CYP2C9: siponimod 7160 to 7166

*1 = no CYP2C9 gene variant, normal activity, *2 = CYP2C9 gene variant with decreased activity, *3 = CYP2C9 gene variant with strongly decreased activity, AUC = area under the concentration-time curve, $AUC_{0\text{-last}}$ = AUC from time zero to the time of the last quantifiable concentration, Cl_{or} = oral clearance, ECG = electrocardiogram, IM = IM other = intermediate metaboliser, other genotype (decreased CYP2C9 enzyme activity due to the presence of one gene variant with decreased activity other than *2 or *3), NM = normal metaboliser (*1/*1) (normal CYP2C9 enzyme activity), NS = non-significant, PM = PM other = poor metaboliser, other genotype (strongly decreased CYP2C9 enzyme activity due to the presence of two gene variants with decreased activity, of which at least one other than *2 or *3), S = significant, $t_{1/2}$ = half-life

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, then the health care professional should consider the next best option.

Brief summary and justification of choices:
Siponimod is primarily metabolised by CYP2C9, and to a lesser extent by CYP3A4, to hydroxysiponimod. This metabolite is then glucuronidated. Both metabolites have a very limited pharmacological activity (hydroxysiponimod 470-fold less and siponimodglucuronide 10,000-fold less than siponimod).
There are no studies investigating clinical effects of gene variants leading to a reduced CYP2C9 activity other than two studies in volunteers receiving either a lower than normal dose for a maximum of 3 days or receiving a single dose. A study found a numerical increase in AUC of siponimod for *2/*3 and *3/*3 compared to *1/*1 after a single dose (2.0-fold and 3.8-fold respectively). Another study found a numerical increase in AUC of siponimod for *1/*3 compared to *1/*2 after a single dose (1.3-fold). The SmPC reported calculated numerical increases in AUC of siponimod compared to *1/*1 of 1.0-fold for *1/*2, 1.6-fold for *1/*3, 1.3 fold for *2/*2, 1.9-fold for *2/*3 and 2.8-fold for *3/*3, using modelling with *in vitro* data and the data from the two studies.
Based on the size of the effect for *3/*3 in the small study, the KNMP concludes that there is enough evidence for a gene-drug interaction. In general, the KNMP only provides therapeutic recommendations if clinical effects have been found or if the drug has a narrow therapeutic range. For siponimod, no clinical effects have been found and there are no indications for a narrow therapeutic range. As mentioned in Gardin 2019 Clin Pharmacokinet, siponimod doses up to 20 mg/day (10 times the normal dose of 2 mg/day) were well tolerated for a period of 28 days. However, the risk for bradycardia was diminished by starting with a low dose (0.25 mg/day). In addition, siponimod decreases the lymphocyte count and thus increases infection risk, suggesting a high likelihood for a negative long term effect of overdosing. Finally, long term treatment with other immune-modulating drugs for multiple sclerosis has been associated with progressive multifocal leukoencephalopathy, a life-threating adverse event caused by activation of the JC virus in the brain. Thus, although there is not enough evidence for pharmacotherapeutic recommendations for siponimod, there is not enough evidence to reject them either. For this reason, the KNMP decided to adopt the pharmacotherapeutic recommendations in the SmPC of siponimod (i.e. yes/no-interactions for *1/*2 and *2/*2, and yes/yes-interactions for *1/*3, *2/*3 and *3/*3). The pharmacotherapeutic recommendation for *1/*3 and *2/*3 is to use halve of the normal maintenance dose and take into account an increased effect of moderate CYP3A4 inducers. The pharmacotherapeutic recommendation for *3/*3 is to avoid siponimod. There are no data for IM and PM, the genotypes involving one or two alleles with reduced activity other than *2 and *3. Because the *6 allele has no activity, these phenotypes include genotypes with an effect size equal to or larger than *1/*3 and *3/*3. Therefore, for these phenotypes the same therapeutic recommendations are given as for *1/*3 and *3/*3, respectively (yes/yes-interactions).
You can find an overview of the observed kinetic and clinical effects in the background information text of the gene-drug interactions in 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:
Due to the absence of publications of multiple dose studies using the recommended therapeutic dose or case reports with patients with a CYP2C9 genotype leading to reduced CYP2C9 activity in medical journals, and thus the absence of evidence of an increase in adverse events code ≥ D (grade ≥ 3) in these patients, the clinical implication of the gene-drug interaction scores only 2 out of the maximum of 10 points (with pre-emptive genotyping considered to be poten-

tially beneficial for scores ranging from 0 to 2 points) (see also the clinical implication score tables at the end of this risk analysis).
However, there is not enough evidence to reject the warnings and recommendations in the SmPC. In addition, the Clinical Implication Score is mainly (for 80%) based on studies published in medical journals and therefore not suited to determine the clinical implication for gene-drug interactions for which data are predominantly provided by pre-registration studies. For these reasons, the KNMP decided to ignore the Clinical Implication Score and adopt the genotyping recommendation in the SmPC. The SmPC indicates that genotyping must be performed before starting siponimod to guide drug and dose selection. This would amount to genotyping being essential for drug safety according to the nomenclature of the KNMP Pharmacogenetics Working Group.

The table below follows the KNMP definition for NM, PM and IM, unless stated otherwise. The definition of NM, PM and IM used in the table below may therefore differ from the definition used by the authors in the article.

Source	Code	Effect				Comments
ref. 1 Gardin A et al. Siponimod pharmacokinetics, safety, and tolerability in combination with the potent CYP3A4 inhibitor itraconazole in healthy subjects with different CYP2C9 genotypes. Eur J Clin Pharmacol 2019;75:1565-74. PubMed PMID: 31392364.	3	30 healthy volunteers, selected for their CYP2C9 genotype, received a single dose of siponimod 0.25 mg. Co-medication and herbal and dietary supplements were excluded. Genotyping: - 17x *1/*2 - 13x *1/*3 Results:				Author's conclusion: "Geometric mean AUC_{inf}, $T_{1/2}$, and median T_{max} were higher while systemic clearance was lower in cohort 2 (CYP2C9 *1*3) than cohort 1 (CYP2C9 *1*2)."
		Results compared to *1/*1 in Gardin 2019, Clin Pharmacokinet:				
			*1/*3	*1/*2	value for *1/*1 in Gardin 2019, Clin Pharmacokinet	
		adverse events	No major differences were observed in cardiac monitoring data between *1/*2 and *1/*3. The absolute lymphocyte count reduction was consistent with the magnitude of absolute lymphocyte count reduction observed in previous clinical studies (with not for their CYP2C9 genotype selected subjects). No volunteers experienced severe adverse events and no deaths were reported. No clinically relevant changes were observed in clinical laboratory variables or vital signs.			
	*1/*2: AA *1/*3: AA	AUC siponimod	x 1.41 (NS)	x 1.10 (NS)	70.5 h.ng/ml	AUC siponimod compared to *1/*1 in another study: *1/*2: 110% *1/*3: 141%
		$AUC_{0\text{-last}}$ hydroxysiponimod	x 0.58 (NS)	x 0.98 (NS)	1.09 h.ng/ml	
		AUC siponimodglucuronide	x 0.99 (NS)	x 1.10 (NS)	32.7 h.ng/ml	
		$t_{1/2}$ siponimod	x 1.46 (NS)	x 0.99 (NS)	28.1 h	
		$t_{1/2}$ hydroxysiponimod	x 1.48 (NS)	x 0.91 (NS)	37.9 h	
		$t_{1/2}$ siponimodglucuronide	x 1.29 (NS)	x 1.02 (NS)	32.9 h	
		Cl_{or} siponimod	x 0.74 (NS)	x 0.96 (NS)	3.6 L/h	
		Significance of the differences between *1/*3 and *1/*2 was not determined. Significance of the differences for *1/*2 or *1/*3 compared to *1/*1 in other studies was not determined.				
		Note: Genotyping was for *2 and *3. These are the most important gene variants in this population from the USA.				

<table>
<tr>
<td>ref. 2
Gardin A et al. Effect of fluconazole coadministration and CYP2C9 genetic polymorphism on siponimod pharmacokinetics in healthy subjects. Clin Pharmacokinet 2019;58:349-61.
PubMed PMID: 30088221.</td>
<td>3

*2/*3: AA

*3/*3: AA</td>
<td>24 healthy volunteers, selected for their CYP2C9 genotype, received a single dose of siponimod 0.25 mg (single dose study). Volunteers with *1/*1 genotype were matched by body weight (± 10%) to the volunteers with a variant genotype. In a second study, the 12 volunteers with a variant genotype received siponimod for 3 days (0.25 mg on day 1 and 2, 0.5 mg on day 3) (multiple dose study).
Co-medication, herbal and dietary supplements and smoking were excluded. For siponimod, the time to steady state is 6 days, but siponimod exposure is dose-proportional and time-independent.
A sample size of six subjects per genotype group was estimated to provide 80% power to detect a 1.5-fold change in pharmacokinetic parameters (C_{max}, $AUC_{0\text{-}last}$ and $AUC_{0\text{-}\infty}$) of siponimod at a significance level of 0.05, with an assumption of an intersubject coefficient of variance of 25% based on historical data. With six completed subjects per group, the 90% confidence interval of the pharmacokinetics parameters ratio was expected to be within 72–138% of the observed mean, with 90% coverage probability. The SimCYP simulations (based on in vitro data) predicted a 4.5-fold higher (based on AUC_{∞}) siponimod exposure in subjects with a *3/*3 versus *1/*1 genotype.

Genotyping:
- 12x *1/*1
- 6x *2/*3
- 6x *3/*3

Results:
<table>
<tr><td colspan="4">Single dose study: results compared to NM:</td></tr>
<tr><td></td><td>*3/*3</td><td>*2/*3</td><td>value for *1/*1</td></tr>
<tr><td>adverse events</td><td colspan="3">16.7% of the volunteers reported at least one adverse event. Headache was the most prevalent adverse events (8.3%). Only one adverse event in either the single dose or multiple dose study was considered to be possibly related to siponimod (somnolence). The low incidence of adverse events did not allow for any meaningful comparison across genotype or phenotype groups (NS).
No deaths or serious adverse events were reported during the study, and no systematic, clinically relevant alterations of laboratory, vital signs or ECG data were identified. No clinically significant changes were observed with respect to body weight throughout the study.</td></tr>
<tr><td>AUC siponimod</td><td>x 3.84 (NS)</td><td>x 2.04 (NS)</td><td>70.5 h.ng/ml</td></tr>
<tr><td>$AUC_{0\text{-}last}$ hydroxysiponimod</td><td>-</td><td>x 0.61 (NS)</td><td>1.09 h.ng/ml</td></tr>
<tr><td>AUC siponimodglucuronide</td><td>x 0.34 (NS)</td><td>x 0.88 (NS)</td><td>32.7 h.ng/ml</td></tr>
<tr><td>$AUC_{0\text{-}24h}$ siponimod</td><td>x 1.35 (NS)</td><td>x 1.41 (NS)</td><td>31.6 h.ng/ml</td></tr>
<tr><td>$AUC_{0\text{-}24h}$ hydroxysiponimod</td><td>x 0 (NS)</td><td>x 0.56 (NS)</td><td>0.618 h.ng/ml</td></tr>
<tr><td>$AUC_{0\text{-}24h}$ siponimodglucuronide</td><td>x 0.09 (NS)</td><td>x 0.51 (NS)</td><td>12.1 h.ng/ml</td></tr>
<tr><td>$t_{1/2}$ siponimod</td><td>x 4.48 (NS)</td><td>x 1.81 (NS)</td><td>28.1 h</td></tr>
<tr><td>$t_{1/2}$ hydroxysiponimod</td><td>-</td><td>x 2.77 (NS)</td><td>37.9 h</td></tr>
<tr><td>$t_{1/2}$ siponimodglucuronide</td><td>x 2.92 (NS)</td><td>x 1.67 (NS)</td><td>32.9 h</td></tr>
</table>
</td>
<td>Author’s conclusion:
”Changes in siponimod pharmacokinetics, when coadministered with fluconazole at steady-state and in subjects with different CYP2C9 genotypes, indicate that the reduced CYP2C9 enzymatic activity does not affect the absorption phase of siponimod but prolongs the elimination phase.”

AUC siponimod compared to *1/*1:
*2/*3: 204%
*3/*3: 384%</td>
</tr>
</table>

<table>
<tr>
<td>ref. 2, continuation</td>
<td></td>
<td>
<table>
<tr><td>Cl_{or} siponimod</td><td>x 0.25 (NS)</td><td>x 0.47 (NS)</td><td>3.6 L/h</td></tr>
<tr><td colspan="4">Significance was not determined.</td></tr>
</table>

<table>
<tr><td colspan="3">Multiple dose study: results compared to non-genotype selected healthy volunteers on the same dosing regime in an earlier study:</td></tr>
<tr><td></td><td>*3/*3</td><td>*2/*3</td></tr>
<tr><td>adverse events</td><td colspan="2">Three (25%) of the 12 volunteers with a variant genotype experienced at least one adverse event. Headache was the most prevalent adverse event (8.3%). Only one adverse event in either the single dose or multiple dose study was considered to be possibly related to siponimod (somnolence). The low incidence of adverse events did not allow for any meaningful comparison across genotype or phenotype groups (NS).
No deaths or serious adverse events were reported during the study, and no systematic, clinically relevant alterations of laboratory, vital signs or ECG data were identified. No clinically significant changes were observed with respect to body weight throughout the study.</td></tr>
<tr><td>$AUC_{0\text{-}24h}$ siponimod at day 3</td><td>x 1.7 (NS)</td><td>x 1.4 (NS)</td></tr>
<tr><td colspan="3">$AUC_{0\text{-}24h}$ siponimod at day 3 was 3.5-fold higher for *3/*3 and 2.8-fold higher for *2/*3 than after a single oral dose of siponimod 0.25 mg.</td></tr>
<tr><td colspan="3">Significance was not determined.</td></tr>
</table>

Note: Genotyping was for *2 and *3. These are the most important gene variants in this mixed Australian/French/Jordanese/American volunteer group.
</td>
<td></td>
</tr>
<tr>
<td>ref. 3
Assessment report Mayzent (siponimod) 14-11-2019

and

Huth F et al. Prediction of the impact of cytochrome P450 2C9 genotypes on the drug-drug interaction potential of siponimod with physiologically-based pharmacokinetic modeling: a comprehensive approach for drug label recommendations.</td>
<td></td>
<td>These documents confirm that Gardin 2019, Eur J Clin Pharmacol and Gardin 2019, Clin Pharmacokinet were the only studies with humans with a variant genotype used for the pharmacogenetic recommendations in the SmPC of siponimod. Thus, recommendation for *2/*2 were only based on in vitro data and modelling, and data for *1/*2 and *1/*3 were mostly based on in vitro data and modelling. In addition, recommendations are only based on pharmacokinetics, not on clinical effects.</td>
<td></td>
</tr>
</table>

Clin Pharmacol Ther 2019;106:1113-24. PubMed PMID: 31199498.			
ref. 4 SmPC Mayzent (siponimod) 14-08-23.	0 *3/*3: A *2/*2: A *1/*2: AA	Dose: Before initiation of treatment, patients must be genotyped for CYP2C9 to determine their CYP2C9 metaboliser status. In patients with a CYP2C9*3*3 genotype, siponimod should not be used. In patients with a CYP2C9*2*3 or *1*3 genotype, the recommended maintenance dose is 1 mg taken once daily (1 x 1 mg or 4 x 0.25 mg). Additional exposure of 0.25 mg on day 5 does not compromise patient safety. The recommended maintenance dose of siponimod in all other CYP-2C9 genotype patients is 2 mg. Mayzent is taken once daily. Contraindications: Patients homozygous for CYP2C9*3 (CYP2C9*3*3) genotype (poor metaboliser). Warnings: Before initiation of treatment with siponimod, patients should be genotyped for CYP2C9 to determine their CYP2C9 metaboliser status. Patients homozygous for CYP2C9*3 (CYP2C9*3*3 genotype: approximately 0.3 to 0.4% of the population) should not be treated with siponimod. Use of siponimod in these patients results in substantially elevated siponimod plasma levels. The recommended maintenance dose is 1 mg daily in patients with a CYP2C9*2*3 genotype (1.4-1.7% of the population) and in patients with a *1*3 genotype (9-12% of the population) to avoid increased exposure to siponimod. Drug interactions: The co-administration of fluconazole (moderate CYP2C9/CYP3A4 dual inhibitor) 200 mg daily at steady state and a single dose of siponimod 4 mg in healthy volunteers with a CYP2C9*1*1 genotype led to a 2-fold increase in the area under the curve (AUC) of siponimod. According to evaluation of the drug interaction potential using physiologically based pharmacokinetic (PBPK) modelling, a maximum of a 2-fold increase in the AUC of siponimod is predicted across genotypes with any type of CYP3A4 and CYP2C9 inhibitors except for patients with a CYP2C9*2*2 genotype. In CYP2C9*2*2 patients, a 2.7-fold increase in the AUC of siponimod is expected in the presence of moderate CYP2C9/CYP3A4 inhibitors. Because of an expected reduction in siponimod exposure, the appropriateness and possible benefit of the treatment should be considered when siponimod is combined with moderate CYP3A4 inducers (e.g. modafinil) or strong CYP3A4 inducers in patients with a CYP2C9*1*3 or *2*3 genotype. A significant reduction of siponimod exposure (by up to 51%) is expected under these conditions according to evaluation of the drug interaction potential using PBPK modelling. Pharmacodynamics: With continued daily dosing, the lymphocyte count continues to decrease, reaching a nadir median (90% CI) lymphocyte count of approximately 0.560 (0.271-1.08) cells/nL in a typical CYP2C9*1*1 or *1*2 non-Japanese SPMS patient, corresponding to 20-30% of baseline. Pharmacokinetics: CYP2C9 is polymorphic and the genotype influences the fractional contributions of the two oxidative metabolism pathways to overall elimination. PBPK modelling indicates a differential CYP2C9 genotype-dependent inhibition and induction of CYP3A4 pathways. With decreased CYP2C9 metabolic activity in the respective genotypes, a larger effect of the CYP3A4 perpetrators on siponimod exposure is	

<table>
<tr>
<td>ref. 4, continuation</td>
<td>*1/*3: A
*2/*3: A</td>
<td>anticipated.
The CYP2C9 genotype influences siponimod CL/F. Two population pharmacokinetic analyses indicated that CYP2C9*1*1 and *1*2 subjects behave as normal metabolisers, *2*2 and *1*3 subjects as intermediate metabolisers and *2*3 and *3*3 subjects as poor metabolisers. Compared to CYP2C9*1*1 subjects, individuals with the CYP2C9*2*2, *1*3, *2*3 and *3*3 genotypes have 20%, 35-38%, 45-48% and 74% smaller CL/F values, respectively. Siponimod exposure is therefore approximately 25%, 61%, 91% and 284% higher in CYP2C9 *2*2, *1*3, *2*3 and *3*3 subjects, respectively, as compared to *1*1 subjects (see Table).
There are other less frequent occurring polymorphisms for CYP2C9. The pharmacokinetics of siponimod have not been evaluated in such subjects. Some polymorphisms such as *5, *6, *8 and *11 are associated with decreased or loss of enzyme function. It is estimated that CYP2C9 *5, *6, *8 and *11 alleles have a combined frequency of approximately 10% in populations with African ancestry, 2% in Latinos/Hispanics and <0.4% in Caucasians and Asians.
Table CYP2C9 genotype effect on siponimod CL/F and systemic exposure
<table>
<tr><th>CYP2C9 genotype</th><th>Frequency in Caucasians</th><th>Estimated CL/F (L/h)</th><th>% of CYP2C9*1*1 CL/F</th><th>% exposure increase versus CYP2C9 *1*1</th></tr>
<tr><td colspan="5">Normal metabolisers</td></tr>
<tr><td>CYP2C9*1*1</td><td>62-65</td><td>3.1-3.3</td><td>100</td><td>-</td></tr>
<tr><td>CYP2C9*1*2</td><td>20-24</td><td>3.1-3.3</td><td>99-100</td><td>-</td></tr>
<tr><td colspan="5">Intermediate metabolisers</td></tr>
<tr><td>CYP2C9*2*2</td><td>1-2</td><td>2.5-2.6</td><td>80</td><td>25</td></tr>
<tr><td>CYP2C9*1*3</td><td>9-12</td><td>1.9-2.1</td><td>62-65</td><td>61</td></tr>
<tr><td colspan="5">Poor metabolisers</td></tr>
<tr><td>CYP2C9*2*3</td><td>1.4-1.7</td><td>1.6-1.8</td><td>52-55</td><td>91</td></tr>
<tr><td>CYP2C9*3*3</td><td>0.3-0.4</td><td>0.9</td><td>26</td><td>284</td></tr>
</table>
<u>Physician’s Checklist</u>:
The Physician’s Checklist shall contain the following key messages:
• Potential long-term safety implications in CYP2C9 poor metabolisers:
o Perform genotyping for CYP2C9 before treatment initiation to determine the siponimod maintenance dose. Test requires a DNA sample obtained via blood or saliva (buccal swab). The test identifies two variant alleles for CYP2C9: CYP2C9*2 (rs1799853, c.430C>T) and CYP2C9*3 (rs1057910, c.1075 A>C). Both are single nucleotide polymorphisms. This genotyping can be done using a Sanger sequencing method or PCR-based assay methods. For further clarifications please refer to your local laboratory.
o Do not prescribe siponimod in patients homozygous for CYP2C9*3*3.
o Adjust the maintenance dose to 1 mg in patients with CYP2C9 *2*3 or *1*3 genotypes.
• Bradyarrhythmia (including conduction defects) during treatment initiation:
o Initiate treatment with a titration pack that lasts for 5 days. Start treatment with 0.25 mg on day 1, up-titrated to the maintenance dose of 2 mg or 1 mg on day 6 based on the CYP2C9 metaboliser status.</td>
<td>Cl_{or} siponimod compared to *1/*1:
*1/*2: 99-100%
*1/*3: 62-65%
*2/*2: 80%
*2/*3: 52-55%
*3/*3: 26%</td>
</tr>
<tr>
<td>ref. 5
SmPC Mayzent (siponimod) 10-08-23, USA.</td>
<td>0</td>
<td><u>Dose</u>:
Before initiation of treatment with Mayzent, assess the following:
Test patients for CYP2C9 variants to determine CYP2C9 genotype.
An FDA-cleared or -approved test for the detection of CYP2C9 variants to direct the use of siponimod is not currently available.
After treatment titration, the recommended maintenance dosage of Mayzent is 2 mg taken orally once daily starting on Day 6 (patients</td>
<td></td>
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</table>

ref. 5, continuation		with CYP2C9 genotypes *1/*1, *1/*2, or *2/*2). Dosage adjustment is required in patients with a CYP2C9 *1/*3 or *2/*3 genotype. In patients with a CYP2C9 *1/*3 or *2/*3 genotype, after treatment titration, the recommended maintenance dosage of Mayzent is 1 mg taken orally once daily starting on Day 5. <u>Contraindications</u>: Patients who have a CYP2C9*3/*3 genotype. <u>Drug interactions</u>: Concomitant use of Mayzent and moderate (e.g., modafinil, efavirenz) or strong CYP3A4 inducers is not recommended for patients with CYP2C9*1/*3 and *2/*3 genotype. <u>Use in specific populations</u>: Before initiation of treatment with Mayzent, test patients to determine CYP2C9 genotype. Mayzent is contraindicated in patients homozygous for CYP2C9*3 (i.e., CYP2C9*3/*3 genotype) because of substantially elevated siponimod plasma levels. The *3/*3 genotype is present in approximately 0.5% of white patients and 1% of Asian patients, and is less prevalent in other racial/ethnic groups.	
	*1/*3: A *2/*3: A	Mayzent dosage adjustment is recommended in patients with CYP2C9 *1/*3 or *2/*3 genotype because of an increase in exposure to siponimod. The *1/*3 or *2/*3 genotypes are present in 2% to 20% of the population depending on ancestry.	
	IM: A PM: A	There are other less frequently occurring polymorphisms in CYP2C9. Some polymorphisms, such as *5, *6, *8, and *11, are associated with decreased or loss of enzyme function. The impact of variants other than *2 and *3 on the pharmacokinetics of siponimod has not been evaluated. It is anticipated that variants that result in loss of CYP2C9 function (e.g., *6) will have similar effects on siponimod pharmacokinetics as the *3 variant.	
	*1/*2: AA	<u>Pharmacodynamics</u>: With continued daily dosing, the lymphocyte count continues to decrease, reaching a nadir median (90% CI) lymphocyte count of approximately 0.560 (0.271-1.08) cells/nL in a typical CYP2C9*1*1 or *1*2, non-Japanese patient, corresponding to 20 to 30% of baseline.	
		<u>Pharmacokinetics</u>: CYP2C9 is polymorphic and the genotype influences the fractional contributions of the two oxidative metabolism pathways to overall elimination. Physiologically based PK modeling indicates a differential CYP2C9 genotype-dependent inhibition and induction of CYP3A4 pathways. With decreased CYP2C9 metabolic activity in the respective genotypes, a larger effect of the CYP3A4 perpetrators on siponimod exposure is anticipated. <u>Pharmacogenomics</u>: The CYP2C9 genotype has a significant impact on siponimod metabolism. After a single dose of 0.25 mg siponimod, AUC_{inf} and AUC_{last} was approximately 2-and 4-fold higher in subjects with the CYP2C9 *2/*3 and CYP2C9*3/*3 genotypes, respectively, while there was only a minor increase of C_{max} by 21% and 16%, respectively, compared to normal metabolizers (CYP2C9*1/*1). Mean half-life is prolonged in CYP2C9*2/*3 and CYP2C9*3/*3 carriers (51 hours and 126 hours, respectively).	
	*2/*2: A *3/*3: A	An apparent systemic clearance (CL/F) of about 3.11 L/h was estimated in CYP2C9 normal metabolizer (CYP2C9*1/*1 and CYP2C9 *1/*2) MS patients after multiple oral administrations of siponimod. Cl/F is 2.5, 1.9, 1.6, and 0.9 L/h in subjects with the CYP2C9*2/*2, CYP2C9 *1/*3, CYP2C9*2/*3, and CYP2C9*3/*3 genotypes respectively. The resultant increase in siponimod AUC was approximately 25%, 61%, 91%, and 285% higher in CYP2C9*2/*2, CYP2C9*1/*3, CYP2C9*2/*3, and CYP2C9*3/*3 subjects, respectively, as compared to CYP2C9 *1/*1 subjects. As the apparent clearance estimated for CYP2C9 *1*2	Cl_{or} siponimod compared to *1/*1: *1/*2: appr. 100% *1/*3: 61% *2/*2: 80% *2/*3: 51% *3/*3: 29%

ref. 5, continuation		subjects is comparable to that of CYP2C9*1/*1 subjects, similar siponimod exposure is expected for both genotypes. Variants other than *2 and *3 may also lead to decreased or loss of CYP2C9 function (e.g., *5, *6, *8, *11) and may have substrate-specific effects. The frequency of certain CYP2C9 variants differs based on ancestry. The *2 and *3 variants are more prevalent in patients of European or Asian ancestry, while *5, *6, *8, and *11 are more prevalent in individuals of African ancestry.	

Risk group	*1/*3, *2/*3 and IM with CYP2C9 inhibitors; *1/*3, *2/*3, *3/*3, IM and PM with gene variant leading to reduced CYP3A4 activity; *1/*3, *2/*3, *3/*3, IM and PM with CYP3A4 inhibitors; *1/*3, *2/*3 and IM with CYP3A4 inducers (risk for decreased effectiveness)

Comments:
-

Date of literature search: 16 October 2023.

	Genotype	Code	Gene-drug interaction	Action	Date
KNMP Pharmacogenetics Working Group decision	*1/*2	3 AA	Yes	No	6 November 2023
	*1/*3	3 AA	Yes	Yes	
	*2/*2	--	Yes	No	
	*2/*3	3 AA	Yes	Yes	
	*3/*3	3 AA	Yes	Yes	
	IM	--	Yes	Yes	
	PM	--	Yes	Yes	

Mechanism:
Siponimod is primarily metabolised by CYP2C9 (79% *in vitro*), and to a lesser extent by CYP3A4 (19% *in vitro*), to hydroxysiponimod. This metabolite is then glucuronidated. Both metabolites have a very limited pharmacological activity (hydroxysiponimod 470-fold less and siponimodglucuronide 10,000-fold less than siponimod).

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+	2+
Corresponding Clinical Implication Score:		Potentially beneficial
Score according to the SmPC:		Essential