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ARTICLE

Dutch pharmacogenetics working group guideline for the gene-drug interaction of *ABCG2*, *HLA-B* and Allopurinol, and *MTHFR*, folic acid and methotrexate

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The Dutch Pharmacogenetics Working Group (DPWG) aims to facilitate PGx implementation by developing evidence-based pharmacogenetics guidelines to optimize pharmacotherapy. This guideline describes the gene-drug interaction of *ABCG2* with allopurinol, *HLA-B* with allopurinol, *MTHFR* with folic acid, and *MTHFR* with methotrexate, relevant for the treatment of gout, cancer, and rheumatoid arthritis. A systematic review was performed based on which pharmacotherapeutic recommendations were developed. Allopurinol is less effective in patients with the *ABCG2* p.(Gln141Lys) variant. In *HLA-B*58:01* carriers, the risk of severe cutaneous adverse events associated with allopurinol is strongly increased. The DPWG recommends using a higher allopurinol dose in patients with the *ABCG2* p.(Gln141Lys) variant. For *HLA-B*58:01* positive patients the DPWG recommends choosing an alternative (for instance febuxostat). The DPWG indicates that another option would be to precede treatment with allopurinol tolerance induction. Genotyping of *ABCG2* in patients starting on allopurinol was judged to be 'potentially beneficial' for drug effectiveness, meaning genotyping can be considered on an individual patient basis. Genotyping for *HLA-B*58:01* in patients starting on allopurinol was judged to be 'beneficial' for drug safety, meaning it is advised to consider genotyping the patient before (or directly after) drug therapy has been initiated. For *MTHFR*-folic acid there is evidence for a gene-drug interaction, but there is insufficient evidence for a clinical effect that makes therapy adjustment useful. Finally, for *MTHFR*-methotrexate there is insufficient evidence for a gene-drug interaction.

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INTRODUCTION

The field of pharmacogenetics (PGx) studies the impact of heritable genetic variation on therapeutic effects and side effects of drugs. Germline genetic variations can predict phenotypic variations in drug response between patients which can be used to guide drug selection and (starting) dose. The goal is optimization of drug therapy and preventing adverse drug reactions, resulting in safer and more (cost-)effective pharmacotherapy. Over the past two decades this field has been receiving increasing attention and pharmacogenetics has started to be implemented in daily clinical practice. In 2005 the Dutch Pharmacogenetics Working Group (DPWG) was established by the Royal Dutch Pharmacists Association (KNMP). Its goals are to develop PGx informed therapeutic recommendations based on systematic literature review, and to assist physicians and pharmacists with integrating the recommendations into computerized systems for drug prescription, dispensing, and automated medication surveillance. This has led to risk analyses for 108 gene-drug combinations and 62 guidelines providing therapeutic recommendations for one or more aberrant phenotypes, an overview is available at the KNMP website [1]. Recently, the DPWG guidelines were endorsed by the European Association of Clinical Pharmacology and Therapeutics (EACPT) and the European Association of Hospital Pharmacists (EAHP) [2, 3]. Other initiatives such as the Clinical Pharmacogenetics Implementation Consortium (CPIC) were also established to promote implementation of

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PGx [4, 5]. Several DPWG guidelines have already been published [6–10]. This guideline, to be used in daily clinical practice, advises on four gene-drug interactions concerning drugs that are used in gout, cancer, and rheumatoid arthritis patients: *ABCG2* and allopurinol, *HLA-B* and allopurinol, *MTHFR* and folic acid, and *MTHFR* and methotrexate. This article describes the guideline developed by the DPWG and provides an overview of its pharmacotherapeutic recommendations. It also provides both the content required for enabling local PGx gene curation and for programming therapeutic recommendations into clinical decision support systems. We will first provide background information on the drugs, genes, and (if known) the mechanism that could give rise to a gene-drug interaction. Then the literature search (Supplementary tables 1 through 4) and pharmacotherapeutic recommendations provided by the guideline will be presented. The DPWG has additionally developed the clinical implication score, which is given to every gene-drug interaction requiring therapy adjustment. The objective of this score is to direct clinicians on whether or not to order relevant PGx genotyping tests before initiating therapy.

DRUGS

Allopurinol

Allopurinol is a purine analogue that lowers serum uric acid by inhibiting the enzyme xanthine oxidase (XO) which is a crucial enzyme in the purine metabolism pathway (see Fig. 1). Allopurinol is rapidly metabolized in vivo by XO and aldehyde-oxidase to its main active metabolite oxypurinol, which is responsible for most of the in vivo uric acid lowering effect. Allopurinol is generally safe and well tolerated although adverse effects do occur generally affecting the skin. Skin reactions range from relatively benign maculopapular rash to potentially fatal 'severe cutaneous adverse reactions' (SCAR) such as the Stevens-Johnson syndrome (SJS) and toxic epidermal necrolysis (TEN) [11]. Mortality rates reported in patients developing SCAR may be as high as 10–30% [12, 13]. The reported incidence of SCAR varies from 0.05 to 2.3% [11, 12, 14]. Incidence rates vary greatly between different populations and are also dependent on definitions used.

Methotrexate

Methotrexate is used in the treatment of a wide range of malignant and inflammatory diseases (e.g., rheumatoid arthritis, Crohn's disease, acute lymphatic leukemia, non-Hodgkin's lymphoma). Methotrexate is a folic acid antagonist, it inhibits multiple enzymes involved in nucleotide synthesis including dihydrofolate reductase, and thymidylate synthase (see Fig. 2) [15]. Methotrexate has numerous adverse effects especially in tissues with rapid cell turnover. Most common severe adverse effects are hepatotoxicity, pulmonary toxicity, myelosuppression, nephrotoxicity, mucositis, and increased risk of infection. Folate depletion is thought to be the cause of most of these side-effects, and therefore folate (folinic acid or folic acid) supplementation is often recommended during methotrexate therapy.

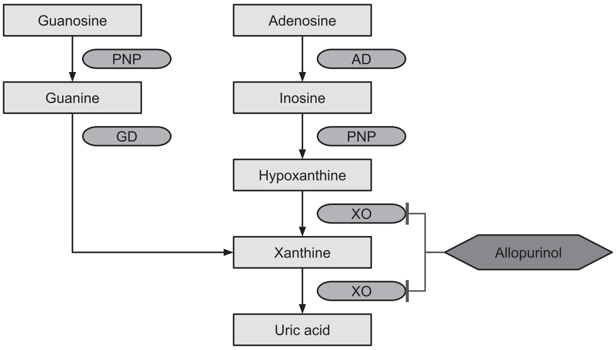

Fig. 1 Overview of purine metabolism and mechanism of action of allopurinol. Rectangles represent substances in purine metabolism pathway; ovals represent enzymes; lines originating from allopurinol represent enzyme inhibition by allopurinol; PNP purine nucleoside phosphorylase; AD adenosine deaminase; GD guanine deaminase; XO xanthine oxidase.

Folic acid

Folic acid is used in the treatment and prevention of several diseases. For example, it is used to reduce risk of methotrexate adverse effects, in the treatment of folate deficiency macrocytic anemia, and recommended in women wishing to become pregnant to prevent the development of neural tube defects [16].

GENES

ATP binding cassette transporter subfamily G member 2. The *ATP Binding Cassette Transporter Subfamily G Member 2 (ABCG2)* gene is located on chromosome 4q22.1 and transcription variant 1 contains 12 exons [17]. The gene encodes the ATP Binding Cassette Transporter Subfamily G Member 2, also known as Breast Cancer Resistance Protein. ABCG2 is an efflux transporter playing an important role in the excretion of uric acid in the kidneys and intestinal tract [18]. Interestingly oxypurinol has also been reported to be a substrate of ABCG2 [19]. Variants that are associated with reduced transporter activity, and likely reduced renal- and gastrointestinal excretion of uric acid, are associated with hyperuricemia and gout [18, 20]. Only the p.(Gln141Lys) variant appears to have a clinical effect. Reports suggest an association between *ABCG2* p.(Gln141Lys) and hyperuricemia and gout, resulting in a higher frequency of the p.(Gln141Lys) variant in hyperuricemic patients [21]. The full HGVS-nomenclature of this gene variant is indicated below (Table 1) with the genotypes and genotype groups distinguished for the three genes in this guideline. The *ABCG2* p.(Gln141Lys) variant is very common in people of East Asian origin (29-50%), common in people of European (9-17%), Latin American (14-22%) and South Asian origin (9%) and rare in people of African origin (2%) (see Supplementary Table 5a).

HLA-B. The *human leukocyte antigen (HLA)* genes, including *HLA-B*, are located on chromosome 6p21. Their function is to present peptides derived from proteins within the cell to mainly cytotoxic (CD8 +) T-cells. HLA proteins, especially HLA-B proteins, also have an essential role in the pathogenesis of delayed hypersensitivity reactions, such as SJS and TEN, to drugs [1]. The specific *HLA* allele involved in this hypersensitivity response is dependent on the drug. For allopurinol, the systematic literature review showed the *HLA-B*58:01* allele to be involved (see Conclusions from the body of evidence, later in this review). The *HLA-B*58:01* sequence is described in GenBank: EU499350.1 [22]. For the polymorphous *HLA* genes, no allele has been assigned as wild-type, so variants cannot be described in the HGVS-nomenclature. To generate an immune response, only very small amounts of HLA are required, predicting the absence of a gene-dose effect. For this reason, the DPWG does not distinguish between heterozygotes and homozygotes of *HLA* alleles (see Table 1). Prevalence of *HLA-B*58:01* carriers varies greatly between populations being as high as 5–31% in South-East Asians other than Japanese while it is 0–9% in Europeans (see Supplementary Table 5b).

Methylenetetrahydrofolate reductase. The *methylenetetrahydrofolate reductase* (*MTHFR*) gene is located on chromosome 1p36.22 and transcription variant 1 contains 12 exons [17]. The gene encodes the enzyme methylenetetrahydrofolate reductase. The *MTHFR* polymorphisms c.665 C > T and c.1286 A > C reduce enzyme activity. These polymorphisms are often referred to in literature as c.677 C > T and c.1298 A > C respectively. To avoid confusion among health care professionals, we use the

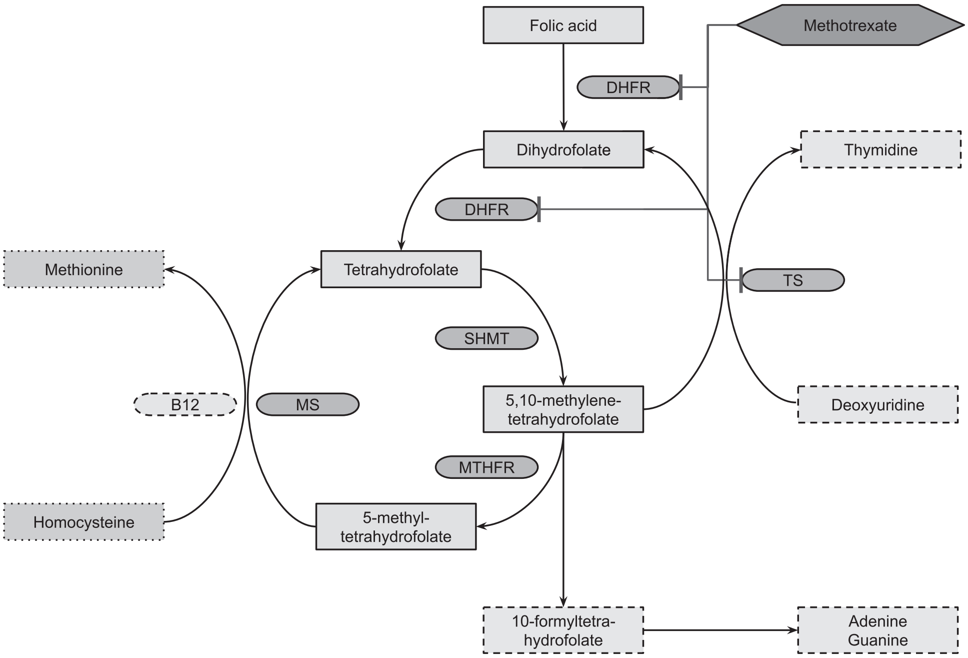

Fig. 2 Overview of folate metabolism and mechanism of action of methotrexate. Rectangles with continous borders represent substances in the folic acid cycle; rectangles with dotted borders represent substances in the methionine cycle; rectangles with dashed borders represent substances needed for DNA synthesis; ovals with continous borders represent enzymes; ovals with dashed borders represent cofactors; lines originating from methotrexate represent enzyme inhibition by methotrexate; B12 vitamin B12; DHFR dihydrofolate reductase; MS methionine synthase; SHMT serine hydroxymethyltransferase; TS thymidylate synthase.

Table 1. Distinguished genotypes and genotype groups for *ABCG2*, *HLA-B* and *MTHFR*.

Gene	Distinguished genotypes or genotype groups		Predicted phenotype
	Assigned name	Description	
ABCG2	*ABCG2* 141QQ (wild type)	No p.(Gln141Lys) variant (homozygous wild type)	Normal ABCG2 efflux transporter activity
	ABCG2 141QK	Heterozygous for the p.(Gln141Lys) variant	Reduced ABCG2 efflux transporter activity
	ABCG2 141KK	Homozygous for the p.(Gln141Lys) variant	Strongly reduced ABCG2 efflux transporter activity
HLA-B	*HLA-B*5801* negative	No *HLA-B*58:01* allele present	Low risk of allopurinol-induced severe cutaneous adverse events
	*HLA-B*5801* positive	Hetero- or homozygous for the *HLA-B*58:01* allele	Increased risk of allopurinol-induced severe cutaneous adverse events
MTHFR	*MTHFR* 677CC (wildtype)	No c.665 C > T variant (homozygous wild type)	Normal MTHFR enzyme activity
	MTHFR 677CT	Heterozygous for the c.665 C > T variant	Reduced MTHFR enzyme activity
	MTHFR 677TT	Homozygous for the c.665 C > T variant	Strongly reduced MTHFR enzyme activity

The gene variants in the table above and the *MTHFR* c.1286 A > C variant, that is not considered relevant, are characterised by the following sequence variations:
ABCG2 p.(Gln141Lys): rs-number: 2231142; NM_001257386.1:c.421 C > A; NP_001244315.1:p.Gln141Lys; NG_032067.2:g.105152 C > A;
*HLA-B*58:01:* The HLA-B*58:01 sequence is described in GenBank: EU499350.1
MTHFR c.665 C > T: rs-number: rs1801133; NM_005957.4:c.665 C > T; NP_005948.3:p.Ala222Val; NG_013351.1:g.14783 C > T
MTHFR c.1286 A > C: rs-number: rs1801131; NM_005957.4:c.1286 A > C; NP_005948.3:p.Glu429Ala; NG_013351.1:g.16685 A > C.

nomenclature from literature in the documents destined for health care professionals (abstracts of articles included in the systematic review and resulting pharmacist and physician texts). For c.665 C > T homozygous individuals enzyme activity was 30%, and for heterozygous individuals activity was 65% compared with wild-type [23]. The impact of c.1286 A > C is less severe with enzyme activity in homozygous individuals being 61% [24]. In addition, linkage disequilibrium has been observed between the two polymorphisms [25, 26]. Any clinical effects of c.1286 A > C may therefore not be independent of the effects of the c.665 C > T polymorphism. For these two reasons, the c.665 C > T polymorphism is more relevant than the c.1286 A > C polymorphism. The DPWG only considers c.665 C > T to be relevant (see Table 1). The full HVGS-nomenclature is included in the legend of Table 1 for c.665 C > T and c.1286 A > C. The frequency of the c.665 C > T variant is approximately 30% in Europeans and East-Asians. In South-Asians and Africans, it is lower with 15% and 11% respectively (see Supplementary Table 5c).

GENE-DRUG INTERACTION

ABCG2-allopurinol

Allopurinol is a uric acid lowering drug and *ABCG2* encodes an efflux transporter playing an important role in excretion of uric acid into the kidneys and intestinal tract. The active allopurinol metabolite oxypurinol also is a substrate of this efflux transporter. Therefore, the effect of gene variant *ABCG2* p.(Gln141Lys), resulting in reduced activity of the efflux transporter, is hard to predict. On the one hand, diminished excretion of oxypurinol would predict higher oxypurinol concentrations and thus a higher effectiveness of allopurinol in p.(Gln141Lys) carriers. On the other hand, it is likely that a stronger inhibition of uric acid production and thus a higher allopurinol dose is required in patients with a diminished uric acid excretion, like p.(Gln141Lys) carriers.

HLA-allopurinol

HLA proteins have an essential role in the pathogenesis of delayed hypersensitivity reactions that have also been observed for allopurinol. So, an analysis of experimental data is required to establish the *HLA* allele or alleles involved.

MTHFR-methotrexate

Methotrexate inhibits dihydrofolate reductase, which converts dihydrofolate to tetrahydrofolate. The toxicity of methotrexate can be reduced by administration of the tetrahydrofolate precursors folic acid or leucovorin [27]. The enzyme MTHFR converts 5,10-methylenetetrahydrofolate to 5-methyltetrahydrofolate, which can in turn be converted to tetrahydrofolate. For this reason, the *MTHFR* c.665 C > T gene variant, that results in reduced MTHFR enzyme activity, should decrease intracellular tetrahydrofolate concentrations, which might increase the effectiveness and/or toxicity of methotrexate.

MTHFR-folic acid

Folic acid is converted to tetrahydrofolate by the enzyme dihydrofolate reductase. The enzyme MTHFR converts 5,10-methylenetetrahydrofolate to 5-methyltetrahydrofolate, which can in turn be converted to tetrahydrofolate. For this reason, the *MTHFR* c.665 C > T gene variant, that results in reduced MTHFR enzyme activity, might decrease intracellular tetrahydrofolate concentrations, and increase the folic acid requirement and counteract folic acid supplementation.

SUPPORTING BODY OF EVIDENCE

The methods used for literature search, assessment, and therapeutic recommendations have previously been described [7, 8]. Briefly, a scientist of the KNMP (MN) performed a systematic review. After literature search relevant articles meeting in- and exclusion criteria were selected and summarized. Based on these summaries therapeutic recommendations were made for clinical practice when a significant gene-drug interaction was found. The performed literature searches with search strings and in- and exclusion criteria can be found in supplementary material 1. All included articles were scored for quality of evidence and clinical impact of the interaction, using the method previously described [8]. In brief, for quality of evidence a five-point scale was used with 0 being the lowest possible quality, and 4 being the highest possible quality (e.g., high quality meta-analysis or study). Clinical impact was scored on a seven-point scale ranging from AA$^{\#}$ (positive effect) to F (highest negative effect). This clinical impact scale (AA$^{\#}$-F) runs parallel to the Common Terminology Criteria for Adverse Events (CTCAE); where CTCAE grade 5 severity is equal to clinical relevance score F (death) and CTCAE grade 1 severity is equal to clinical relevance score B. The clinical relevance score additionally includes the scores AA$^{\#}$, AA, and A. These regard “positive clinical effect”, “no clinical or kinetic effect”, and “significant kinetic effect or not clinically relevant effect”, respectively. The summaries of articles, and their respective scores, reviewed to devise this guideline can be found in supplementary table 1, 2, 3, and 4. The summaries of each article and their respective scores were checked by two independent DPWG members. All summaries and scores were discussed with all members of the DPWG in a meeting. If scores differed, consensus was reached within this meeting and a score was agreed upon.

GENERAL CONCLUSION OF EVIDENCE

ABCG2-allopurinol

Nine articles were included in the systematic review. The summaries of included articles can be found in supplementary table 1, and a detailed overview of observed clinical effects in supplementary table 6. Five of the eight studies and a case-report showed a decreased effectiveness of allopurinol in p.(Gln141Lys) carriers. For example, in one study the percentage of patients with poor response to allopurinol (defined as serum uric acid ≥0.36 mmol/L despite allopurinol >300 mg/day) was 2.0 and 2.8-fold higher compared with wildtype for heterozygotes and homozygotes respectively. In another study the decrease in plasma uric acid for each 100 mg increase in allopurinol dose was 0.8 and 0.4-fold lower compared with for wildtype for heterozygotes and homozygotes respectively. Because of the decreased effectiveness, the DPWG decided that there is a gene-drug interaction.

No other *ABCG2* variants were shown to have a clinically relevant effect on allopurinol therapy.

HLA-allopurinol

Twelve articles and the Summary of Product Characteristics were included in the systematic review. The summaries of included articles can be found in supplementary table 2, and a detailed overview of observed clinical effects in supplementary table 7. *HLA-B*58:01* The included meta-analyses showed that this allele strongly increased the risk for allopurinol-induced SJS/TEN (OR = 84–151), all SCAR (OR = 73–165), and DRESS (OR = 54) both in Asian and European subgroups. In addition, three studies in East-Asians showed that excluding *HLA-B*58:01* positive patients from therapy with allopurinol or starting with an allopurinol tolerance induction protocol for these patients, resulted in reduction of the incidence of allopurinol-induced severe cutaneous adverse events from 0.3% (non-selected patients), 0.9% or 2.0% (patients with chronic renal insufficiency) to 0%. The DPWG concluded that there is a gene-drug interaction.

Other HLA variants. No other *HLA* variants were shown to have a clinically relevant effect on allopurinol therapy.

MTHFR-methotrexate

Sixteen meta-analyses were included. The summaries of included articles can be found in supplementary table 3, and a detailed overview of observed clinical effects in supplementary table 8.

Gene variant c.665C>T. All 6 meta-analyses (5 in patients with rheumatoid arthritis and 1 with haematologic malignancies) investigating the effectiveness of methotrexate found no association with the c.665 C > T gene variant. The 13 meta-analyses investigating adverse events (7 in patients with rheumatoid arthritis, 5 with cancer, and 1 with different indications) did not show consistent results. Four did not show an association, while the other 9 meta-analyses only showed an association in certain subgroups. Between these subgroups there was little consistency across meta-analyses. There are indications for a stronger effect of the c.665 C > T gene variant on adverse events in case of folate supplementation, and on serious adverse events. However, current evidence for these subgroups is also insufficient.

For these reasons, the DPWG decided that there is insufficient evidence for a gene-drug interaction.

Gene variant c.1286A>C. The c.1286 A > C gene variant results in an enzyme in which the activity is less severely reduced than for the c.665 C > T gene variant. As expected, there is not much evidence for an association of this gene variant with the effectiveness or adverse events of methotrexate therapy (see Supplementary table 3).

For this reason, the DPWG decided that there is insufficient evidence for a gene-drug interaction.

***MTHFR*-folic acid**

Ten articles were included. The summaries of included articles can be found in Supplementary table 4, and a detailed overview of observed clinical effects in supplementary table 9.

Gene variant c.665C > T. Studies showed that the effect of therapy with folic acid either was not changed (1 meta-analysis on homocysteine levels, and 1 study on the risk of stroke in hypertensive patients) or was increased (1 meta-analysis on homocysteine and folate levels, and 1 study on the risk of stroke in hypertensive patients) in patients with the c.665 C > T variant that results in reduced MTHFR activity. In the meta-analysis that showed increased effectiveness, the decrease in total homocysteine concentration during folic acid supplementation was 3.3 μM (95% CI: 2.7–3.8) in the homozygous group versus 1.0 μM (95% CI: 0.8–1.2) in the wildtype group, though total homocysteine concentration after folic acid treatment was still higher in the homozygous group (14.1 μM vs 12.1 μM). Patients with the c.665 C > T variant had lower baseline folate concentrations and higher baseline homocysteine concentrations than patients without this variant. For example, in one study baseline folate concentrations were 0.88-fold lower and baseline homocysteine concentrations 1.7-fold higher for homozygous patients compared with wildtype. Folic acid therapy partially corrected for this. Although this correction is only partial, there are no indications for adverse clinical effects, like an increased incidence of neural tube defects, for patients with the c.665 C > T variant after folic acid treatment.

For this reason, the DPWG concluded that there is a *MTHFR*-folic acid interaction, but that there is insufficient evidence for a clinical effect that makes therapy adjustment useful.

Gene variant c.1286A > C. One study showed a decreased risk of treatment failure (OR = 0.26 for homozygous and OR = 0.52 for heterozygous individuals compared with wildtype) with folic acid for hyperhomocysteinaemia in c.1286 A > C carriers (see Supplementary table 4). However, the direction of this effect was opposite to the increased risk found for c.665 C > T carriers, and the reduction in risk for treatment failure was similar for alleles without the c.665 C > T variant, that either had or did not have the c.1286 A > C variant. So, the observed association was likely due to the strong linkage disequilibrium between c.665 C > T and c.1286 A > C, resulting in 99.5% of alleles with a c.665 C > T variant having no c.1286 A > C variant.

For this reason, the DPWG decided that there was not enough evidence for a c.1286 A > C - folic acid interaction.

Other MTHFR variants. No other *MTHFR* variant with a clinically relevant effect on folic acid therapy was found.

PHARMACOTHERAPEUTIC RECOMMENDATIONS

An overview of the (presence of) pharmacotherapeutic recommendations is presented in Table 2. Detailed justifications of choices are included in supplementary tables 6 through 9.

Table 2. Pharmacotherapeutic recommendations (if present).

Gene	Drug	Distinguished genotypes or genotype groups		Pharmacotherapeutic recommendation[a]
		Assigned name	Description	
ABCG2	allopurinol	*ABCG2* 141QK	p.(Gln141Lys) heterozygote	Use 1.25 times the standard dose. This equates to a dose titration schedule of 100, 200, 400, and 500 mg/day instead of the usual schedule of 100, 200, 300, and 400 mg/day.
		ABCG2 141KK	p.(Gln141Lys) homozygote	Use 1.4 times the standard dose. This equates to a dose titration schedule of 100, 300, 400, 600, and 700 mg/day instead of the usual schedule of 100, 200, 300, 400, and 500 mg/day.
HLA-B	allopurinol	*HLA-B*5801* positive	*58:01 carrier	Choose an alternative, such as febuxostat. Another option[b] is to induce allopurinol tolerance first: To induce allopurinol tolerance, the allopurinol dose is increased every 3 days until a dose of 100 mg/day has been achieved on Day 28. The consecutive daily doses in the induction protocol are 50 μg, 100 μg, 200 μg, 500 μg, 1 mg, 5 mg, 10 mg, 25 mg, 50 mg and 100 mg.
MTHFR	methotrexate	*MTHFR* 677CT	c.665 C > T heterozygote	-
		MTHFR 677TT	c.665 C > T homozygote	-
MTHFR	folic acid	*MTHFR* 677CT	c.665 C > T heterozygote	-
		MTHFR 677TT	c.665 C > T homozygote	-

[a] No pharmacotherapeutic recommendation: therapy adjustment is not required or beneficial for this genotype-drug combinations.

[b] The applicability of this option is hampered by the unavailability of commercial (very) low dose formulations of allopurinol.

ABCG2-allopurinol

The DPWG recommends using a higher allopurinol dose in patients with the p.(Gln141Lys) variant. Only one study provided data on the required allopurinol dose for the different p.(Gln141Lys) genotypes. In this study in gout patients the allopurinol dose was increased until the serum uric acid was below 0.36 mmol/l. The required increase in allopurinol dose mentioned in the recommendation was derived from this study (a 1.25-fold higher dose for p.(Gln141Lys) heterozygotes and a 1.4-fold higher dose for p.(Gln141Lys) homozygotes). Articles referred to in this section can be found in supplementary table 6.

*HLA-B*58:01*-allopurinol

For the whole group of *HLA-B*58:01* carriers, the positive predictive value for development of allopurinol-induced severe cutaneous adverse events was 1.6% to 2.0%. For patients with chronic kidney insufficiency this was 8–18%. Because of the relatively high positive predictive values and the relatively high fatality of allopurinol-induced severe cutaneous adverse events of 11%, the DPWG recommends choosing an alternative. The DPWG decided to mention preceding treatment with allopurinol tolerance induction as another option, despite realising that application of this option is hampered by the unavailability of commercial (very) low dose formulations of allopurinol. Articles referred to in this section can be found in supplementary table 7.

Supplementary Table 10 through 13 present an overview of suggested pop-up or look-up texts for electronic prescribing systems for pharmacists and physicians. These can be used to program alerts into the clinical decision support system (CDSS). Complete genotype to distinguished genotype/genotype group translation tables, which can be used to programme the translation of genotype results into the distinguished genotypes/genotype groups in laboratory information systems, can be found in Supplementary Table 14a through c. The guidelines and background information are available on KNMP.nl [1] and will be available on PharmGKB.org.

IMPLICATIONS FOR CLINICAL PRACTICE

To assist the clinician in deciding on whether to order PGx before initiating a new therapy, the DPWG has developed the clinical implementation score. The pre-emptive PGx results for a certain drug–gene pair can be scored as: essential, beneficial, or potentially beneficial (for a more detailed explanation of these terms see supplementary table 15a). The development of these categories and the systematic scoring criteria have been discussed previously [28]. In brief, the implications for clinical practice are based on a list of four criteria regarding the following: the clinical effect associated with the gene-drug interaction, the level of evidence supporting the clinical effect, the effectiveness of the intervention in preventing the clinical effect (which includes the number needed to genotype), and the PGx information included in the drug-label. A clinical implementation score is only provided for gene-drug interactions with a therapeutic recommendation. If a therapeutic recommendation is lacking, pre-emptive genotyping provides no benefit.

ABCG2-allopurinol

The DPWG considers genotyping of *ABCG2* before starting allopurinol to be 'potentially beneficial' for drug effectiveness. Genotyping can be considered on an individual patient basis. For details on how this score is established see supplementary table 15b.

*HLA-B*58:01*-allopurinol

Based on the clinical implication score genotyping was scored as 'essential' in all patient groups. However, in many patients with the *HLA-B*58:01* variant there is no equivalent alternative for allopurinol. Furthermore, because the positive predictive value is far below 50%, a majority of patients will falsely be denied allopurinol (i.e. these patients will not receive the preferred treatment for gout). For these reasons the DPWG decided that genotyping for *HLA-B*58:01* in patients planned to be started on allopurinol is not 'essential' and downgraded the recommendation to 'beneficial'. It is advised to consider genotyping the patient before (or directly after) drug therapy has been initiated to guide drug and dose selection. For details on how these scores are established see Supplementary table 15c.

DIFFERENCES BETWEEN AVAILABLE GUIDELINES

To the best of our knowledge no guidelines are available for the *ABCG2*-allopurinol, *MTHFR*-folic acid, and *MTHFR*-methotrexate gene-drug combinations. Guidelines are available on the *HLA-B*58:01*-allopurinol gene variant-drug interaction; these are described in more detail below.

Therapeutic recommendations

The CPIC gives the same therapeutic recommendations as the DPWG. The CPIC indicates that, given the strong association of *HLA-B*58:01* with allopurinol-induced SCAR, allopurinol is contraindicated in patients who have tested positive for *HLA-B*58:01*. Alternative medication should be given to these patients to avoid the risk of developing SCAR. CPIC classifies this recommendation as strong [29].

The European Medicine Agency CFMPFHU, Pharmacovigilance Working Party mentioned in their 2012 report that, if the patient is a known carrier of *HLA-B*58:01*, the use of allopurinol may be considered if the benefits are thought to exceed risks. Extra vigilance for signs of hypersensitivity syndrome or SJS/TEN is required and the patient should be informed of the need to stop treatment immediately at the first appearance of symptoms [30]. The lack of suitable alternative therapies to allopurinol is mentioned as the rationale for this recommendation.

The American college of Rheumatology in their guidelines for management of gout recommends an alternative to allopurinol in individuals of Korean descent with stage 3 or worse chronic kidney disease, or of Han Chinese or Thai extraction positive for *HLA-B*58:01* [31]. The rationale is that they consider only carriers from these groups to have a high-risk for allopurinol-induced SCAR.

Genotyping recommendations

The European Medicine Agency CFMPFHU, Pharmacovigilance Working Party mentioned in their 2012 report that the sensitivity of prior testing for *HLA-B*58:01* may be as low as 50% in European populations. This suggests that potentially half of European patients that do develop SCAR will not be identified by prior testing. Therefore, they made the following recommendations: The use of genotyping as a screening tool to make decisions about treatment with allopurinol has not been established. Secondly, routine testing for *HLA-B*58:01* is not recommended in any patient [30].

The American college of Rheumatology in their guidelines for management of gout recommended to consider *HLA-B*58:01* screening in selected patients specifically in subpopulations at higher risk for severe allopurinol hypersensitivity reaction (e.g., Koreans with stage 3 or worse chronic kidney disease, and Han Chinese and Thai irrespective of renal function) [31].

Disclaimer

The Pharmacogenetics Working Group of the KNMP (DPWG) 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, then the health care professional should consider the next best option.

DATA AVAILABILITY

All data and material are either included in the supplementary information or publicly available (i.e., the published articles, PubMed). The guidelines and background information are available on KNMP.nl [1] and will be available on PharmGKB.org.

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AUTHOR CONTRIBUTIONS

KP drafted the manuscript and contributed to interpretation of results. EH and GR supervised drafting of the manuscript and contributed to conceiving the work and interpretation of the results. MN contributed to conceiving the work and interpretation of the results, and performed the data extraction. BS drafted and published English versions of clinical decision support texts. NBV, AB, HG, AR, RS, JS, DT, JW, RW, and VD contributed to conceiving the work and interpretation of the results. In addition, all authors revised the manuscript and approved the final version as well as agreed to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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COMPETING INTERESTS

The authors declare no competing interests.

ETHICAL APPROVAL

Ethical approval was not required as no individual patient data was used for this article. Data was extracted from other publications and cannot be traced to any individual patient.

ADDITIONAL INFORMATION

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