relative dosage decreases are similar for thioguanine, mercaptopurine, and azathioprine (**Table 2**).

NUDT15. Through agnostic genome-wide association studies, variants in *NUDT15* have been identified that strongly influence thiopurine tolerance in patients with acute lymphoblastic leukemia (ALL) (17) and those with inflammatory bowel diseases (18). As a nucleoside diphosphatase, NUDT15, catalyzes the conversion of cytotoxic thioguanine triphosphate (TGTP) metabolites to the non-toxic thioguanine monophosphate (TGMP). Defects in NUDT15-mediated degradation of TGTP results in more TGTP available for incorporation into DNA (DNA-TG, the primary antileukemic

metabolite (19)), thus allowing for DNA damage and apoptosis. The SNP (rs 116855232; c.415C>T) causing p.R139C was the first NUDT15 variant linked to thiopurine toxicity. It was shown that this amino acid change results in a nearly complete loss of enzymatic activity and protein stability in vitro. Patients carrying this allele showed excessive DNA-TG and severe myelosuppression (20). In children with ALL, patients homozygous for the p.R139C variant allele tolerated only 8% of the standard dose of mercaptopurine, whereas tolerated dose intensity was 63% and 83.5% for those heterozygous and wildtype for this SNP, respectively (17). While most clinical studies focused on mercaptopurine, in vitro experiments using laboratory models indicated similar influence of NUDT15 on the cytotoxicity of azathioprine and thioguanine (20). Additional variant alleles have been identified with varying prevalence among differing ancestral groups and varying degrees of functional effects (NUDT15 Allele Functionality Table and Frequency Table). The variant p.R139C has been studied most extensively in patients receiving thiopurine therapy, thus, providing the strongest evidence for clinical implementation. Subsequent studies reported additional variants, most of which are rare, and their associations with clinical thiopurine toxicity do not rise to clinical actionability at this point, even though some showed decreased NUDT15 activity in *in vitro*. For this reason, these variants (*4 to *9) are designated as unclear function but may be clarified as more data emerge.

Inherited TPMT deficiency is the primary genetic cause of thiopurine intolerance in Europeans and Africans, whereas risk alleles in *NUDT15* explain the majority of thiopurine-related myelosuppression in Asians and have been found in Hispanics.

Genetic Test Interpretation

Genetic testing analyzes the DNA sequence at specific SNP locations in the *TPMT* and *NUDT15* genes (**Supplement**). Each named star (*) allele is defined by the genotype at one or more specific loci (*TPMT* Allele Definition Table (21, 22) and *NUDT15* Allele Definition Table (21, 23)) and is associated with a level of enzyme activity (*TPMT* Allele Functionality Table (21, 22) and *NUDT15* Allele Functionality Table (21, 23)). Table 1 summarizes the assignment of the likely TPMT and

NUDT15 phenotypes, based on the most common * allele diplotypes, and these assignments are used to link genotypes with thiopurine prescribing recommendations. Of note, the phenotype of "possible intermediate metabolizer" has been introduced to the this guideline to describe an individual carrying one uncertain/unknown function allele PLUS one known no function allele, as this individual should be treated with "at least" the same precautions as would apply to an intermediate metabolizer. Although inactivating *TPMT* and *NUDT15* alleles have been identified in multiple populations (*TPMT* Frequency Table (21, 22) and *NUDT15* Frequency Table (21, 23)), one of the limitations inherent in a commercial genotype-only test is that rare or previously undiscovered variants may not be included.

Available Genetic Test Options

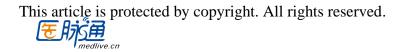
See **Supplementary material** and the Genetic Testing Registry (https://www.ncbi.nlm.nih.gov/gtr/) for more information on commercially available clinical testing options.

Incidental findings

There are no diseases or phenotypic traits that have been linked to variation in *TPMT* or *NUDT15* in the absence of thiopurine treatment (2).

Linking genetic variability to variability in drug-related phenotypes

There is substantial evidence linking *TPMT* and *NUDT15* genotype with phenotypic variability (see **Table S1**). Pre-emptive dose adjustments based on *TPMT* genotype have reduced thiopurine-induced adverse effects without compromising desired antitumor and immunosuppressive therapeutic effects in several clinical settings (**Table S1**). Similarly, retrospective studies strongly indicate that patients with loss-of-function *NUDT15* alleles are at excessive risk of thiopurine toxicity if the standard dose is administered. This body of evidence, rather than randomized clinical trials, provides the basis for most of the dosing recommendations in **Tables 2 and 3**.

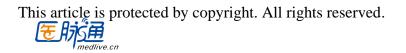


Therapeutic Recommendations

Thiopurines are used to treat malignant and non-malignant conditions, and thus the approach to dosing adjustments and the propensity to initiate therapy at higher vs. lower starting doses based on TPMT/NUDT15 status may differ depending on the clinical indication.. Thiopurines have a unique role in the treatment of many malignancies. The "normal" starting doses of thiopurines are generally "high" because they have been derived from trials which have been heavily weighted by the ~90% of the population who are wild-type for *TPMT* and *NUDT15* and receive maximal tolerable doses by the standards of anticancer treatment (hence, full doses should be given to those who are normal metabolizers for TPMT and NUDT15, **Tables 2 and 3**). Because the level of thiopurine tolerance is highly correlated with genetic ancestry (17), the "normal" starting doses can also vary by geographic regions and clinical practice.

TPMT recommendation. If starting doses are already high (e.g., 75 mg/m² of mercaptopurine), as is true in some ALL treatment regimens, lower than normal starting doses should be considered in TPMT intermediate metabolizers (11, 15, 24, 25) and markedly reduced doses (10-fold reduction) should be used in TPMT poor metabolizers (26) (**Table 2**). This approach has decreased the risk of acute toxicity without compromising relapse rate in ALL (27). Even at these markedly reduced dosages, erythrocyte TGN concentrations in TPMT poor metabolizers remain well above those tolerated and achieved by the majority of patients (who are TPMT normal metabolizers) (4, 26).

In some nonmalignant conditions, alternative agents may be chosen for TPMT intermediate or poor metabolizers rather than reduced doses of thiopurines; if thiopurines are used, full starting doses are recommended for TPMT normal metabolizers, reduced doses (30-80% of target dose) in TPMT intermediate metabolizers (28, 29), and substantially reduced doses (or use of an alternative agent) in TPMT poor metabolizers (**Table 2**) (4, 30).



Some of the clinical data upon which dosing recommendations are based (**Table 2**) rely on measures of TPMT phenotype rather than genotype; however, because *TPMT* genotype is strongly linked to TPMT phenotype (5-7, 31), these recommendations apply regardless of the method used to assess TPMT status.

NUDT15 recommendation. Similar to *TPMT*, tolerated mercaptopurine dosage is also correlated with the number of non-functional alleles of the *NUDT15* gene (17, 18). In fact, the degree of thiopurine intolerance (e.g., for mercaptopurine) is also largely comparable between carriers of *TPMT* vs. *NUDT15* decreased function alleles (17), although there remains a paucity of multi-ethnic studies examining both *TPMT* and *NUDT15* variants. Therefore, our *NUDT15* recommendations parallel those for *TPMT*. For NUDT15 normal metabolizers (*NUDT15*1/*1*), starting doses do not need to be altered. For NUDT15 intermediate metabolizers (e.g., *NUDT15*1/*3*, **Table 2**), reduced starting doses should be considered to minimize toxicity, particularly if the starting doses is high (e.g., 75 mg/m²/day for mercaptopurine). For NUDT15 poor metabolizers (e.g., *3/*3), substantially reduced doses (e.g., 10 mg/m²/day of mercaptopurine) or the use of an alternative agent should be used (**Table 2**) (20).

As for TPMT, there has been some variability in the tolerated thiopurine dosages within NUDT15 intermediate metabolizers, with a minority of individuals who do not seem to require significant dose reduction (17, 20). Therefore, genotype-guided prescribing recommendations apply primarily to starting doses; subsequent dosing adjustments should be made based on close monitoring of clinical myelosuppression (or disease-specific guidelines). In contrast, a full dose of mercaptopurine poses a severe risk of prolonged hematopoietic toxicity in NUDT15 poor metabolizers and preemptive dose reductions are strongly recommended (32, 33).

The NUDT15 poor metabolizer phenotype is observed at a frequency of about one in every 50 patients of East Asian descent, which is more common than the TPMT poor metabolizer phenotype in Europeans, and thus genotyping NUDT15 in the Asian populations may be of particular clinical importance. NUDT15 deficiency is also more prevalent in individuals of Hispanic ethnicity, particularly those with high levels of Native American genetic ancestry (17).

TPMT and NUDT15 recommendation. Figure 2 outlines the recommended course of action if both *TPMT* and *NUDT15* genotypes are known. There have been reports of patients with intermediate metabolizer status for both TPMT and NUDT15 (i.e., compound intermediate metabolizers), and there was a trend for a lower thiopurine tolerance in these individuals compared to intermediate metabolizers for only TPMT or NUDT15. The two genes are independent: the likelihood of an individual being an intermediate metabolizer for both genes depends upon the population frequencies for variant alleles. For example, given estimates of no function alleles for *NUDT15* of 11% and of no function alleles for *TPMT* of 2%, the frequency of the compound intermediate phenotype is estimated at 0.2%. However, the evidence for a different starting dosage recommendation for the compound intermediate metabolizers remains limited.

Recommendations for Incidental Findings

Not applicable.

Other considerations

If test results are available for only one gene (*TPMT* or *NUDT15*, but not both), prescribing recommendations based on that gene's results may be implemented, with the caveat that the other gene's results are missing and may have important implications. The higher frequency of decreased function *NUDT15* variants among individuals of Asian and Hispanic backgrounds and of TPMT variants in those with European and African backgrounds should be considered. In addition, there

may be other reasons underlying poor tolerance to thiopurines that are not related to *TPMT* or to *NUDT15* genetic variation.

Complementary clinical laboratory tests are available to measure thiopurine metabolites in erythrocytes: TGNs (for mercaptopurine, azathioprine, and thioguanine) and MeMPNs (or MeTIMP) for those on mercaptopurine or azathioprine (see **Supplement** for details on associations with TPMT). Erythrocyte TGNs or MeMPNs are not related to *NUDT15* genotypes (34-36) because clinical assays do not distinguish among the mono-, di-, and tri-phosphate forms of active TGNs, but there is evidence that intermediate and poor metabolizers for NUDT15 accumulate higher level of DNA-TG than normal metabolizers given the same mercaptopurine dosage (20). Thus, currently available erythrocyte therapeutic drug monitoring tests do not distinguish NUDT15 metabolizer phenotypes.

Implementation of this guideline. The guideline supplement contains resources that can be used within electronic health records (EHRs) to assist clinicians in applying genetic information to patient care for the purpose of drug therapy optimization (see *Resources to incorporate pharmacogenetics into an electronic health record with clinical decision support* sections of supplement).

POTENTIAL BENEFITS AND RISKS FOR THE PATIENT

The benefits of pre-emptive TPMT testing are that doses that are customized based on TPMT status reduce the likelihood of acute myelosuppression without compromising disease control (4, 8, 24, 25). The risks would be that a proportion of TPMT intermediate metabolizers may spend a period of time at lower thiopurine doses than they can eventually tolerate, because only ~30-60% of *TPMT* heterozygous patients receiving conventional thiopurine doses experience severe myelosuppression (4, 8, 11). However, because steady state is reached in 2-4 weeks, any period of "under-dosing" should be short, and using this approach, at least in ALL and in inflammatory bowel disease, outcomes were not compromised (4, 8, 24, 25, 28).

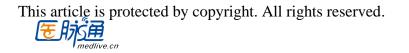
Similar benefits are expected with pre-emptive *NUDT15* genotyping, especially for Asian patients, given that these variants have comparable effects as risk alleles in *TPMT*. At least in ALL, leukemia cells with loss-of-function *NUDT15* alleles are also more sensitive to mercaptopurine (20) and thus in theory *NUDT15* genotyped guided dosing would not compromise anti-leukemic efficacy of this drug.

A possible risk to the patient is an error in genotyping (4). Some *TPMT* and/or *NUDT15* variants may not be included in the genotype test used and patients with these variants may be assigned a "wildtype" (*1) genotype by default. Thus, an assigned "wild-type" allele could potentially harbor a no or decreased function variant. Because genotypes are life-long test results, any such error could stay in the medical record for the life of the patient.

CAVEATS: APPROPRIATE USE AND/OR POTENTIAL MISUSE OF GENETIC TESTS

Most of the time, thiopurines are given orally daily for a period of at least several months. Genotypebased starting doses are just that—starting doses, and in most diseases, titration to the desired degree (or lack thereof) of myelosuppression is required. Thus, clinicians must continue to evaluate markers of disease progression and/or of myelosuppression to adjust thiopurine doses up or down from the genotype-directed starting doses. One caveat is that some serious long-term adverse effects (secondary tumors) have been associated with defective TPMT activity without necessarily causing serious acute myelosuppression; whether capping doses of thiopurines in those with a TPMT defect will decrease the risk of the late effect of secondary cancer is not known. Some adverse reactions to thiopurines, such as pancreatitis and hepatotoxicity, are not related to low TPMT activity.

The discovery and clinical implementation of *NUDT15* variants in thiopurine dosing is relatively recent and the exact impact of *NUDT15* genotype-guided dose adjustments on toxicity and efficacy are less clear compared to *TPMT*.



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Figure Legend

Figure 1: Metabolism of azathioprine, thioguanine, and mercaptopurine (41). Permission has been given by PharmGKB and Stanford to use figure (https://www.pharmgkb.org/pathway/PA2040). Pathway images and data are available under a Creative Commons BY-SA 4.0 license.

Figure 2. Recommended Starting Doses of Thiopurines by TPMT and NUDT15 phenotype

^a Whether a dose reduction is recommended from the starting dose depends on the level of the standard starting dose; for example, if the standard starting dose of mercaptopurine is 75 mg/m/day or higher, then a lower starting dose may be considered in intermediate metabolizers and would be recommended in poor metabolizers, whereas if the starting dose is 50 mg/m/day or lower, a reduced starting dose may not be necessary in intermediate metabolizers.

See Table 2 for recommendation.

b

For patients who are IM for both TPMT and NUDT15, further dose reduction might be needed compared to those who are only IM with respect to one gene (TPMT or NUDT15).

- Clinical Pharmacogenetics Implementation Consortium (CPIC) guideline for thiopurine dosing based on TPMT and NUDT15 genotypes: 2018 update
- CPIC Thiopurine Supplemental Tables

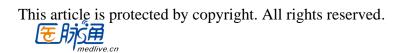


Table 1. Assignment of likely TPMT and NUDT15 phenotypes based on genotypes

Assignment of likely TPMT phenotypes based on genotypes

Likely Phenotype	Genotypes	Examples of diplotypes
Normal metabolizer	an individual carrying normal function alleles	*1/*1
Intermediate metabolizer	an individual carrying one normal function allele PLUS	*1/*2, *1/*3A, *1/*3B, *1/*3C,
	one no function allele	*1/*4
Possible Intermediate	an individual carrying one uncertain/unknown function	*2/*8, *3A/*7
metabolizer	allele PLUS one no function allele	
Poor metabolizer	an individual carrying two no function alleles	*3A/*3A, *2/*3A, *3A/*3C,
		*3C/*4, *2/*3C, *3A/*4
Indeterminate	An individual carrying two uncertain/unknown function	*6/*8
	alleles	
	OR	
	one normal function allele plus one uncertain allele	
	function allele	*1/*8
Assignment of likely NUD	T15 phenotypes based on genotypes	1

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an individual carrying two normal function alleles	*1/*1	
an individual carrying one normal function allele PLUS one no function allele	*1/*2, *1/*3	
an individual carrying one uncertain function allele PLUS one no function allele	*2/*5, *3/*6	
an individual carrying two no function alleles	*2/*2, *2/*3, *3/*3	
one normal function allele PLUS one uncertain function allele OR	*1/*4, *1/*5	
	an individual carrying one normal function allele PLUS one no function allele an individual carrying one uncertain function allele PLUS one no function allele an individual carrying two no function alleles one normal function allele PLUS one uncertain function allele an individual carrying two no function alleles an individual carrying two no function alleles	

¹ see TPMT and NUDT15 frequency table (21-23) for estimates of phenotype frequencies among different ethnic/geographic groups

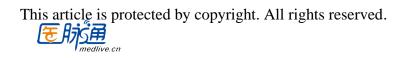


Table 2. Recommended Dosing of Thiopurines by TPMT phenotype

		Mercaptopurine		Azathiopri	ne	Thioguanine		
Phenotype	Implications	Dosing	Classificati	Dosing	Classific	Implications	Dosing	Classificat
	for	recommendations	on of	recommendations	ation of	for	recommendations for	ion of
	mercaptopurin	for mercaptopurine	recommen	for azathioprine	recomme	thioguanine	thioguanine	recommen
	e and		dations		ndations	phenotypic		dations
	azathioprine					measures		
	phenotypic							
	measures							
TPMT Normal	Lower	Start with normal	Strong	Start with normal	Strong	Lower	Start with normal	Strong
metabolizer	concentrations	starting dose ^a (e.g.		starting dose ^a (e.g.		concentrations	starting dose ^a (e.g.	
	of TGN	75 mg/m ² /day or		2-3 mg/kg/day)		of TGN	40-60 mg/m ² /day)	
	metabolites,	1.5 mg/kg/day) and		and adjust doses of		metabolites,	and adjust doses of	
	higher	adjust doses of		azathioprine based		but note that	thioguanine and of	
	MeTIMP, this	mercaptopurine		on disease-specific		TGN after	other	
	is the 'normal'	(and of any other		guidelines. Allow		thioguanine	myelosuppressive	

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	pattern	myelosuppressive		2 weeks to reach		are 5-10X	therapy without any	
		therapy) without		steady-state after		higher than	special emphasis on	
	Normal risk of	any special		each dose		TGN after	thioguanine. Allow 2	
	thiopurine-	emphasis on		adjustment (4, 30,		mercaptopurin	weeks to reach	
	related	mercaptopurine		37).		e or	steady-state after	
	leukopenia,	compared to other				azathioprine	each dose adjustment	
	neutropenia,	agents. Allow at					(4, 16).	
	myelosuppress	least 2 weeks to				Normal risk		
	ion	reach steady-state				of thiopurine-		
		after each dose				related		
		adjustment (4, 27,				leukopenia,		
		30).				neutropenia,		
						myelosuppres		
						sion		
ТРМТ	Moderate to	Start with reduced	Strong	Start with reduced	Strong	Moderate to	Start with reduced	Moderate
Intermediate	high	starting doses		starting doses		high	doses (50% to 80%	

metabolizer	concentrations	(30%-80% of	(30%-80% of	concentrations	of normal dose) if
OR	of TGN	normal dose) if	normal dose) if	of TGN	normal starting dose ^a
TPMT Possible	metabolites;	normal starting	normal starting	metabolites;	is \geq 40-60 mg/m ² /day
intermediate	low	$dose^a$ is ≥ 75	dose ^a is 2-3	but note that	(e.g. 20-48
metabolizer	concentrations	$mg/m^2/day \; or \geq 1.5$	mg/kg/day, (e.g.	TGN after	mg/m ² /day) and
	of MeTIMP	mg/kg/day (e.g.	0.6 - 2.4	thioguanine	adjust doses of
		start at 25-60	mg/kg/day), and	are 5-10X	thioguanine based on
	Increased risk	mg/m ² /day or 0.45-	adjust doses of	higher than	degree of
	of thiopurine-	1.2 mg/kg/day) and	azathioprine based	TGN after	myelosuppression
	related	adjust doses of	on degree of	mercaptopurin	and disease-specific
	leukopenia,	mercaptopurine	myelosuppression	e or	guidelines. Allow 2-
	neutropenia,	based on degree of	and disease-specific	azathioprine	4 weeks to reach
	myelosuppress	myelosuppression	guidelines. Allow		steady-state after
	ion	and disease-	2-4 weeks to reach	Increased risk	each dose
		specific guidelines.	steady-state after	of thiopurine-	adjustment. If
		Allow 2-4 weeks to	each dose	related	myelosuppression

	2	<u>u</u>	20
reach steady-state	adjustment (4, 30,	leukopenia,	occurs, and
after each dose	37, 38).	neutropenia,	depending on other
adjustment. If		myelosuppres	therapy, emphasis
myelosuppression		sion	should be on
occurs, and			reducing thioguanine
depending on other			over other agents (4,
therapy, emphasis			16).
should be on			
reducing			
mercaptopurine			
over other agents			
(4, 11, 15, 24, 25,			
27, 30, 38, 39).			
If normal starting			
dose is already			
<75mg/m2/day or			
	adjustment. If myelosuppression occurs, and depending on other therapy, emphasis should be on reducing mercaptopurine over other agents (4, 11, 15, 24, 25, 27, 30, 38, 39). If normal starting dose is already	after each dose37, 38).adjustment. If"""""""""""""""""""""""""""""""""	after each dose37, 38).neutropenia,adjustment. Ifmyelosuppressionsionoccurs, andin the sign of the si

		< 1.5mg/kg/day,						
		dose reduction may						
		not be						
		recommended.						
TPMT Poor	Extremely	For malignancy,	Strong	For non-malignant	Strong	Extremely	Start with drastically	Strong
metabolizer	high	start with		conditions,		high	reduced doses (16)	
	concentrations	drastically reduced		consider		concentrations	(reduce daily dose ^a	
	of TGN	doses (reduce daily		alternative non-		of TGN	by 10-fold and dose	
	metabolites;	dose ^a by 10-fold		thiopurine		metabolites;	thrice weekly instead	
	fatal toxicity	and reduce		immunosuppressan		fatal toxicity	of daily) and adjust	
	possible	frequency to thrice		t therapy.		possible	doses of thioguanine	
	without dose	weekly instead of				without dose	based on degree of	
	decrease; no	daily, e.g. 10		For malignancy,		decrease	myelosuppression	
	MeTIMP	mg/m ² /day given		start with			and disease-specific	
	metabolites	just 3 days/week)		drastically reduced		Greatly	guidelines. Allow 4-	
		and adjust doses of		doses (reduce daily		increased risk	6 weeks to reach	

0	Greatly	mercaptopurine	dose ^a by 10-fold	of thiopurine-	steady-state after
ir	ncreased risk	based on degree of	and dose thrice	related	each dose
0	of thiopurine-	myelosuppression	weekly instead of	leukopenia,	adjustment. If
re	elated	and disease-	daily) and adjust	neutropenia,	myelosuppression
16	eukopenia,	specific guidelines.	doses of	myelosuppres	occurs, emphasis
n	neutropenia,	Allow 4-6 weeks to	azathioprine based	sion	should be on
n	nyelosuppress	reach steady-state	on degree of		reducing thioguanine
ic	on	after each dose	myelosuppression		over other agents.
		adjustment. If	and disease-		For non-malignant
		myelosuppression	specific guidelines.		conditions, consider
		occurs, emphasis	Allow 4-6 weeks		alternative non-
		should be on	to reach steady-		thiopurine
		reducing	state after each		immunosuppressant
		mercaptopurine	dose adjustment		therapy (4).
		over other agents.	(28, 30, 37, 38,		
		For non-malignant	40).		

conditions,			
consider alternative			
non-thiopurine			
immunosuppressan			
t therapy (4, 26, 30,			
38).			

*Normal starting doses vary by race/ethnicity and treatment regimens. If standard dose is below normal recommended dose, dose reduction might not be recommended for intermediate metabolizers.

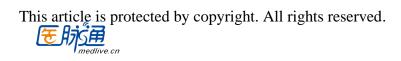


Table 3. Recommended Dosing of Thiopurines by 1	NUDT15 phenotype
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		Mercaptopurine		Azathioprine		Thioguanine	
Phenotype	Implications for	Dosing	Classification	Dosing	Classificatio	Dosing	Classificatio
	thiopurine	recommendations	of	recommendation	n of	recommendations	n of
	phenotypic	for	recommendat	s for	recommend	for thioguanine	recommendat
	measures	mercaptopurine	ions	azathioprine	ations		ions
NUDT15	Normal risk of	Start with normal	Strong	Start with	Strong	Start with normal	Strong
Normal	thiopurine-	starting dose ^a		normal starting		starting dose ^a (40-	
metabolizer	related	(e.g.,		dose ^a (e.g., 2-3		60 mg/day).	
	leukopenia,	75mg/m2/day or		mg/kg/day) and		Adjust doses of	
	neutropenia,	1.5mg/kg/day)		adjust doses of		thioguanine and	
	myelosuppressi	and adjust doses		azathioprine		of other	
	on	of mercaptopurine		based on		myelosuppressive	
		(and of any other		disease-specific		therapy without	
		myelosuppressive		guidelines.		any special	

		therapy) without		Allow 2 weeks		emphasis on	
		any special		to reach steady-		thioguanine.	
		emphasis on		state after each		Allow 2 weeks to	
		mercaptopurine		dose adjustment		reach steady-state	
		compared to other		(4, 30, 37).		after each dose	
		agents. Allow at				adjustment (4,	
		least 2 weeks to				16).	
		reach steady-state					
		after each dose					
		adjustment (4, 27,					
		30).					
NUDT15	Increased risk of	Start with reduced	Strong	Start with	Strong	Start with reduced	Moderate
Intermediate	thiopurine-	starting doses		reduced starting		doses (50% to	
metabolizer	related	(30%-80% of		doses (30%-80%		80% of normal	
OR	leukopenia,	normal dose) if		of normal dose)		dose) if normal	

Possible	neutropenia,	normal starting	if normal	starting dose ^a is
NUDT15	myelosuppressio	dose ^a is \geq 75	starting dose ^a is	≥40-60
Intermediate	n	$mg/m^2/day \ or \geq$	2-3 mg/kg/day,	mg/m ² /day (e.g.
metabolizer		1.5 mg/kg/day	(e.g. 0.6 – 2.4	20-48 mg/m ² /day)
		(e.g. start at 25-60	mg/kg/day), and	and adjust doses
		mg/m ² /day or	adjust doses of	of thioguanine
		0.45-1.2	azathioprine	based on degree
		mg/kg/day) and	based on degree	of
		adjust doses of	of	myelosuppression
		mercaptopurine	myelosuppressio	and disease-
		based on degree	n and disease-	specific
		of	specific	guidelines. Allow
		myelosuppression	guidelines.	2-4 weeks to
		and disease-	Allow 2-4 weeks	reach steady-state
		specific	to reach steady-	after each dose

myelosuppression
myelosuppression
occurs, and
depending on
other therapy,
emphasis should
be on reducing
thioguanine over
other agents (4,
16).

		If normal starting dose is already <75mg/m2/day or < 1.5mg/kg/day, dose reduction may not be recommended.				
NUDT15 Poor metabolizer		For malignancy, initiate dose at 10 mg/m ² /day and adjust dose based on myelosuppression	For non- malignant conditions, consider alternative non- thiopurine	Strong	Reduce doses to 25% of normal dose ^a and adjust doses of thioguanine based on degree of	Strong
	myelosuppressio n	and disease- specific	immunosuppres sant therapy.		myelosuppression and disease-	

guidelines. Allow		specific
4-6 weeks to	For malignant	guidelines. Allow
reach steady state	conditions, start	4-6 weeks to
after each dose	with drastically	reach steady-state
adjustment. If	reduced normal	after each dose
myelosuppression	daily doses ^a	adjustment. In
occurs, emphasis	(reduce daily	setting of
should be on	dose by 10-fold)	myelosuppression
reducing	and adjust doses	, emphasis should
mercaptopurine	of azathioprine	be on reducing
over other agents.	based on degree	thioguanine over
	of	other agents. For
For non-	myelosuppressi	non-malignant
malignant	on and disease-	conditions,
conditions,	specific	consider

consider	guidelines.	alternative non-
alternative non-	Allow 4-6	thiopurine
thiopurine	weeks to reach	immunosuppressa
immunosuppressa	steady-state	nt therapy (4).
nt therapy (4, 26,	after each dose	
30, 38).	adjustment (28,	
	30, 37, 38, 40).	

^aNormal starting doses vary by race/ethnicity and treatment regimens. If standard dose is below normal recommended dose, dose

reduction might not be recommended for intermediate metabolizers.

