CYP2D6: nortriptyline 1557/1558/1559

AUC = area under the concentration-time curve, Cl_{or} = oral clearance, C_{ss} = plasma concentration in steady state, EM = extensive metaboliser (gene dose 1.5-2.5) (normal CYP2D6 enzyme activity), HNT = 10-hydroxynortriptyline, IM = intermediate metaboliser (gene dose 0,5-1) (decreased CYP2D6 enzyme activity), MR = metabolic ratio, NS = non-significant, NT = nortriptyline, PM = poor metaboliser (gene dose 0) (absent CYP2D6 enzyme activity), S = significant, SmPC = Summary of Product Characteristics, $t_{1/2}$ = half-life, TCA = tricyclic antidepressant, TDM = therapeutic drug monitoring, UM = ultra-rapid metaboliser (gene dose ≥ 3) (increased CYP2D6 enzyme activity)

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:
Nortriptyline is mainly metabolised by CYP2D6 to the active metabolite E-10-hydroxynortriptyline. This metabolite is approximately half as potent as nortriptyline itself, but the therapeutic range of nortriptyline is only based on the nortriptyline concentration (50-150 ng/ml).
Nortriptyline is converted by CYP2D6 and CYP2C19 to the inactive metabolite desmethylnortriptyline.
Genetic variants in CYP2D6 can result in a decreased CYP2D6 enzyme activity (intermediate metabolisers (IM)), an absent CYP2D6 enzyme activity (poor metabolisers (PM)) or an increased CYP2D6 enzyme activity (ultra-rapid metabolisers (UM)).
Kinetic studies showed differences in nortriptyline exposure for patients with CYP2D6 gene variants (Hodgson 2014, Lee 2006, Murphy 2001, Morita 2000, Yue 1998, Dalen 1998 and Dahl 1996). Case reports suggest an increased risk of toxic plasma concentrations and adverse events in PM and IM (Lee 2004, Chen 1996 and Bertilsson 1981). However this could not be confirmed in a study with 20 IM and 20 PM (Hodgson 2015), a study with 10 IM and 3 PM (Berm 2016) and a study with 4 PM (Roberts 2004). A case report suggests an increased risk of subtherapeutic plasma concentrations and ineffectiveness in UM (Bertilsson 1993). However, this could not be confirmed in a study with 11 UM. Because nortriptyline has a narrow therapeutic range, changes in exposure are likely to have therapeutic consequences. For this reason and despite the contradictory evidence from the literature, the KNMP Pharmacogenetics Working Group decides that a gene-drug interaction is present and that dose adjustments are required for PM, IM and UM (yes/yes-interactions).
Justification of recommendations per CYP2D6 phenotype
Dose adjustments were calculated on the basis of the AUC or Css for nortriptyline.
PM: The weighted mean of the calculated dose adjustment is a dose reduction to 35% of the standard dose (30%-41%; median 38%). This was translated to 40% to be more achievable in clinical practice. Effectiveness and side effects and/or the plasma concentration should be monitored when performing dose adjustments.
IM: The weighted mean of the calculated dose adjustment is a dose reduction to 58% of the standard dose (36%-74%; median 54%). This was translated to 60% to be more achievable in clinical practice. Effectiveness and side effects and/or the plasma concentration should be monitored when performing dose adjustments.
UM: The weighted mean of the calculated dose adjustment is a dose increase to 174% of the standard dose (130%-185%; median 170%). This was translated to 170% to be more achievable in clinical practice. Effectiveness and side effects and/or the plasma concentration should be monitored when performing dose adjustments. As the adjustment of UMs is difficult [Ther Drug Monit 1985;7:478-80] and the cardiotoxic metabolite can accumulate [Ther Drug Monit 1985;7:478-80], the recommendation is to choose a different antidepressant that is not metabolised by CYP2D6 if a dose increase is unwanted due to the cardiotoxic metabolites or if the dose increase does not give the desired results.
Note: The dose calculations do not take into consideration the active metabolite E-10-hydroxynortriptyline. The reason for this is that the effectiveness determination is normally performed based on nortriptyline alone. If this metabolite is taken into consideration, then the calculated dose adjustment is smaller.
You can find an overview of the observed kinetic and clinical consequences per phenotype in the background information text of the gene-drug interactions on the KNMP Kennisbank. You might also have access to this background information text via your pharmacy or physician electronic decision support system.

Recommendation concerning pre-emptive genotyping, including justification of choices:
The Dutch Pharmacogenetics Working Group considers genotyping before starting nortriptyline to be potentially beneficial for the prevention of side effects and for drug efficacy. Genotyping can be considered on an individual patient basis. If, however, the genotype is available, the Dutch Pharmacogenetics Working Group recommends adhering to the gene-drug guideline.
The clinical implication of the gene-drug interaction scores 0 out of the maximum of 10 points (with pre-emptive genotyping considered to be potentially beneficial for scores ranging from 0 to 2 points) (see also the clinical implication score tables at the end of this risk analysis):
No severe clinical effects were observed in users of nortriptyline with a variant phenotype. The maximum severity code was C corresponding to CTCAE grade 2. This results in a score of 0 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 (only points for CTCAE grade ≥ 3).
The lack of a severe clinical effect also results in a score of 0 of the maximum of 3 points for the second and third criterion of the clinical implication score: the level of evidence supporting an associated clinical effect grade ≥ 3 and the number needed to genotype (NNG) in the Dutch population to prevent one clinical effect code ≥ D (grade ≥ 3).
The American Summary of Product Characteristics (SmPC) of imipramine mentions the CYP2D6 PM phenotype, but the Dutch SmPC does not. This results in 0 out of the maximum of 2 points for the fourth and last criterion of the clinical implication score, the pharmacogenetics information in the SmPC (only points for at least one genotype/phenotype mentioned in the (Dutch) SmPC).

The table below follows the KNMP definitions for EM, PM, IM and UM. Therefore, the definitions of EM, PM, IM and UM used in the table below may differ from the definition used by the authors in the article.

<table>
<tr><th>Source</th><th>Code</th><th>Effect</th><th>Comments</th></tr>
<tr><td>ref. 1
Berm E et al.
Relation between CYP2D6 genotype, phenotype and therapeutic drug concentrations among nortriptyline and venlafaxine users in old age psychiatry.
Pharmacopsychiatry 2016;49:186-190.
PubMed PMID: 27101231.</td><td>3

PM: AA
IM: AA</td><td>38 patients with major depressive disorder aged 60 years or older with a mean age of 72 years were treated with nortriptyline. Dosing was adapted based on clinical effects and therapeutic drug monitoring. Therapeutic drug monitoring was performed 3, 5 and 12 weeks after start of nortriptyline (in respectively 38, 33 and 27 patients). The reason for patients not completing therapeutic drug monitoring was loss to follow-up.
Co-medication with psychotropic drugs was restricted to oxazepam, temazepam, haloperidol or risperidone, but somatic medication with effect on CYP2D6 was not excluded.

Genotyping:
- 25x EM
- 10x IM
- 3x PM

Results:
<table>
<tr><td colspan="4">Results compared to EM:</td></tr>
<tr><td colspan="2"></td><td>PM</td><td>IM</td><td>value for EM</td></tr>
<tr><td rowspan="3">% of patients with a supratherapeutic nortriptyline plasma concentration (> 150 ng/ml)</td><td>3 weeks</td><td colspan="2">NS for PM versus IM versus EM</td><td>16%</td></tr>
<tr><td>5 weeks</td><td colspan="2">trend for an increase for PM versus IM versus EM (p = 0.07) (NS)</td><td>12%</td></tr>
<tr><td>12 weeks</td><td colspan="2">NS for PM versus IM versus EM</td><td>20%</td></tr>
</table>
Note: Genotyping was for *3 and *4. Next to gene multiplication, these are the most important gene variants in this Dutch population.</td><td>Authors' conclusion: 'Genotype information could be used as a valuable tool, in addition to therapeutic drug monitoring, to prevent supratherapeutic drug levels of nortriptyline or venlafaxine in elderly patients with a PM genotype.'</td></tr>
<tr><td>ref. 2
Hodgson K et al.
Exploring the role of</td><td>4</td><td>284 patients with moderate to severe unipolar disorder were treated with nortriptyline for 12 weeks. Adverse event data were available for 251 patients. Nortriptyline</td><td>Authors' conclusion: 'In this sample where antidepres-</td></tr>
</table>

<table>
<tr>
<td>drug-metabolising enzymes in antidepressant side effects. Psychopharmacology (Berl) 2015;232:2609-17. PubMed PMID: 25761838.

ref. 2, continuation</td>
<td>PM: AA
IM: AA
UM: AA</td>
<td>was initiated at 50 mg/day and titrated to a target dose of 100 mg/day within the first 2 weeks unless adverse events limited dose increase, and could be further increased to 150 mg/day (and up to 200 mg/day if there was clinical agreement that a higher dose was needed). Dose titration was informed by assessments of depressive symptoms and adverse events.
The presence or absence of 21 adverse events was assessed weekly with the self-report Antidepressant Side Effect Checklist (ASEC), which was also administered prior to treatment. Associations with total adverse event burden were tested using linear models, whilst the weekly presence/absence of each specific adverse event was examined using logistic models. When considering the 21 different adverse events, Bonferroni correction for multiple testing was applied (significance if $p < 0.002381$ (i.e. $< 0.05/21$)). To ensure that reports of adverse events were not confounded by the severity of depression, MADRS scores were entered as a covariate in all analyses, along with baseline reports of adverse events, age, sex, linear and quadratic effects of time and centre of recruitment. When testing CYP2D6 genotype as a predictor, both CYP2D6-inhibiting medication and dose of nortriptyline were used as covariates.
Finally, CYP2D6 genotype was considered as a predictor of time to study discontinuation, using a survival Cox proportional hazards model. Covariates of age, sex, centre, baseline depression and baseline total adverse event score were included in the model.
Co-medication with psychotropic drugs was restricted to occasional use of hypnotics. Other medication was not excluded, but CYP2D6-inhibiting co-medication was controlled for. In week 8, no patients used CYP2D6 inducers and 4.7% used weak CYP2D6 inhibitors (combined oral contraceptive pill, amiodarone or ranitidine).
The smallest sample size included in these analysis was the 168 patients taking nortriptyline with both plasma concentration and dose information available. It was calculated that in this sample, it is possible to detect an effect size explaining 4.7% of the variance in outcome with 80% power, at a p value threshold of 0.05. This corresponds to 0.52 points on the ASEC when measuring total adverse event burden. For study drop-out, hazard ratios of 0.64 (or 1.56) could be detected at $p < 0.05$ with 80% power.

Genotyping:
- 238x EM+IM (gene dose 1.5-2 and gene dose 1/0)
- 20x IM (gene dose 0.5 and gene dose 0.5/0.5)
- 20x PM
- 6x UM

Results:
There was no association of CYP2D6 genotype with:
- total number of adverse events (NS, also NS for PM versus IM+EM+UM)
- the following specific adverse events (all NS):
<table>
<tr><td>dry mouth</td><td>problems with urination</td></tr>
<tr><td>drowsiness</td><td>palpitations</td></tr>
<tr><td>insomnia (difficulty sleeping)</td><td>feeling light-headed on standing</td></tr>
<tr><td>blurred vision</td><td>feeling like the room is spinning around</td></tr>
</table>
</td>
<td>sant dosage is titrated using clinical judgement, P450 genotypes do not explain differences between patients in side effects with antidepressants.’</td>
</tr>
</table>

<table>
<tr>
<td>ref. 2, continuation</td>
<td></td>
<td>
<table>
<tr><td>headache</td><td>sweating</td></tr>
<tr><td>constipation</td><td>increased body temperature</td></tr>
<tr><td>diarrhoea</td><td>tremor</td></tr>
<tr><td>increased appetite</td><td>disorientation</td></tr>
<tr><td>decreased appetite</td><td>yawning</td></tr>
<tr><td>nausea or vomiting</td><td>weight gain</td></tr>
<tr><td>problems with sexual function</td><td></td></tr>
</table>
- study discontinuation (NS)

Note: In this study there was also no correlation of plasma nortriptyline concentrations in week 8 with the total number of adverse events and with specific adverse events other than dry mouth. Nine European centres participated in the study and a significant effect of the centre of recruitment on nortriptyline concentrations was observed.

Note: Genotyping was for 33 variants with the Roche AmpliChip P450.
</td>
<td></td>
</tr>
<tr>
<td>ref. 3
Hodgson K et al. Genetic differences in cytochrome P450 enzymes and antidepressant treatment response.
J Psychopharmacol 2014;28:133-41.
PubMed PMID: 24257813.</td>
<td>4</td>
<td>Efficacy data for 334 patients from the same study as Hodgson 2015 were analysed. Of the 79% of patients who did not drop out of the study before week 8, nortriptyline plasma concentrations were measured for 161 patients and 10-hydroxynortriptyline plasma concentrations for 158 patients. The mean nortriptyline dose in week 8 was 104.9 mg/day, resulting in a mean nortriptyline plasma concentration of 92.6 ng/ml.
The severity of depressive symptoms was measured weekly, using the Montgomery-Åsberg Depression Rating Scale (MADRS). Prior to treatment, mean scores on the MADRS were 28.76.
Trough plasma concentrations of nortriptyline and 10-hydroxynortriptyline were measured in week 8. For ease of interpretation, standardised plasma concentration measurements were calculated, with a mean of 0 and a standard deviation of 1.
Significant differences were observed in treatment response outcomes in patients with and without plasma concentration measurements who remained in the study until at least week 8. Patients with plasma concentration measurements available were more likely to have responded to treatment than those without plasma concentration measurements (S).
All analyses were performed with linear mixed effect models including age, sex, cytochrome CYP2D6-inhibiting co-medication and centre of recruitment as covariates. Daily dose of drug was entered as a covariate into the model investigating the effect of CYP2D6 genotype on standardised plasma concentrations to consider dose-independent effects (CYP2D6 genotype was found to be unrelated to nortriptyline dose). Baseline depression severity, linear and quadratic effects of time, and individual were included as covariates in the model investigating the effect of CYP2D6 genotype on treatment response.
Co-medication with CYP2D6 inhibitors had a significant effect on both nortriptyline and 10-hydroxynortriptyline plasma concentrations, but all analyses corrected for this co-medication and results were similar when patients taking CYP2D6 inhibitors were excluded from the analysis.</td>
<td>Authors' conclusion: 'While there is a significant relationship between the CYP450 genotype and serum concentrations of escitalopram and nortriptyline, the genotypes are not predictive of differences in treatment response for either drug.'</td>
</tr>
</table>

<table>
<tr>
<td>ref. 3, continuation</td>
<td>PM: A
IM: A
UM: A</td>
<td>Uher 2012 calculated that, for studies addressing predictors of antidepressant treatment outcomes, continuous biomarkers (such as serum levels) should explain at least 6.3% of the variance in treatment response in order to be clinically significant. It was calculated that a sample size of n = 120 would be needed to detect an effect size of this magnitude with p = 0.05, and power of 80%. This study exceeds this sample size, and thus is adequately powered to detect clinically significant associations between serum levels of antidepressant and treatment response.

Genotyping (calculated with the percentages for all patients (treated with nortriptyline or escitalopram)):
- 273x EM+IM (gene dose 1.5-2 and gene dose 1/0)
- 26x IM (gene dose 0.5 and gene dose 0.5/0.5)
- 24x PM
- 11x UM

Results:
<table><tr><td>Results for PM versus IM versus (EM+gene dose 1/0) versus UM:</td></tr><tr><td>- no difference in treatment response (NS)</td></tr><tr><td>- increase in the (dose-corrected) nortriptyline plasma concentration (S)</td></tr><tr><td>- decrease in the (dose-corrected) 10-hydroxynortriptyline plasma concentration (S)</td></tr><tr><td>- no difference in nortriptyline dose (NS)</td></tr></table>
Note: In this study there was also no correlation of nortriptyline plasma concentrations with treatment response (NS). Higher 10-hydroxynortriptyline plasma concentrations were associated with poorer treatment response, but not after correction for drug dose. Because dose titration was based on depressive-symptoms and adverse events, higher drug doses were prescribed to patients failing to adequately respond to treatment. Nine European centres participated in the study and a significant effect of the centre of recruitment on nortriptyline dose and dose-corrected nortriptyline concentrations was observed.</td>
<td></td>
</tr>
<tr>
<td>ref. 4
Lee SY et al.
Sequence-based CYP2D6 genotyping in the Korean population.
Ther Drug Monit 2006;28:382-7.</td>
<td>3

IM: A

UM: AA</td>
<td>16 Korean volunteers (12x EM (3x *1/*1, 8x *1/*10, 1x *2/*10), 3x IM (2x *10/*10, 1x *5/*10), 1x UM (*2N/*10)) received a single dose of nortriptyline 15 mg.

IM versus EM:
- AUC NT increased from 743.2 to 1898.4 µg.h/L (S by 155%)

UM versus EM:
- AUC NT decreased from 743.2 to 572.0 µg.h/L (NS by 23%)</td>
<td>AUC NT versus EM:
IM: 255%
UM: 77%</td>
</tr>
<tr>
<td>ref. 5
Lee S et al.
A case report of a poor metabolizer of CYP2D6 presented with unusual responses to nortriptyline medication.
J Korean Med Sci 2004;19:750-2.</td>
<td>2

IM: C</td>
<td>- A patient receiving nortriptyline 150 mg/day developed side effects (dry mouth, constipation, dizziness), C_{ss} is 471 µg/L. No side effects when the dose was reduced to 50 mg/day. Genotype: *5/*10.</td>
<td></td>
</tr>
</table>

ref. 6 Roberts RL et al. No evidence of increased adverse drug reactions in cytochrome P450 CYP2D6 poor metabolizers treated with fluoxetine or nortriptyline. Hum Psychopharmacol 2004;19:17-23.	3 PM: AA	60 patients, 56x EM+IM (carriers of *1, *2, *9 or *10), 4x PM (*4/*4 or *4/*5) received NT 25-75 mg for 3 days, after which the dose was adjusted based on side effects, concentrations and clinical effect, study duration was 6 weeks, no relevant co-medication; - PM and EM+IM had equal levels of side effects after 6 weeks. - PM had a slightly lower dose after 6 weeks than EM+ IM. NOTE: the aim of the study (examining whether PMs suffered more side effects, measured after 3 and 6 weeks) was obscured by the fact that the dose could be adjusted during the study based on side effects.	Authors' conclusion: 'These findings suggest that inability to efficiently metabolize antidepressants that are CYP2D6 substrates does not necessarily lead to increased occurrence of antidepressant-associated adverse drug reactions.'
ref. 7 Dalen P et al. Disposition of debrisoquine and nortriptyline in Korean subjects in relation to CYP2D6 genotypes, and comparison with Caucasians. Br J Clin Pharmacol 2003;55:630-4.	3	10 healthy volunteers, 5x *1/*1, 5x *1/*10, a single dose of 25 mg nortriptyline, no co-medication; - *1/*10: for nortriptyline, an increase in the AUC versus *1/*1 from 1591 to 1672 nM·h (NS by 5%), decrease in Cl_{or} from 1.9 to 1.0 L/kg/h (NS by 47%). For HNT, a decrease in AUC HNT from 2,317 to 2,143 (NS by 8%). Increase in AUC ratio NT/HNT from 0.69 to 0.77 (NS by 12%).	
ref. 8 Murphy GM et al. CYP2D6 genotyping with oligonucleotide microarrays and nortriptyline concentrations in geriatric depression. Neuropsychopharmacol 2001;25:737-43	3 IM: A	36 geriatric patients, 18x EM (5x *1/*1, 12x *1/*2, 1x *1/*10) and 18x IM (2x *1/*3, 4x *1/*4, 1x *5/*10, 3x *2/*10, 2x *2/*2, 4x *2/*4,1x *3/*4, 1x *4/*4), NT dosed according to target concentration of 50-150 µg/L, with co-medication; - IM: increase in C_{ss}[b] nortriptyline versus EM from 1.3 to 2.9 ng/mL (S by 123%), decrease in dose from 66.9 to 43.3 mg (S by 30%). NOTE: it is not clear to what extent the co-medication had an effect on CYP2D6.	C_{ss} NT versus EM: IM: 223%
ref. 9 Kvist EE et al. Quantitative pharmacogenetics of nortriptyline: a novel approach. Clin Pharmacokinet 2001;40:869-77.	3 PM: AA IM: AA UM: AA	20 patients and 20 healthy study subjects, 5x no functional allele, 17x 1 functional allele, 12x 2 functional alleles, 6x 3 or more functional alleles (*1 and *2: functional alleles, *3, *4 and *5: completely dysfunctional alleles), patients received 50 mg 2-3 times daily, healthy study subjects received a single dose of 25-50 mg, no co-medication; For nortriptyline versus two functional alleles: - no functional allele: decreased Cl_{or} from 65.5 to 25.1 L/h (NS by 62%) - 1 functional allele: decreased Cl_{or} from 65.5 to 45.3 L/h (NS by 31%) - 3 functional alleles: increased Cl_{or} from 65.5 to 85.7 L/h (NS by 31%) - 4 functional alleles: increased Cl_{or} from 65.5 to 105.9 L/h (NS by 62%) - 13 functional alleles: increased Cl_{or} from 65.5 to 278.7 L/h (NS by 325%) The number of functional CYP2D6 alleles explains 21% of the inter-individual variation in Cl_{or} and 34% of the inter-individual variation in the C_{ss} nortriptyline. NOTE: genotyping performed, but only the number of	Cl_{or} NT versus EM: PM: 38% IM: 69% UM: 185%

		functional alleles is presented	
ref. 10 Morita S et al. Steady-state plasma levels of nortriptyline and its hydroxylated metabolites in Japanese patients: impact of CYP2D6 genotype on the hydroxylation of nortriptyline. J Clin Psychopharmacol 2000;20:141-9.	4 IM: A	41 patients, 7x *1/*1, 8x *1/*2, 16x *1/*10, 1x *2/*10, 3x *1/*5, 5x *10/*10, 1x *5/*10, nortriptyline 15-120 mg/day, no relevant co-medication; - 2 mutations (*10/*10, *10/*5): increase in C_{ss}^{b} NT versus no mutation from 70.3 to 147 ng/mL/mg/kg (S by 109%), decrease in C_{ss}^{b} HNT from 89.6 to 59.8 ng/mL/mg/kg (S by 33%). Increase in ratio NT/HNT from 0.82 to 2.71 (S by 230%). - 1 mutation (*1/*10, *2/*10,*1/*5): increase in Css^{b} NT versus no mutation from 70.3 to 98.4 ng/mL/mg/kg (S by 40%), increase in C_{ss}^{b} HNT from 89.6 to 107 ng/mL/mg/kg (NS by 19%). Increase in ratio NT/HNT from 0.82 to 1.04 (NS by 27%).	C_{ss}^{b} NT versus EM (*1/*1+*1/*2+*1/*10+*2/*10+*1/*5): IM: 170%
ref. 11 Yue QJ et al. Pharmacokinetics of nortriptyline and its 10-hydroxy metabolite in Chinese subjects of different CYP2D6 genotypes. Clin Pharmacol Ther 1998;64:384-90.	3 IM: A	15 healthy volunteers, 5x *1/*1, 5x *1/*10, 5x *10/*10, a single dose of 25 mg nortriptyline, no co-medication; - *10/*10: increase in the AUC NT versus *1/*1 from 1817 to 4002 nM·h (NS by 120%), decrease in Cl_{or} NT from 1.86 to 0.80 L/h/kg (NS by 57%). Decrease in AUC HNT from 2,273 to 1,704 nM·h (S by 25%). Increase in ratio AUC NT/HNT from 0.82 to 2.51 (by 244%). - *1/*10: increase in the AUC NT versus *1/*1 from 1,817 to 2,492 nM·h (NS by 37%), decrease in Cl_{or} NT from 1.86 to 1.39 L/h/kg (NS by 25%). Increase in AUC HNT from 2,273 to 2,975 nM·h (NS by 31%). Increase in ratio AUC NT/HNT from 0.82 to 0.94 (by 15%).	AUC NT versus EM (*1/*1+*1/*10): IM: 186%
ref. 12 Dalen P et al. 10-Hydroxylation of nortriptyline in white persons with 0, 1, 2, 3, and 13 functional CYP2D6 genes. Clin Pharmacol Ther 1998;63:444-52.	3 PM: A IM: A UM: A	20 healthy volunteers, 4x *4/*4, 5x *1/*1, 3x *1/*4, 2x *1/*5, 5x *2x2/*2, 1x *2x13/*1, a single dose of 25 mg NT (UM 50 mg), no co-medication; - 0 functional alleles: increase in AUC NT versus EM from 1,295 to 4,301 nM·h (S by 232%), t½ is 54.5 h. Decrease in AUC HNT from 1,711 to 1,537 nM·h (NS by 10%), t½ HNT is 52.2 h. Increase in ratio AUC NT/HNT from 0.77 to 2.89 (S by 275%). - 1 functional allele: increase in AUC NT versus EM from 1,295 to 3,617 nM·h (S by 179%), t½ is 47.5 h. Increase in AUC HNT from 1,711 to 1,856 nM·h (NS by 8%), t½ HNT is 39.7 h. Increase in ratio AUC NT/HNT from 0.77 to 2.06 (S by 168%). - 3 functional alleles: decrease in AUC NT versus EM from 1,295 to 860 nM·h (NS by 34%), t½ is 18.1 h. Increase in AUC HNT from 1,711 to 2,731 nM·h (NS by 60%), t½ HNT is 17.6 h. Decrease in ratio AUC NT/HNT from 0.77 to 0.32 (S by 58%). - 13 functional alleles: decrease in AUC NT versus EM from 1,295 to 267 nM·h (NS by 79%), t½ is 19 h. Increase in AUC HNT from 1,711 to 3,442 nM·h (NS by 101%), t½ HNT is 9.5 h. Decrease in ratio AUC NT/HNT from 0.77 to 0.08 (NS by 90%).	AUC NT versus EM: PM: 332% IM: 279% UM: 59%
ref. 13 Dahl M et al. Steady-state plasma levels of nortriptyline and its 10-hydroxy metabolite: relationship to the CYP2D6 genotype.	3 PM: A	21 patients, 7x *1/*1, 13x *1/*3 or *1/*4 or *1/*5, 1x *4/*4, nortriptyline 150 mg/day (1 person 100 mg/day), co-medication unknown; - PM: increase in C_{ss} NT versus *1/*1 from 2.60 to 6.40 (S by 146%), decrease in C_{ss} HNT from 5.20 to 4.50 (S by 13%), increase in ratio C_{ss} NT/HNT from 0.5 to 1.4 (S by 180%).	C_{ss} NT versus EM: PM: 246%. IM: 135%.

Psychopharmacol 1996;123:315-9. **ref. 13, continuation**	IM: A	- IM (*1/*3, *1/*4, *1/*5): increase in C_{ss} NT versus *1/*1 from 2.60 to 3.50 (NS by 35%), decrease in C_{ss} HNT from 5.20 to 3.50 (S by 33%), increase in ratio C_{ss} NT/HNT from 0.5 to 1.0 (S by 100%).	
ref. 14 Chen S et al. The cytochrome P450 2D6 (CYP2D6) enzyme polymorphism: screening costs and influence on clinical outcomes in psychiatry. Clin Pharmacol Ther 1996;60:522-34	1 IM: C PM: C	Side effects occurred in 8 patients (4x *1/*1, 1x *1/*3, 1x *1/*4, 1x *3/*9, 1x *4/*4) receiving nortriptyline 10-75 mg/day; co-medication unknown; Side effects that occurred in IM and PM following administration of NT: - *1/*3: 25-50 mg/day: nervousness, tinnitus - *1/*4: 75-100 mg/day: instability of the knees and nervousness - *3/*9: 10 mg/day: drowsiness, sluggishness - *4/*4: 10 mg/day: anxiety, agitation, nervousness NOTE: no analysis to determine whether the listed side effects could also be symptoms of the condition	
ref. 15 Bertilsson L et al. Molecular basis for rational megaprescribing in ultrarapid hydroxylators of debrisoquine. Lancet 1993;341:63.	2 UM: C	For one patient, the nortriptyline dose had to be increased to 500 mg/day (3-5x the standard dose) in order to achieve therapeutic plasma concentrations and a response. The patient was found to have a CYP2D6 duplication.	
ref. 16 Bertilsson L et al. Slow hydroxylation of nortriptyline and concomitant poor debrisoquine hydroxylation: clinical implications. Lancet 1981;1:560-1.	2 PM: C	A patient exhibited dizziness and hypotension 2 days after starting a low dose of nortriptyline (75 mg/day). Eight days after starting treatment, she complained about increasing fatigue and dizziness and appeared confused. C_{ss} NT was 1,300 nmol/L (normally 200-600 nmol/L at this dose). After twelve days of nortriptyline 25 mg/day, the C_{ss} NT was 742 nmol/L. The side effects disappeared once the dose was reduced to 20 mg/day.	
ref. 17 SmPC Nortrilen (nortriptyline) 01-04-17.		<u>Pharmacokinetic properties</u>: The metabolism is subject to genetic polymorphism (CYP2D6).	
ref. 18 SmPC Pamelor (nortriptyline) 28-07-14, USA.	0 PM: A	<u>Drug interactions</u>: Drugs metabolized by P450 2D6 The biochemical activity of the drug metabolizing isozyme cytochrome P450 2D6 (debrisoquin hydroxylase) is reduced in a subset of the Caucasian population (about 7% to 10% of Caucasians are so-called “poor metabolizers”); reliable estimates of the prevalence of reduced P450 2D6 isozyme activity among Asian, African, and other populations are not yet available. Poor metabolizers have higher than expected plasma concentrations of tricyclic antidepressants (TCAs) when given usual doses. Depending on the fraction of drug metabolized by P450 2D6, the increase in plasma concentration may be small, or quite large (8-fold increase in plasma AUC of the TCA).	

[a] corrected for the dose

[a] corrected for the dose and body weight

[#]: the calculations were based on the assumption that the metabolite 10-hydroxynortriptyline is half as potent as the mother substance nortriptyline.

Risk group	IM with CYP2D6 inhibitor, UM with CYP2D6 inducer

Comments:

- After 2010, case reports were not included in the risk analysis, because they did not add enough to the evidence.
- Cost-effectiveness:
  - Berm EJ et al. A model based cost-effectiveness analysis of routine genotyping for CYP2D6 among older, depressed inpatients starting nortriptyline pharmacotherapy. PLoS One 2016;11:e0169065. PubMed PMID: 28033366.

    Routine genotype-guided therapy for old aged Dutch depressed inpatients starting nortriptyline pharmacotherapy is not cost-effective (costs of € 1,333,000 per quality adjusted life year (QALY) gained) at current genotyping costs (€ 190 per test) compared to not-genotype-guided nortriptyline pharmacotherapy. However at test costs below € 40, genotype-guided therapy could be cost-effective (costs equal to or less than € 50,000 per QALY gained). Genotype-guided therapy consisted of a starting dose of 40% of the normal starting dose for CYP2D6 PM, 100% of the normal starting dose for CYP2D6 IM and EM and 160% of the normal starting dose for CYP2D6 UM.

    At genotyping test costs < € 35 per test, genotyping was both better (i.e. resulting in more QALYs) and cheaper than not genotyping.

    Varying all other input parameters at genotyping test costs of € 17 per test, showed a 95% probability that genotyping was cost-effective. At genotyping test costs of € 190, the probability was < 1%.

    If also the starting dose for IM was changed (to 60% of the normal starting dose), the calculated costs were € 2,380,626 per QALY gained and genotype-guided therapy could be cost-effective at genotyping test costs below € 68.

    Direct medical costs were calculated from a health-care insurance payers perspective. Costs were calculated during the dose titration phase (the first 12 weeks of therapy including a maximum of 3 dose adaptations based on therapeutic drug monitoring (TDM) (at 12 days, 43 days and 81 days after start of nortriptyline)). Patients were assumed to be discharged from hospital when antidepressant dose titration was completed. Patients discontinuing nortriptyline were assumed to receive tranylcypromine thereafter. For 1000 patients, not-genotype guided therapy resulted in total costs of € 7,374,826 and a loss of 4.57 QALYs. Genotype-guided therapy resulted in total costs of € 7,528,292 and a loss of 4.46 QALYs. Thus genotype-guided therapy resulted in costs of € 153,466 for a gain of 0.12 QALYs. Costs included in the calculation were hospitalisation costs of € 255.62/day (including costs for medication and the first TDM measurement), TDM costs of € 23.11/measurement, costs for ambulant contact with the psychiatrist of € 190.62, nortriptyline costs of € 0.08 for 10 mg, € 0.15 for 25 mg and € 0.29 for 50 mg, tranylcypromine costs of € 0.96 per 40 mg, and costs of € 188.20 for the genotyping test (genotyping of CYP2D6 *2, *3, *4, *5 and gene duplication). Proportions of therapeutic, sub-, and supratherapeutic plasma concentrations, and the reduction in inadequately dosed patients by starting dose adjustment (35% for both PM and UM) were derived from Jornil J et al. Risk assessment of accidental nortriptyline poisoning: the importance of cytochrome P450 for nortriptyline elimination investigated using a population-based pharmacokinetic simulator. Eur J Pharm Sci 2011;44: 265-72. Genotype frequencies (8% for PM, 11% for IM, 2% for UM and 79% for EM), average duration of inpatient care (28.6 days), shorter hospital stay when correctly dosed (13.0%), patients who discontinue therapy after the first dose evaluation (22%) and after second dose evaluation (8.5%) were also derived from literature. Assumptions made were that PMs and IMs could only receive a dose that was either correct or too high and UMs could only receive a dose that was either correct or too low. In addition, after dose adjustment, dosing could not become incorrect in an opposite way.

    Main parameters which influenced the costs per QALY gained were the genotyping test costs, the improvement in the duration of hospitalization among correctly dosed patients, mean duration of hospitalization, and the proportion of patients who discontinued nortriptyline pharmacotherapy.

    For the scenario in which also the starting dose for IMs was adjusted (to 60% of standard dose), the percentage of IMs being supratherapeutically dosed was assumed to be the average of the EM and PM group (56%) and the effect of dose adaptation was assumed to be the same as for PM and UM (35% reduction of incorrectly dosed patients). With respect to clinical validity, It has been reported that 38% of CYP2D6 IMs are false positive, i.e. have normal CYP2D6 enzyme activity despite the genetic variation (Rebsamen MC et al. The AmpliChip CYP450 test: cytochrome P450 2D6 genotype assessment and phenotype prediction. Pharmacogenomics J 2009;9:34-41). This means, these patients would be dosed too low when dose adaptations would be made based on genotype. So, although inclusion of dose adaptations for IMs was found to increase the costs savings from € 153,466 to € 122,346 by reduced inpatient care of correctly dosed IMs, it decreased the QALY gain from 0.12 to 0.05 per 1000 patients, due to false positive IM genotypes which resulted in more subtherapeutic dosed patients. Consequently, the costs per QALY gained increased to € 2,400,000 and the cost-effectiveness decreased. At test costs below € 68, genotype-guided therapy could be cost-effective in this scenario (costs equal to or less than € 50,000 per QALY gained). At genotyping test costs < € 66 per test, genotyping was both better (i.e. resulting in more QALYs) and cheaper than not genotyping in this scenario.
- Existing guideline:

Hicks JK et al. Clinical pharmacogenetics implementation consortium guideline (CPIC) for CYP2D6 and CYP2C19 genotypes and dosing of tricyclic antidepressants: 2016 update. Clin Pharmacol Ther 2016 Dec 20 [Epub ahead of print]. PubMed PMID: 27997040.
CPIC uses the same definition for PM as we do. However, CPIC uses different definitions for EM (gene dose 1-2), IM (gene dose 0.5) and UM (gene dose > 2). In addition, CPIC changed the name of EM to normal metaboliser (NM). The summary below uses the KNMP definitions for EM, PM, IM and UM.
CPIC states that the recommended starting dose of nortriptyline does not need dose adjustment for those with gene dose 1-2. In addition, CPIC states that a 25% reduction of the recommended dose may be considered for patients with a CYP2D6 gene dose of 0.5. As a reference for this percentage reduction they mention the 2011 publication of our dosing recommendations in Clinical Pharmacology and Therapeutics. However, this dosing recommendation is primarily based on patients with gene dose 1. In addition, for IM we recommended a nortriptyline dose reduction of 50% in that publication, which was decreased to 40% in 2012. Because patients with a CYP2D6 activity score of 1.0 are inconsistently categorised as intermediate or normal metabolisers in the literature, making these studies difficult to evaluate, CPIC classified the strength of the recommendation for gene dose 0.5 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).
CPIC states that CYP2D6 ultra-rapid metabolisers + gene dose 2.5 have a higher probability of failing nortriptyline pharmacotherapy due to subtherapeutic plasma concentrations, and alternate agents are preferred. CPIC mentions a documented case of a CYP2D6 ultrarapid metaboliser receiving large doses of nortriptyline in order to achieve therapeutic concentrations (Bertilsson L. et al. Extremely rapid hydroxylation of debrisoquine: a case report with implication for treatment with nortriptyline and other tricyclic antidepressants. Ther Drug Monit 1985:7;478-80. (This concerns the same case as in Bertilsson 1993)). CPIC indicates that this case had very high plasma concentrations of the nortriptyline hydroxy-metabolite, which may increase the risk for cardiotoxicity. CPIC states that, if nortriptyline is warranted, there are insufficient data in the literature to calculate a starting dose for a patient with CYP2D6 ultra-rapid metaboliser or gene dose 2.5 status, and therapeutic drug monitoring is strongly recommended.
Based on Bertilsson 1981, CPIC indicates that adverse effects are more likely in CYP2D6 poor metabolisers due to elevated tricyclic plasma concentrations; therefore, alternate agents are preferred. If a tricyclic is warranted, CPIC recommends to consider a 50% reduction of the usual dose, and strongly recommends therapeutic drug monitoring.
The therapeutic recommendations for nortriptyline are indicated below:

Dosing recommendations for nortriptyline for conditions requiring higher doses such as depression based on CYP2D6 phenotype[a]		
Phenotype	Therapeutic recommendation	Classification of recommendation
UM + gene dose 2.5	Avoid nortriptyline use due to potential lack of efficacy. Consider alternative drug not metabolised by CYP2D6. If nortriptyline is warranted, consider titrating to a higher target dose (compared to normal metabolisers).[b] Utilise therapeutic drug monitoring to guide dose adjustments.	Strong
gene dose 1-2	Initiate therapy with recommended starting dose.[c]	Strong
gene dose 0.5	Consider a 25% reduction of recommended starting dose.[c] Utilise therapeutic drug monitoring to guide dose adjustments.[b]	Moderate
PM	Avoid tricyclic use due to potential for side effects. Consider alternative drug not metabolised by CYP2D6. If nortriptyline is warranted, consider a 50% reduction of recommended starting dose.[c] Utilise therapeutic drug monitoring to guide dose adjustments.[b]	Strong

[a] Dosing recommendations only apply to higher initial doses of nortriptyline for treatment of conditions such as depression. For conditions at which lower initial doses are used, such as neuropathic pain, CPIC recommends no dose modifications for PM or IM, because it is less likely that PM or gene dose 0.5 will experience adverse effects due to supratherapeutic plasma concentrations of nortriptyline. However, CPIC indicates that these patients should be monitored closely for side effects. In addition, if larger doses of TCA are warranted, CPIC recommends following the gene-based dosing guidelines in the table above. For UM+gene dose 2.5, CPIC recommends considering an alternative agent. Based on predicted and observed pharmacokinetic data in those with depression, CYP2D6 UM+gene dose 2.5 may be at an increased risk of failing nortriptyline therapy for neuropathic pain due to lower than expected drug concentrations (Dworkin RH et al. Pharmacologic management of neuropathic pain: evidence-based recommendations. Pain 2007;13: 237-51).
[b] Titrate dose to observed clinical response with symptom improvement and minimal (if any) side effects.
[c] Patients may receive an initial low dose of nortriptyline, which is then increased over several days to the recommended steady-state dose. The starting dose in this guideline refers to the recommended steady-state dose.

As evidence linking CYP2D6 genotype with nortriptyline phenotype, CPIC mentions Berm 2016, Hodgson 2014, Chua 2013, Piatkov 2011, Bijl 2008, Lee 2006, Kawanishi 2004, Lee 2004, Kvist 2001, Laine 2001, Murphy 2001, Morita 2000, Dalen 1998, Yue 1998, Chen 1996, Dahl 1996, Bertilsson 1993, Bertilsson 1985,

Nordin 1985, Woolhouse 1984, Bertilsson 1981, Mellstrom 1981 and Bertilsson 1980. All these studies, except for Bertilsson 1980, Mellstrom 1981, Woolhouse 1984, Nordin 1985, Bertilsson 1985, Laine 2001, Kawanishi 2004, Bijl 2008, and the case reports of Piatkov 2011 and Chua 2013, are included in our risk analysis. Chua 2013 and Piatkov 2011 were not included in our risk analysis because we did not include case reports published after 2010. Bijl 2008 and Kawanishi 2004 were not included in our risk analysis because only a minority of the patients in the studies used nortriptyline (35 of the 1198 patients (among whom 807 TCA users) in Bijl 2008 and 1 of the 8 UM in Kawanishi 2004), and results were not reported separately for nortriptyline. Laine 2001 was not included in our risk analysis, because the study compares UM to UM with paroxetine not to EM. Bertilsson 1985 was not included in our risk analysis, because it describes the same case as Bertilsson 1993. Bertilsson 1980, Mellstrom 1981, Woolhouse 1984 and Nordin 1985 were not included in our risk analysis, because phenotyping was not used to distinguish and compare different pharmacogenetic genotypes. In addition to the studies considered by CPIC, our risk analysis includes the studies of Hodgson 2015, Roberts 2004 and Dalen 2003. CPIC indicates that these studies provide a high level of evidence for a decreased nortriptyline metabolism in PM and in gene dose 0.5 compared to gene dose 1-2 (based on 5 references for PM and on 2 references for gene dose 0.5). Likewise, CPIC indicates that these studies provide a high level of evidence for an increased nortriptyline metabolism in UM + gene dose 2.5 compared to gene dose 1-2 (based on 4 references, including Laine 2001). In addition, CPIC indicates that these studies provide a high level of evidence for a correlation between the number of CYP2D6 variant alleles and for a correlation of debrisoquine hydroxylation with nortriptyline metabolism (8 references, including Chua 2013, for the number of variant alleles, and the 4 references Bertilsson 1980, Mellstrom 1981, Woolhouse 1984 and Nordin 1985 for the debrisoquine hydroxylation). CPIC indicates that these studies provide a moderate level of evidence for the requirement of a decreased dose of nortriptyline in PM (3 references, including Bijl 2008 and including Hodgson 2014 that does not report an association) and an increased dose of nortriptyline in UM + gene dose 2.5 (Bertilsson 1993) compared to gene dose 1-2. In addition, CPIC indicates that these studies provide a moderate level of evidence for an increased risk of side effects in carriers of no function and decreased function alleles compared to carriers of other alleles (5 references, including Piatkov 2011 and including Hodgson 2014 that does not report an association) and for a decreased response in UM + gene dose 2.5 receiving nortriptyline (3 references including Kawanishi 2004 and including Bertilsson 1985 and Bertilsson 1993 that describe the same case). Finally, CPIC indicates a moderate level of evidence from a pharmacokinetic model using published data for the intrinsic clearance of nortriptyline being a linear function of the number of functional CYP2D6 alleles (Kvist 2001).
CPIC also took other gene-based dosing recommendations in consideration, including the 2008 and 2011 publications of our dosing recommendations in Clinical Pharmacology and Therapeutics.
CPIC also provides therapeutic recommendations based on both CYP2D6 and CYP2C19 genotypes. For CYP2D6 UM+gene dose 2.5 and for CYP2D6 PM the therapeutic recommendations for the different CYP-2C19 phenotypes are similar, reflecting the stronger influence of the CYP2D6 phenotype compared to the CYP2C19 phenotype. CPIC indicates that further studies are needed to develop moderate or strong dosing recommendations for TCAs when considering combined CYP2D6/CYP2C19 phenotypes. At the moment, insufficient data are available. Based on Steimer 2005, CPIC mentions that patients carrying at least one CYP2D6 no function allele and two CYP2C19 normal function alleles had an increased risk of experiencing side effects when administered amitriptyline. This would argue for a therapeutic recommendation also for patients with CYP2D6 gene dose 1, which is the predominant phenotype in this patient group.
On 3-10-2018, there was not a more recent version of the recommendations present on the PharmGKB- and on the CPIC-site.

Date of literature search: 26 September 2018.

	Phenotype	Code	Gene-drug interaction	Action	Date
KNMP Pharmacogenetics Working Group decision	PM	3 C	yes	yes	19 November 2018
	IM	4 C	yes	yes	
	UM	3 C	yes	yes	

Mechanism:
Nortriptyline is mainly metabolised by CYP2D6 to the active metabolite E-10-hydroxynortriptyline. This metabolite is approximately half as potent as nortriptyline itself. A CYP2D6 genetic polymorphism may cause a change in the plasma concentration of nortriptyline and its active metabolite.
Nortriptyline is converted by CYP2D6 and CYP2C19 to the inactive metabolite desmethylnortriptyline.
The therapeutic range is 50-150 ng/ml and values higher than 250 ng/ml are considered to be toxic. The Z-hydroxy-metabolites can cause cardiotoxicity and plasma concentrations of Z-hydroxynortriptyline higher than 40 ng/ml are considered to be toxic.

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 genotype 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**
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+	0+
Corresponding Clinical Implication Score:		Potentially beneficial