NUDT15: thioguanine 7033/7034

ALL = acute lymphoblastic leukaemia, IM = intermediate metaboliser (e.g. *1/*2, *1/*3) (reduced NUDT15 enzyme activity), NM = normal metaboliser (*1/*1) (normal NUDT15 enzyme activity), NUDT15 = nudix hydroxylase 15 (enzyme involved in hydrolysis of diphosphate bonds and thus the conversion of 6-thio-deoxyguanosine triphosphate (6-thio-dGTP) into 6-deoxythioguanosine monophosphate (6-thio-dGMP)), PM = poor metaboliser (e.g. *2/*2, *2/*3, *3/*3) (absent or strongly reduced NUDT15 enzyme activity), 6-TGN = 6-thioguanine nucleotides (the active metabolites of azathioprine/6-mercaptopurine), 6-thio-dGTP = 6-thio-deoxyguanosine triphosphate (the fully activated metabolite of azathioprine/6-mercaptopurine), TPMT = thiopurine S-methyltransferase (enzyme involved in formation of inactive thiopurine metabolites).

Disclaimer: The Pharmacogenetics Working Group of the KNMP formulates the optimal recommendations for each phenotype group based on the available evidence. If this optimal recommendation cannot be followed due to practical restrictions, e.g. therapeutic drug monitoring or a lower dose is not available, the health care professional should consider the next best option.

Brief summary and justification of choices:
NUDT15 reverses the final step in the formation of the active metabolite of thioguanine. It converts 6-thiodeoxyguanosine triphosphate (6-thio-dGTP), which is incorporated into DNA, into 6-thiodeoxyguanosine monophosphate (6-thio-dGMP). For this reason, lower metabolic activity of NUDT15 leads to increased intracellular concentrations of the active metabolite 6-thio-dGTP. This increases the risk of adverse events such as myelosuppression.
There were no studies in humans confirming an increased thioguanine toxicity risk in patients with a genetically reduced NUDT15 enzyme activity (intermediate metabolisers (IM) and poor metabolisers (PM)). However, for azathioprine and mercaptopurine, which have the same active metabolite as thioguanine, 14 studies with more than 175 patients and 6 meta-analyses confirm IM and PM to have an increased risk for leukopenia and/or dose reduction due to adverse events like myelosuppression and infection. In addition, the thioguanine concentration killing 50% of acute lymphoblastic leukaemia blasts *in vitro* was significantly lower for cells derived from IM patients than for cells derived from NM patients (Moriyama T et al. NUDT15 polymorphisms alter thiopurine metabolism and hematopoietic toxicity. Nat Genet 2016;48:367-73. PubMed PMID: 26878724). For these reasons, the KNMP Pharmacogenetics Working Group decided that it was justified to extrapolate the data for azathioprine and 6-mercaptopurine to thioguanine and decided that therapy adjustment is required for the NUDT15-thioguanine interactions (yes/yes-interactions).
Therapeutic recommendations
Because *in vivo* data for thioguanine are lacking and extrapolation from the data on either azathioprine and 6-mercaptopurine or TPMT phenotypes was decided to be justified, the therapeutic recommendations for thioguanine were based on the azathioprine and 6-mercaptopurine and/or TPMT data.
Because there is no assay for determining 6-thio-dGTP levels in patient cells, only maximum tolerated doses can be used to calculate dose adjustments for patients with NUDT15 variants. Ten studies with more than 175 patients or with data for at least 2 PM and/or at least 25 IM (Maeda 2022, Mao 2021, Tanaka 2021, Wang 2021, Park 2019, Yi 2017, Kim 2017, Chao 2017, Liang 2016 and Yang 2015) and one meta-analysis (Yin 2017) report tolerated doses of azathioprine and/or 6-mercaptopurine (see the risk analysis for azathioprine and 6-mercaptopurine for full references and details). Calculations were only based on the ten studies because the meta-analysis also includes two of the studies. In addition, the meta-analysis does not specify the criteria for inclusion of studies for dose calculations and also included the studies of Moriyama 2016 and Yang 2014, which do not provide clear data on tolerated doses. Eight of the studies used for dose calculations concern acute lymphoblastic leukaemia (ALL) therapy (Mao 2021, Tanaka 2021, Wang 2021, Park 2019, Yi 2017, Kim 2017, Chao 2017, Liang 2016, and Yang 2015). The other two studies concern inflammatory bowel disease therapy, but provide only data for IM (Maeda 2022 and Chao 2017).

PM: The weighted mean of the calculated dose adjustment for 57 PM from 8 ALL studies is a dose reduction to 16% (8-48%, median 16%). In ALL therapy, 6-mercaptopurine and methotrexate dose are usually lowered alternately in case of leukopenia or other side effects. This indicates that using 16% of the normal 6-mercaptopurine dose in combination with normal methotrexate doses most probably will still result in leukopenia and the need for dose adjustment. For this reason, the KNMP Pharmacogenetics Working Group decided to

recommend starting with the lower range of the observed tolerated doses, i.e. 8% of the normal dose, which was rounded off to 10% to make application in clinical practice more feasible. This would make the dosing recommendation the same as for TPMT PM. This corresponds to the similar effect of variants of these two genes on the tolerated thiopurine dose as observed in two studies comparing NUDT15 and TPMT directly (Liang 2016 and Yang 2015).
Because the dose adjustment could not be calculated, but had to be extrapolated, the KNMP Pharmacogenetics Working Group decided to recommend adjusting the dose in second instance and choosing another drug in first instance.

IM: The weighted mean of the calculated dose adjustment for 497 IM from 10 studies is a dose reduction to 71% (52-88%, median 73%). In ALL therapy, 6-mercaptopurine and methotrexate dose are usually lowered alternately in case of leukopenia or other side effects. In addition, in one of the inflammatory bowel disease studies, the tolerated dose was only calculated in patients tolerating azathioprine or 6-mercaptopurine for more than 6 months (60% of the NM, 47% of the IM and none of the PM) (Chao 2017). This suggests that in this study, the patients most sensitive to thiopurines were not included in the calculation. This indicates that using 70% of the normal 6-mercaptopurine dose either in combination with normal methotrexate doses or in the more sensitive halve of IM patients most probably will still result in leukopenia and the need for dose adjustment. For this reason, the KNMP Pharmacogenetics Working Group decided to recommend starting with the lower range of the observed tolerated doses, i.e. 52% of the normal dose, which was rounded off to 50% to make application in clinical practice more feasible. This would make the dosing recommendation the same as for azathioprine and 6-mercaptopurine, that have the same active metabolite as thioguanine..
For oncolytics, toxicity and efficacy are strongly coupled, and it is unknown whether starting with a dose reduction based on genotype results in the same efficacy as reducing the dose based on toxicity. For this reason and because some TPMT IM tolerate the full thiopurine dose, the KNMP Pharmacogenetics Working Group recommendation for these patients is to either start with 50% of the normal thioguanine dose or to start with the normal dose and reduce to 50% when adverse events necessitate a dose reduction. In determining the starting dose, next to the IM phenotype, the physician needs to take into account the comorbidity (e.g. the sensitivity for infections), the patient wishes (taking into account the above mentioned uncertainty) and the estimation of the aggression of the tumour (e.g. based on tumour genetics).

You can find a detailed overview of the observed kinetic and clinical effects for azathioprine and mercaptopurine in the background information text of the corresponding gene-drug interactions in the KNMP Kennisbank. You might also have access to this background text via your pharmacy or physician electronic decision support system.

Recommendation concerning pre-emptive genotyping, including justification of choices:

The KNMP Pharmacogenetics Working Group considers genotyping before starting thioguanine to be essential for drug safety. Genotyping must be performed before drug therapy has been initiated to guide drug and dose selection.

Data in humans are lacking for thioguanine, but extrapolation of data from azathioprine and 6-mercaptopurine was considered to be justified (see Brief summary and justification of choices above). For this reason, for determination of the clinical implication score for thioguanine, the severity of the clinical effect, the evidence supporting this severity and the percentage of patients with leukopenia due to a NUDT15 variant were derived from azathioprine/6-mercaptopurine. This resulted in the clinical implication of the NUDT15-thioguanine interaction scoring 6 out of the maximum of 10 points for patients of European and African descend and 7 out of the maximum of 10 points for patients of Asian or (Latin-)American descend (with pre-emptive genotyping considered to be essential for scores ranging from 6 to 10 points):

The risk of serious life-threatening toxicity of azathioprine and 6-mercaptopurine (code E corresponding to grade 4) is increased for patients with a genotype resulting in diminished NUDT15 enzyme activity (IM and PM). This results in 1 out of the maximum of 2 points for the first criterion of the clinical implication score, the clinical effect associated with the gene-drug interaction (1 point for CTCAE grade 3 or 4).

The increased risk for serious toxicity (code D-E corresponding to grade 3-4) has been shown in 13 studies (Mao 2021, Tanaka 2021, Wang 2021, Park 2019, Zhu 2018, Yi 2018, Kim 2018, Chao 2017, Liang 2016, Zhu 2016, Moriyama 2016, Yang 2015, and Yang 2014) and 2 meta-analyses (Zhang 2018 and Yin 2017) on azathioprine and 6-mercaptopurine. This results in the maximum score of 3 points for the second criterion of the clinical implication score, the level of evidence supporting the associated clinical effect grade ≥ 3 (3 points for three or more publications with level of evidence score ≥ 3).

The number needed to genotype was deduced from the increase in the percentage of IM patients with azathioprine/6-mercaptopurine induced leukopenia. IM is the most prevalent variant genotype. For IM, an additional 23-77% of patients developed leukopenia compared to NM (Zhu 2018, Zhang 2018, and Chao 2017). The lower limit of 23% was used for the calculation. For Asians, Hispanics and Americans, the prevalence of IM has been reported to be 9.4-20.7%. This would amount to 2-5% of patients in these populations in which leukopenia is due to a NUDT15 variant and thus could have been prevented by reducing the risk to that in NM by lowering the dose, i.e. a number needed to genotype of 20-50. For Europeans and Africans, the prevalence of IM has been reported to be 0.6-1.5%. This would amount to 0.14-0.35% of patients developing leukopenia due to a NUDT15 variant in these

populations, i.e. a number needed to genotype of 300-700. The calculated number needed to genotype of 20-50 for Asians and (Latin-) Americans results in 2 out of the maximum of 3 points for the third criterion of the clinical implication score, the number needed to genotype (NNG) to prevent one clinical effect grade ≥ 3 (2 points for 10 < NNG ≤ 100). The calculated number needed to genotype of 300-700 for Europeans and Africans results in 1 out of the maximum of 3 points (1 point for 100 < NNG ≤ 1000).
The Summary of Product Characteristics (SmPC) indicates that patients with a variant NUDT15 gene have an increased risk for severe toxicity of thioguanine, such as early leukopenia and alopecia, at conventional doses of thiopurine therapy. This results in 1 out of the maximum of 2 points for the fourth and last criterion of the clinical implication score, the pharmacogenetics information in the SmPC (1 point for at least one genotype/phenotype mentioned in the SmPC, but not mentioned as a contra-indication and no recommendation to genotype).

The table below uses the KNMP nomenclature for NM, PM and IM. As a result, the definitions of NM, PM and IM in the table below can differ from the definitions used by the authors in the source.

Source	Code	Effect	Comments
ref. 1 SmPC Lanvis (thio-guanine) 08-06-18	0 IM: E PM: E	<u>Dosing</u>: Patients with an inherited mutated NUDT15 gene have an increased risk for severe toxicity of thioguanine. In general, these patients require a dose reduction, especially if they are homozygous for the NUDT15 variant. Genotypic testing for NUDT15 variants before starting thioguanine therapy can be considered. At least, careful monitoring of blood cell counts is necessary. <u>Warning</u>: Patients with an inherited mutated NUDT15 gene have an increased risk for severe toxicity of thioguanine, such as early leukopenia and alopecia, at conventional doses of thiopurine therapy. In general, these patients require a dose reduction, especially if they are homozygous for the NUDT15 variant. The frequency of NUDT15 c.415C>T has an ethnic variability of approximately 10% in East-Asians, 4% in Latin-Americans, 0.2% in Europeans and 0% in Afri-cans. At least, careful monitoring of blood cell counts is necessary. <u>Pharmacodynamics</u>: Recent studies indicate a strong association between the NUDT15 variant NUDT15 c.415C>T [p.Arg139Cys] (also known as NUDT15 R139C [rs116855232]), which is believed to result in loss of function of the NUDT15 enzyme, and thio-purine mediated toxicity, such as leukopenia and alopecia. Patients homozygous for the NUDT15 variant (NUDT15 T risk alleles) have a very high risk of thiopurine toxicity compared to patients homozygous for C. The frequency of NUDT15 c.415C>T has an ethnic variabi-lity with an increased risk in the Asian and Latin-American population. Genotypic analysis of the NUDT15 genotype should be performed. The prescribing physician is advised to deter-mine if decreasing the dose is necessary based on the genetic profile of the patient, eventually in combination with the profile of adverse events occurring during treatment. Patients with variants in both the NUDT15 and the TPMT enzyme have significantly less tolerance for thiopurines than patients with risk alleles of only one of these two genes. The exact mechanism of NUDT15 associated thiopurine-related toxicity is not clear.	
ref. 2 SmPC Tabloid (thio-guanine), USA, 23-05-18.	0	<u>Clinical pharmacology</u>: Several published studies indicate that patients with reduced TPMT or NUDT15 activity receiving usual doses of mercap-topurine, accumulate excessive cellular concentrations of	

ref. 2, continuation		active 6-TGNs, and are at higher risk for severe myelosuppression. In a study of 1028 children with ALL, the approximate tolerated mercaptopurine dosage range for patients with TPMT and/ or NUDT15 deficiency on mercaptopurine maintenance therapy (as a percentage of the planned dosage) was as follows: heterozygous for either TPMT or NUDT15, 50-90%; heterozygous for both TPMT and NUDT-15, 30-50%; homozygous for either TPMT or NUDT15, 5-10%. NUDT15 deficiency is detected in <1% of patients of European or African ancestry. Among patients of East Asian ancestry (i.e., Chinese, Japanese, Vietnamese), 2% have two loss-of-function alleles of the NUDT15 gene, and approximately 21% have one loss-of-function allele. The p.R139C variant of NUDT15 (present on the *2 and *3 alleles) is the most commonly observed, but other less common loss-of-function NUDT15 alleles have been observed. Warnings:	
	PM: E	Evaluate patients with repeated severe myelosuppression for thiopurine S-methyltransferase (TPMT) or nucleotide diphosphatase (NUDT15) deficiency. TPMT genotyping or phenotyping (red blood cell TPMT activity) and NUDT15 genotyping can identify patients who have reduced activity of these enzymes. Patients with homozygous TPMT or NUDT-15 deficiency require substantial dosage reductions. Bone marrow suppression could be exacerbated by co-administration with drugs that inhibit TPMT, such as olsalazine, mesalazine, or sulphasalazine. Precautions: Consider testing for TPMT and NUDT15 deficiency in patients who experience severe bone marrow toxicities or repeated episodes of myelosuppression. Dose: Patients with homozygous deficiency of either TPMT or NUDT15 enzyme typically require 10% or less of the standard thioguanine dosage. Reduce initial dosage in patients who are known to have homozygous TPMT or NUDT15 deficiency. Most patients with heterozygous TPMT or	
	IM: AA	NUDT15 deficiency tolerate recommended thioguanine doses, but some require dose reduction based on toxicities. Patients who are heterozygous for both TPMT and NUDT15 may require more substantial dosage reductions. Reduce the dosage based on tolerability.	

Risk group	TPMT IM or PM, use of TPMT inhibitors (aminosalicylates: mesalazine, olsalazine or sulphasalazine, furosemide, acetylsalicylic acid) or xanthine oxidase inhibitors (allopurinol, febuxostat), use of inhibitors of de novo purine synthesis (methotrexate). Note: results regarding the effect of the aminosalicylates are contradictory. Five studies clearly showed no in vivo drug interaction (Szumlanski CL et al. Sulphasalazine inhibition of thiopurine methyltransferase: possible mechanism for interaction with 6-mercaptopurine and azathioprine. Br J Clin Pharmacol 1995;39:456-9; Dewit O et al. Interaction between azathioprine and aminosalicylates: an in vivo study in patients with Crohn's disease. Aliment Pharmacol Ther 2002;16:79-85; Dilger K et al. Monitoring of thiopurine methyltransferase activity in postsurgical patients with Crohn's disease during 1 year of treatment with azathioprine or mesalazine. Ther Drug Monit 2007;29:1-5; de Graaff P et al. Influence of 5-aminosalicylic acid on 6-thioguanosine phosphate metabolite levels: a prospective study in patients under steady thiopurine therapy. Br J Pharmacol 2010;160: 1083-91; Reinisch W et al. Azathioprine versus mesalazine for prevention of postoperative clinical recurrence in patients with Crohn's disease with endoscopic recurrence: effi-

	cacy and safety results of a randomised, double-blind, double-dummy, multicentre trial. Gut 2010;59:752-9).

Comments:

- Other guidelines:
  - Relling MV et al. Clinical Pharmacogenetics Implementation Consortium guideline for thiopurine dosing based on TPMT and NUDT15 genotypes: 2018 update. Clin Pharmacol Ther 2019;105:1095-105. PMID: 30447069, and update guideline on the CPIC-site February 2019 (*9 function has been changed from "uncertain function" to "no function").
    CPIC defines NUDT15 IM and NUDT15 PM as we do (one or two no function alleles, respectively), but considers only *2, *3 and *9 to be no function alleles. CPIC considers the other alleles, including *4 through *8 to be alleles with uncertain function. CPIC groups combinations of one allele with uncertain function and one no function allele in the phenotype 'possible IM' instead of in the IM phenotype. In addition, CPIC groups combinations of two uncertain function alleles and combinations of one normal function and one uncertain function allele in the phenotype 'indeterminate'.
    CPIC indicates that thioguanine is mainly used for myeloid leukaemia. CPIC did not perform a literature review for thioguanine separately, but only for all thiopurines together. In addition, for thioguanine, CPIC only refers to the in vitro experiments using laboratory models in Moriyama 2016 indicating similar influence of NUDT15 on the cytotoxicity of azathioprine and thioguanine. So, the thioguanine recommendations are based on data on azathioprine/6-mercaptopurine.
    CPIC indicates that agnostic genome-wide association studies identified variants in NUDT15 that strongly influence thiopurine tolerance in patients with acute lymphoblastic leukaemia (ALL) and those with inflammatory bowel diseases (Yang 2015 and Yang 2014). In addition, CPIC indicates that patients carrying gene variant 415C>T (allele *2 or *3) showed excessive thioguanine incorporation into DNA and severe myelosuppression (Moriyama 2016), and that in children with ALL, patients homozygous for this variant 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 variant, respectively (Yang 2015).
    CPIC states that there is substantial evidence linking NUDT15 genotype with phenotypic variability. In addition, 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 (Yang 2014; Tanaka Y et al. Susceptibility to 6-MP toxicity conferred by a NUDT15 variant in Japanese children with acute lymphoblastic leukaemia. Br J Haematol 2015;171:109-15; Chiengthong K et al. NUDT15 c.415C>T increases risk of 6-mercaptopurine induced myelosuppression during maintenance therapy in children with acute lymphoblastic leukemia. Haematologica 2016;101:e24-6; Asada A et al. NUDT15 R139C-related thiopurine leukocytopenia is mediated by 6-thioguanine nucleotide-independent mechanism in Japanese patients with inflammatory bowel disease. J Gastroenterol 2016;51:22-9; Lee YJ et al. NUDT15 variant is the most common variant associated with thiopurine-induced early leukopenia and alopecia in Korean pediatric patients with Crohn's disease. Eur J Gastroenterol Hepatol 2016;28:475-8; Wong FC et al. NUDT15 variant and thiopurine-induced leukopenia in Hong Kong. Hong Kong Med J 2016;22:185-7; Kakuta Y et al. NUDT15 R139C causes thiopurine-induced early severe hair loss and leukopenia in Japanese patients with IBD. Pharmacogenomics J 2016;16:280-5; Ailing Z et al. Further evidence that a variant of the gene NUDT15 may be an important predictor of azathioprine-induced toxicity in Chinese subjects: a case report. J Clin Pharm Ther 2016;41:572-4; Zhu 2016; Soler, A.M. et al. TPMT and NUDT-15 genes are both related to mercaptopurine intolerance in acute lymphoblastic leukaemia patients from Uruguay. Br J Haematol 2018;181:252-5; Yin 2017; Shah SA et al. Nucleoside diphosphate-linked moiety X-type motif 15 C415T variant as a predictor for thiopurine-induced toxicity in Indian patients. J Gastroenterol Hepatol 2017;32:620-4; Tanaka Y et al. Interaction between NUDT15 and ABCC4 variants enhances intolerability of 6-mercaptopurine in Japanese patients with childhood acute lymphoblastic leukemia. Pharmacogenomics J 2018;18:275-80; Zhang 2018).
    CPIC indicates that tolerated mercaptopurine dose is correlated with the number of nonfunctional alleles of the NUDT15 gene (Yang 2015 and Yang 2014). CPIC states that, in fact, the degree of thiopurine intolerance (e.g., for mercaptopurine) is largely comparable between carriers of TPMT vs. NUDT15 decreased function alleles (Yang 2015), although there remains a paucity of multi-ethnic studies examining both TPMT and NUDT15 variants. Therefore, CPICs NUDT15 recommendations parallel those for TPMT.
    CPIC indicates that starting doses do not need to be altered for NUDT15 normal metabolisers (NUDT15 *1/*1), that reduced starting doses should be considered to minimise toxicity, particularly if the starting doses are high (e.g., 75 mg/m^2 per day for mercaptopurine) for NUDT15 intermediate metabolisers (e.g., NUDT15*1/*3), and that substantially reduced doses (e.g., 10 mg/m^2 per day of mercaptopurine) or the use of an alternative agent should be considered for NUDT15 poor metabolizers (e.g., NUDT15*3/*3) (Moriyama 2016).
    In addition, CPIC indicates that, as for TPMT, there is substantial variability in the tolerated thiopurine doses within NUDT15 intermediate metabolisers, with a minority of individuals who do not seem to

require significant dose reduction (Yang 2015, Moriyama 2016). Therefore, the genotype-guided prescribing recommendations apply primarily to starting doses; subsequent dose adjustments should be made based on close monitoring of clinical myelosuppression (or disease-specific guidelines). CPIC indicates that, in contrast, a full dose of mercaptopurine poses a severe risk of prolonged hematopoietic toxicity in NUDT15 poor metabolisers and pre-emptive dose reductions are strongly recommended (Zhu 2018 and Ailing Z et al. Further evidence that a variant of the gene NUDT15 may be an important predictor of azathioprine-induced toxicity in Chinese subjects: a case report. J Clin Pharm Ther 2016;41:572-4).
CPIC indicates that the NUDT15 poor metaboliser phenotype is observed at a frequency of about 1 in every 50 patients of East Asian descent, which is more common than the TPMT poor metaboliser phenotype in Europeans, and, thus, genotyping NUDT15 in the Asian populations may be of particular clinical importance. In addition, CPIC indicates that NUDT15 deficiency is also more prevalent in individuals of Hispanic ethnicity, particularly those with high levels of Native American genetic ancestry (Yang 2015).
For thioguanine, CPIC classifies the recommendation for PM as strong (i.e. "the evidence is high quality and the desirable effects clearly outweigh the undesirable effects") and the recommendation for IM and possible IM as moderate (i.e. "There is a close or uncertain balance as to whether the evidence is high quality and the desirable clearly outweigh the undesirable effects").
The therapeutic recommendations for thioguanine are indicated below:

Dosing recommendations for thioguanine by NUDT15 phenotype		
Phenotype	Therapeutic recommendation	Classification of recommendation
IM (one no function allele: *1/*2, *1/*3, or *1/*9) or possible IM (one allele with uncertain function (allele other than *1, *2, *3, or *9) and one no function allele: *2, *3, or *9)	Start with reduced doses (50-80% of normal dose) if normal starting dose[a] is ≥ 40-60 mg/m^2/day (e.g., 20-48 mg/m^2/day) and adjust doses of thioguanine based on degree of myelosuppression and disease-specific guidelines. Allow 2-4 weeks to reach steady-state after each dose adjustment. If myelosuppression occurs, and depending on other therapy, emphasis should be on reducing thioguanine over other agents[b].	Moderate[c]
PM (two no function alleles: *2/*2, *2/*3, *3/*3, *2/*9, *3/*9, or *9/*9)	Reduce doses to 25% of normal dose[a] and adjust doses of thioguanine based on degree of myelosuppression and disease-specific guidelines. Allow 4–6 weeks to reach steady-state after each dose adjustment. In setting of myelosuppression, emphasis should be on reducing thioguanine over other agents. For non-malignant conditions, consider alternative non-thiopurine immunosuppressant therapy[d].	Strong[e]

[a] 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 metabolisers.
[b] Ford LT et al. Thiopurine S-methyltransferase (TPMT) assessment prior to starting thiopurine drug treatment: a pharmacogenomic test whose time has come. J Clin Pathol 2010;63:288-95; McBride KL et al. Severe 6-thioguanine-induced marrow aplasia in a child with acute lymphoblastic leukemia and inhibited thiopurine methyltransferase deficiency. J Pediatr Hematol Oncol 2000;22:441-5.
[c] The classification moderate indicates that "there is a close or uncertain balance" as to whether the evidence is high quality and the desirable clearly outweigh the undesirable effects.
[d] Ford LT et al. Thiopurine S-methyltransferase (TPMT) assessment prior to starting thiopurine drug treatment: a pharmacogenomic test whose time has come. J Clin Pathol 2010;63:288-95.
[e] The classification strong indicates that the evidence is high quality and the desirable effects clearly outweigh the undesirable effects.

Recommendations for patients having also a TPMT variant
CPIC states that there have been reports of patients with intermediate metaboliser status for both TPMT and NUDT15 (i.e., compound intermediate metabolisers), and that there was a trend for a lower thiopurine tolerance in these individuals compared with intermediate metabolisers for only TPMT or NUDT15. However, CPIC indicates that the evidence for a different starting dose recommendation for the compound intermediate metabolisers remains limited.
The therapeutic recommendations for patients having also a TPMT variant are indicated below:

Dosing recommendations for thioguanine for patients with a genetically reduced activity for both TPMT and NUDT15		
NUDT15 phenotype	TPMT phenotype	Therapeutic recommendation

IM	IM	Consider dose reduction[a]. See TPMT IM and NUDT15 IM recommendation[b].
IM	PM	Dose reduction recommended[a]. See TPMT PM recommendation.
PM	IM	Dose reduction recommended[a]. See NUDT15 PM recommendation.
PM	PM	Dose reduction recommended[a]. See TPMT PM recommendation.

[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^2/day or higher, then a lower starting dose may be considered in intermediate metabolisers and would be recommended in poor metabolisers, whereas if the starting dose is 50 mg/m^2/day or lower, a reduced starting dose may not be necessary in intermediate metabolisers.

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

On 7-7-2023, there was not a more recent version of the recommendations present on the CPIC-site.

National Cancer Institute Common Toxicity Criteria (NCI-CTC)

	grade 1 = B	**grade 2 = C**	**grade 3 = D**	**grade 4 = E**	**grade 5 = F**
Diarrhoea	Increased stool frequency by < 4; slight increase in stoma output	Increased stool frequency by 4-6; moderate increase in stoma output; no effect on daily activities	Increased stool frequency by ≥ 7; incontinence; IV fluid ≥ 24 hours; hospitalisation; severe increase in stoma output; effect on daily activities	Life-threatening consequences (e.g. haemodynamic collapse)	Death
Neutropenia	> 1.5x10^9/L	< 1.5-1.0x10^9/L	< 1.0-0.5x10^9/L	< 0.5x10^9/L	Death
Leukopenia	> 3.0x10^9/L	< 3.0-2.0x10^9/L	< 2.0-1.0x10^9/L	< 1.0x10^9/L	Death
Thrombocytopenia	> 75x10^9/L	75-50x10^9/L	50-25x10^9/L	< 25x10^9/L	Death
Febrile neutropenia	-	-	Present	Life-threatening consequences (e.g. septic shock, hypotension, acidosis, necrosis)	Death

ULN = upper limit of normal

Date of literature search: 4 August 2023.

	Phenotype	**Code**	**Gene-drug interaction**	**Action**	**Date**
KNMP Pharmacogenetics Working Group decision	IM	0 E	yes	yes	25 September 2023
	PM	0 E	yes	yes	

Mechanism:

NUDT15 reverses the final step in the formation of the active metabolite of thioguanine. It converts 6-thiodeoxyguanosine triphosphate (6-thio-dGTP), which is incorporated into DNA, into 6-thiodeoxyguanosine monophosphate (6-thio-dGMP). For this reason, lower metabolic activity of NUDT15 leads to increased intracellular concentrations of the active metabolite 6-thio-dGTP. This increases the risk of adverse events such as myelosuppression.

Clinical Implication Score:

Table 1: Definitions of the available Clinical Implication Scores

Potentially beneficial	PGx testing for this gene-drug pair is potentially beneficial. Genotyping can be considered on an individual patient basis. If, however, the genotype is available, the DPWG recommends adhering to the gene-drug guideline	0-2 +
Beneficial	PGx testing for this gene-drug pair is beneficial. It is advised to consider genotyping the patient before (or directly after) drug therapy has been initiated to guide drug and dose selection	3-5 +
Essential	PGx testing for this gene-drug pair is essential for drug safety or efficacy. Genotyping must be performed before drug therapy has been initiated to guide drug and dose selection	6-10 +

Table 2: Criteria on which the attribution of Clinical Implication Score is based

Clinical Implication Score Criteria	**Possible Score**	**Given Score**	
		European, African	**Asian, (Latin-) American**
Clinical effect associated with gene-drug interaction (drug- or diminished efficacy-induced)			
• CTCAE Grade 3 or 4 (clinical effect score D or E)	+	+	+
• CTCAE Grade 5 (clinical effect score F)	++		
Level of evidence supporting the associated clinical effect grade ≥ 3			
• One study with level of evidence score ≥ 3	+		
• Two studies with level of evidence score ≥ 3	++		
• Three or more studies with level of evidence score ≥ 3	+++	+++	+++
Number needed to genotype (NNG) in the Dutch population to prevent one clinical effect grade ≥ 3			
• 100 < NNG ≤ 1000	+	+	
• 10 < NNG ≤ 100	++		++
• NNG ≤ 10	+++		
PGx information in the Summary of Product Characteristics (SmPC)			
• At least one genotype/phenotype mentioned	+	+	+
OR			
• Recommendation to genotype	++		
OR			
• At least one genotype/phenotype mentioned as a contra-indication in the corresponding section	++		
Total Score:	10+	6+	7+
Corresponding Clinical Implication Score:		Essential	Essential