2.6.5.10 薬物動態試験：In Vitro における代謝（続き）

2.6.5.10 Pharmacokinetics: Metabolism In Vitro (continued)

Test Article：Ixazomib

Report No.：4.2.2.4-8

Study system: Human cDNA CYP isozyme-expressing[redacted]				
Method: Human rCYP-expressing[redacted](20 pmol/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4) were incubated with CYP isozyme-specific substrates [phenacetin (CYP1A2), bupropion (CYP2B6), amodiaquine (CYP2C8), diclofenac (CYP2C9), *S*-mephenytoin (CYP2C19), dextromethorphan (CYP2D6), or testosterone (CYP3A4/5)] or ixazomib (0.1, 0.5, or 10 µmol/L) for 0 or 15 minutes in the presence of NADPH/ 3 mmol/L $MgCl_2$. The samples were then centrifuged for 10 minutes and the supernatants were analyzed using LC/MS/MS to determine the relative contributions of the different CYP isozymes to the metabolism of ixazomib.				
Time:	0 or 15 minutes			
Concentration:	0.1, 0.5, and 10 µmol/L ixazomib			
CYP Isozyme or Control	Ixazomib Concentration (µmol/L)	Relative Contributions to CYP Metabolism		
		Disappearance Rate (pmol/min/pmol)[a]	Total Measurable Metabolite Formation Rate (pmol/min/pmol)[a]	% Contribution[b]
100 mmol/L potassium phosphate buffer	0.1	0.11	0.17856	N/A
	0.5	0.23	0.15653	N/A
	10	0.00	1.65841	N/A
Control protein[c][redacted]	0.1	0.08	0.17968	N/A
	0.5	0.25	0.21587	N/A
	10	4.58	3.40890	N/A
Control protein[c]	0.1	0.24	0.30069	N/A
	0.5	0.31	0.29925	N/A
	10	7.28	6.39578	N/A

[redacted]cDNA = complementary deoxyribonucleic acid; CYP = cytochrome P450; LC/MS/MS = liquid chromatography with tandem mass spectrometry; N/A = not applicable; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate reduced tetrasodium salt hydrate; NC = not calculated;[redacted]= human CYP isozyme-expressing recombinant microsomes [redacted]USA)].

a Based on a standard curve from the parent compound.

b Mean percent contribution calculated using the results from the parent disappearance (substrate depletion) approach and the metabolite formation approach.

c Control proteins were non-CYP-expressing[redacted]

Additional information: At 0.1 and 0.5 µmol/L substrate concentrations, which are closer to clinical circulating concentrations after administration of ixazomib citrate (equivalent to 4.0 mg of ixazomib), non CYP mediated clearance was observed and seemed to play a major role in ixazomib clearance in vitro. The rate of ixazomib disappearance and the rate of formation of measurable metabolites were similar in control incubations that contained no active isozymes and ones that had active CYP isozymes present when the same protein amounts were used. These data indicate that, at clinically relevant concentrations of ixazomib, non-CYP proteins may contribute to the clearance of ixazomib, and no specific CYP isozyme predominantly contributed to the clearance of ixazomib.

2.6.5.10 薬物動態試験：In Vitro における代謝（続き）

2.6.5.10 Pharmacokinetics: Metabolism In Vitro (continued)

Test Article：Ixazomib

Report No.：4.2.2.4-8

Study system: Human cDNA CYP isozyme-expressing [redacted]				
Method: Human rCYP-expressing [redacted] (20 pmol/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4) were incubated with CYP isozyme-specific substrates [phenacetin (CYP1A2), bupropion (CYP2B6), amodiaquine (CYP2C8), diclofenac (CYP2C9), *S*-mephenytoin (CYP2C19), dextromethorphan (CYP2D6), or testosterone (CYP3A4/5)] or ixazomib (0.1, 0.5, or 10 µmol/L) for 0 or 15 minutes in the presence of NADPH/ 3 mmol/L $MgCl_2$. The samples were then centrifuged for 10 minutes and the supernatants were analyzed using LC/MS/MS to determine the relative contributions of the different CYP isozymes to the metabolism of ixazomib.				
Time:	0 or 15 minutes			
Concentration:	0.1, 0.5, and 10 µmol/L ixazomib			
CYP Isozyme or Control	Ixazomib Concentration (µmol/L)	Relative Contributions to CYP Metabolism		
		Disappearance Rate (pmol/min/pmol)[a]	Total Measurable Metabolite Formation Rate (pmol/min/pmol)[a]	% Contribution[b]
1A2	0.1	0.14	0.18661	NC
	0.5	0.40	0.18160	NC
	10	13.06	8.08620	26.1
2B6	0.1	0.13	0.18132	NC
	0.5	0.58	0.28105	NC
	10	16.72	10.65273	16.0
2C8	0.1	0.13	0.19135	NC
	0.5	0.39	0.18120	NC
	10	8.40	5.12785	6.0

[redacted]cDNA = complementary deoxyribonucleic acid; CYP = cytochrome P450; LC/MS/MS = liquid chromatography with tandem mass spectrometry; N/A = not applicable; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate reduced tetrasodium salt hydrate; NC = not calculated; [redacted] = human CYP isozyme-expressing recombinant microsomes [redacted] USA)].

a Based on a standard curve from the parent compound.

b Mean percent contribution calculated using the results from the parent disappearance (substrate depletion) approach and the metabolite formation approach.

Additional information: At 0.1 and 0.5 µmol/L substrate concentrations, which are closer to clinical circulating concentrations after administration of ixazomib citrate (equivalent to 4.0 mg of ixazomib), non CYP mediated clearance was observed and seemed to play a major role in ixazomib clearance in vitro. The rate of ixazomib disappearance and the rate of formation of measurable metabolites were similar in control incubations that contained no active isozymes and ones that had active CYP isozymes present when the same protein amounts were used. These data indicate that, at clinically relevant concentrations of ixazomib, non-CYP proteins may contribute to the clearance of ixazomib, and no specific CYP isozyme predominantly contributed to the clearance of ixazomib.

2.6.5.10 薬物動態試験：In Vitro における代謝（続き）

2.6.5.10 Pharmacokinetics: Metabolism In Vitro (continued)

Test Article：Ixazomib

Report No.：4.2.2.4-8

Study system: Human cDNA CYP isozyme-expressing [redacted]				
Method: Human rCYP-expressing [redacted] (20 pmol/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4) were incubated with CYP isozyme-specific substrates [phenacetin (CYP1A2), bupropion (CYP2B6), amodiaquine (CYP2C8), diclofenac (CYP2C9), *S*-mephenytoin (CYP2C19), dextromethorphan (CYP2D6), or testosterone (CYP3A4/5)] or ixazomib (0.1, 0.5, or 10 µmol/L) for 0 or 15 minutes in the presence of NADPH/ 3 mmol/L $MgCl_2$. The samples were then centrifuged for 10 minutes and the supernatants were analyzed using LC/MS/MS to determine the relative contributions of the different CYP isozymes to the metabolism of ixazomib.				
Time:	0 or 15 minutes			
Concentration:	0.1, 0.5, and 10 µmol/L ixazomib			
CYP Isozyme or Control	Ixazomib Concentration (µmol/L)	Relative Contributions to CYP Metabolism		
		Disappearance Rate (pmol/min/pmol)[a]	Total Measurable Metabolite Formation Rate (pmol/min/pmol)[a]	% Contribution[b]
2C9	0.1	0.14	0.19435	NC
	0.5	0.20	0.11700	NC
	10	2.54	0.70957	< 1
2C19	0.1	0.14	0.18071	NC
	0.5	0.01	0.09653	NC
	10	11.96	5.70687	4.8
2D6	0.1	0.13	0.20341	NC
	0.5	0.27	0.13417	NC
	10	11.25	5.23299	4.8

[redacted] cDNA = complementary deoxyribonucleic acid; CYP = cytochrome P450; LC/MS/MS = liquid chromatography with tandem mass spectrometry; N/A = not applicable; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate reduced tetrasodium salt hydrate; NC = not calculated; [redacted] = human CYP isozyme-expressing recombinant microsomes [redacted] USA)].

a Based on a standard curve from the parent compound.

b Mean percent contribution calculated using the results from the parent disappearance (substrate depletion) approach and the metabolite formation approach.

Additional information: At 0.1 and 0.5 µmol/L substrate concentrations, which are closer to clinical circulating concentrations after administration of ixazomib citrate (equivalent to 4.0 mg of ixazomib), non CYP mediated clearance was observed and seemed to play a major role in ixazomib clearance in vitro. The rate of ixazomib disappearance and the rate of formation of measurable metabolites were similar in control incubations that contained no active isozymes and ones that had active CYP isozymes present when the same protein amounts were used. These data indicate that, at clinically relevant concentrations of ixazomib, non-CYP proteins may contribute to the clearance of ixazomib, and no specific CYP isozyme predominantly contributed to the clearance of ixazomib.

2.6.5.10 薬物動態試験：In Vitro における代謝（続き）

2.6.5.10 Pharmacokinetics: Metabolism In Vitro (continued)

Test Article：Ixazomib

Report No.：4.2.2.4-8

Study system: Human cDNA CYP isozyme-expressing[redacted]				
Method: Human rCYP-expressing[redacted](20 pmol/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4) were incubated with CYP isozyme-specific substrates [phenacetin (CYP1A2), bupropion (CYP2B6), amodiaquine (CYP2C8), diclofenac (CYP2C9), *S*-mephenytoin (CYP2C19), dextromethorphan (CYP2D6), or testosterone (CYP3A4/5)] or ixazomib (0.1, 0.5, or 10 µmol/L) for 0 or 15 minutes in the presence of NADPH/ 3 mmol/L $MgCl_2$. The samples were then centrifuged for 10 minutes and the supernatants were analyzed using LC/MS/MS to determine the relative contributions of the different CYP isozymes to the metabolism of ixazomib.				
Time:	0 or 15 minutes			
Concentration:	0.1, 0.5, and 10 µmol/L ixazomib			
CYP Isozyme or Control	Ixazomib Concentration (µmol/L)	Relative Contributions to CYP Metabolism		
		Disappearance Rate (pmol/min/pmol)[a]	Total Measurable Metabolite Formation Rate (pmol/min/pmol)[a]	% Contribution[b]
3A4	0.1	0.12	0.17964	NC
	0.5	0.40	0.20126	NC
	10	11.71	6.00217	42.3

[redacted]cDNA = complementary deoxyribonucleic acid; CYP = cytochrome P450; LC/MS/MS = liquid chromatography with tandem mass spectrometry; N/A = not applicable; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate reduced tetrasodium salt hydrate; NC = not calculated; [redacted]= human CYP isozyme-expressing recombinant microsomes [redacted]USA)].

a Based on a standard curve from the parent compound.

b Mean percent contribution calculated using the results from the parent disappearance (substrate depletion) approach and the metabolite formation approach.

Additional information: At 0.1 and 0.5 µmol/L substrate concentrations, which are closer to clinical circulating concentrations after administration of ixazomib citrate (equivalent to 4.0 mg of ixazomib), non CYP mediated clearance was observed and seemed to play a major role in ixazomib clearance in vitro. The rate of ixazomib disappearance and the rate of formation of measurable metabolites were similar in control incubations that contained no active isozymes and ones that had active CYP isozymes present when the same protein amounts were used. These data indicate that, at clinically relevant concentrations of ixazomib, non-CYP proteins may contribute to the clearance of ixazomib, and no specific CYP isozyme predominantly contributed to the clearance of ixazomib.

2.6.5.10 薬物動態試験：In Vitro における代謝（続き）

2.6.5.10 Pharmacokinetics: Metabolism In Vitro (continued)

Test Article : [^{14}C]Ixazomib

Report No. : 4.2.2.4-9

Study system: Human liver microsomes

Method: Human liver microsomes (0.5 mg/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4) were preincubated with CYP isozyme-specific substrates [phenacetin (CYP1A2), tolbutamide (CYP2C9), *S*-mephenytoin (CYP2C19), dextromethorphan (CYP2D6), and testosterone (CYP3A4)] or [^{14}C]ixazomib (10 µmol/L) and known CYP isozyme-specific inhibitors [furafylline (CYP1A2), sulfaphenazole (CYP2C9), benzylnirvanol (CYP2C19), quinidine (CYP2D6), and azamulin (CYP3A4)] for 2 minutes at 37°C, followed by a 30 minutes reaction at 37°C, with the addition of 3 mmol/L $MgCl_2$ and 2 mmol/L NADPH. Supernatants were analyzed using reverse-phase HPLC with an online [redacted] radiochemical detector, and simultaneous equations were used to determine the relative contributions of the different CYP isozymes to the metabolism of ixazomib.

Time:	30 minutes
Concentration:	10 µmol/L [^{14}C]ixazomib

Substrate specificity of prototypical CYP isozyme inhibitors and ixazomib in human liver microsomes:

		Average percentage of inhibition of substrate metabolism (%)[a]					
CYP Isozyme	Inhibitor	Phenacetin (CYP1A2)	Tolbutamide (CYP2C9)	*S*-mephenytoin (CYP2C19)	Dextromethorphan (CYP2D6)	Testosterone (CYP3A4)	Ixazomib
1A2	Furafylline	87.0	-9.0	2.0	1.0	-11.0	22.0
2C9	Sulfaphenazole	1.0	91.0	-1.0	5.0	-6.0	10.0
2C19	Benzylnirvanol	2.0	-1.0	74.0	-1.0	-7.0	-3.8
2D6	Quinidine	-12.0	-11.0	-21.0	93.0	-11.0	5.4
3A4	Azamulin	-4.0	-26.0	-27.0	4.0	85.0	25.9
Relative percent contribution[b]		CYP1A2	CYP2C9	CYP2C19	CYP2D6	CYP3A4	
		30.7%	12.1%	< 1%	14.7%	34.2%	

[redacted] CYP = cytochrome P450; HPLC = high-performance liquid chromatography; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate reduced tetrasodium salt hydrate.

a A negative number may suggest a stimulation of the metabolism or may be a result of assay variability.

b The chemical inhibitors did not completely inhibit the metabolism of the CYP isozyme-selective substrates, and cross-reactivities were observed in some cases. To correct for this, simultaneous equations were used when calculating the percent contribution of each of the 5 major CYP isozymes to the metabolism of ixazomib.

Additional information: CYP3A4 and CYP1A2 were the major CYP isozymes contributing to the metabolism of ixazomib, followed by CYP2D6 and CYP2C9. The relative contribution of CYP2C19 was negligible.

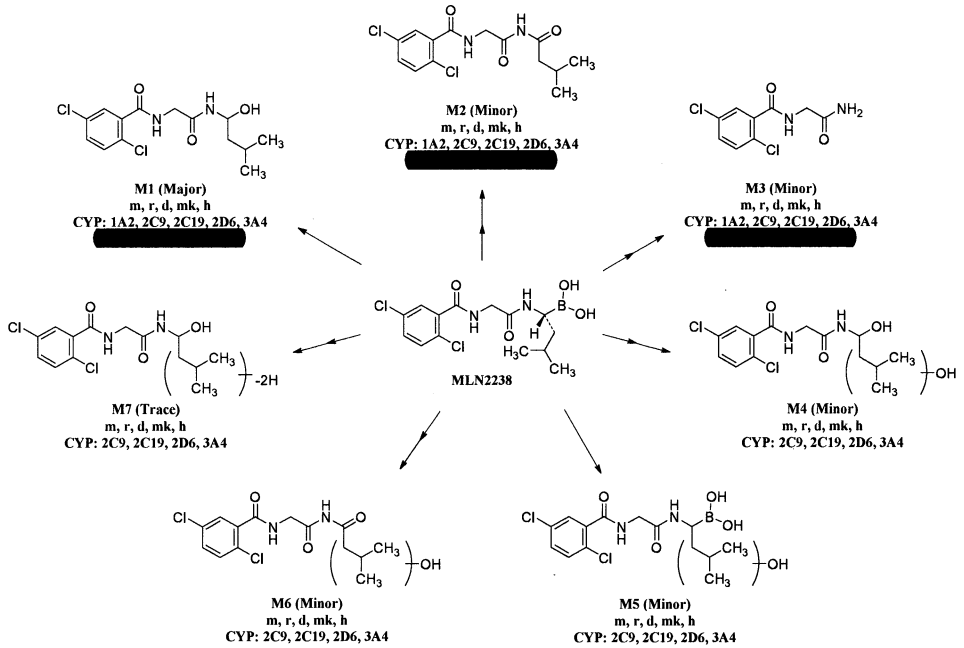
2.6.5.11 薬物動態試験：推定代謝経路

2.6.5.11 Pharmacokinetics: Possible Metabolic Pathways

Test Article: Ixazomib

Report No.: 4.2.2.4-2

Proposed Hepatic Metabolism Pathways of Ixazomib in CD-1 Mouse, Sprague-Dawley Rat, Beagle Dog, Cynomolgus Monkey, and Human Microsomal Incubations and CYP Isozyme Mapping



CYP = cytochrome P450; d = dogs; h = humans; m = mice; mk = monkeys; MLN2238 = ixazomib; r = rats.

Note: CYP isozymes, [redacted] contributing to the metabolism of ixazomib and its metabolites are noted under each metabolite structure.

Additional information: Seven major metabolites of ixazomib were identified. No metabolite unique to humans was identified in vitro. Oxidative deboronation to the hemiaminal metabolite, M1 was the major biotransformation pathway evident in the liver microsomes of all species. Other minor oxidative pathways observed included the further oxidation of M1 to M2; *N*-dealkylation of the boronic acid-containing alkyl moiety of the parent molecule to M3; a combination of these pathways with subsequent oxidation of the isopentyl moiety to generate M4, M5, and M6; and dehydrogenation of the isopentyl moiety to generate M7.

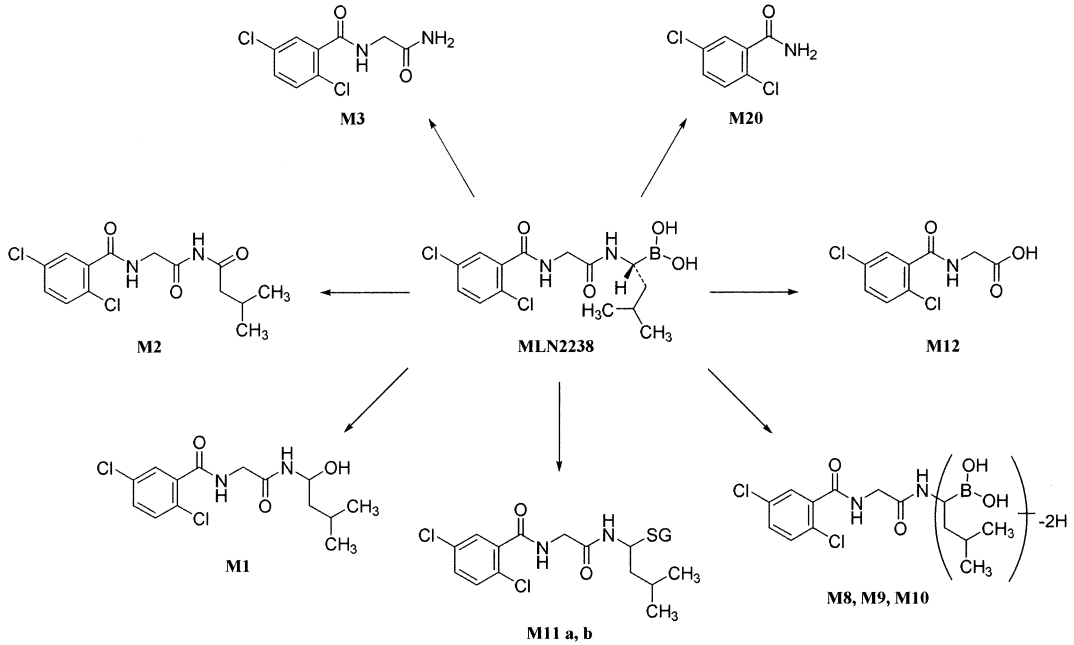
2.6.5.11 薬物動態試験：推定代謝経路（続き）

2.6.5.11 Pharmacokinetics: Possible Metabolic Pathways (continued)

Test Article: [^{14}C]Ixazomib citrate

Report No.: 4.2.2.4-3

Proposed Primary Metabolites of [^{14}C]Ixazomib Citrate Identified in Rat and Human Liver Microsomes and Hepatocytes



MLN2238 = ixazomib.

Additional information: Although the in vitro metabolism work with [^{14}C]ixazomib citrate was not performed specifically with the purpose of quantitation and complete characterization of metabolites, the radiochromatograms obtained provided relative quantitation of metabolites in vitro for comparison between rat and human hepatic metabolism. In rat liver microsomes, metabolites M1, M2, M3, M8 through M12, and M15 through M20 were observed; M1, M3, M8, M12, M15, and M16 were found to be significant metabolites. In human liver microsomes (HLMs), M1 (major), M2, M3, M8, M12, M15, M19, and M20 were observed. In rat hepatocytes, M1, M3, and M8 were the major metabolites, while M9, M10, and M11 were found in much lesser quantities. In human hepatocytes, M1 and M3 were the most dominant metabolites, while M8 was found in small quantities. The structures of metabolites M13 through M19 are tentative and are not shown in this figure.

2.6.5.12 薬物動態試験：薬物代謝酵素の誘導/阻害

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes

Test Article：Ixazomib citrate

Report No.：4.2.2.4-10

Type of study: Cytochrome P450 induction

Method: Cultured human hepatocytes were treated QD for 2 to 3 days with vehicle (DMSO), ixazomib citrate [50, 100, 500, 1000, 2000, 3000, or 5000 ng/mL for hepatocytes [redacted] (treated for 3 days); 500, 2000, or 5000 ng/mL for hepatocytes [redacted] and [redacted] (treated for 2 days)], or known human CYP isozyme inducers (omeprazole, phenobarbital, and rifampin). After treatment, cells were harvested and microsomes were isolated and analyzed by LC/MS/MS for markers of CYP isozyme-specific activity (ie, phenacetin *O*-dealkylation, bupropion hydroxylation, and testosterone 6β-hydroxylation). Immunoreactive protein levels for CYP1A2, CYP2B6, and CYP3A4 were also measured, and the potential for ixazomib citrate to cause cytotoxicity was assessed based on the release of LDH into the culture medium (a measure of cell membrane integrity).

Effects of ixazomib citrate or prototypical inducers on CYP isozyme activity:

Treatment	Conc	Phenacetin *O*-dealkylation (CYP1A2)			Bupropion Hydroxylation (CYP2B6)			Testosterone 6β-hydroxylation (CYP3A4/5)		
		Isozyme Activity (pmol/mg protein/min)[a]	Fold Increase[a]	Percent Positive Control[a]	Isozyme Activity (pmol/mg protein/min)[a]	Fold Increase[a]	Percent Positive Control[a]	Isozyme Activity (pmol/mg protein/min)[a]	Fold Increase[a]	Percent Positive Control[a]
DMSO	0.1% (v:v)	68.3 ± 35.9	1.00 ± 0.53[b]	0 ± 0	51.9 ± 29.2	1.00 ± 0.56[b]	0 ± 0	2430 ± 850	1.00 ± 0.35[b]	0 ± 0
Ixazomib citrate	500 ng/mL	33.0 ± 20.8	0.499 ± 0.218	–2.79 ± 2.54	19.2 ± 8.9	0.384 ± 0.042	–7.09 ± 3.34	886 ± 405	0.361 ± 0.058	–12.8 ± 7.1
Ixazomib citrate	2000 ng/mL	27.1 ± 22.1	0.404 ± 0.263	–3.21 ± 2.71	21.0[c]	0.332[c]	–5.97[c]	676 ± 642	0.274 ± 0.195	–13.9 ± 6.1
Ixazomib citrate	5000 ng/mL	32.4[c]	0.471[c]	–4.30[c]	22.2[c]	0.343[c]	–5.78[c]	893[c]	0.351[c]	–14.5[c]
Omeprazole	100 µmol/L	1600 ± 910	28.1 ± 17.2	100 ± 0	308 ± 186	6.30 ± 3.42	N/A	5060 ± 2120	2.53 ± 1.90	N/A
Phenobarbital	750 µmol/L	123 ± 46	1.93 ± 0.58	N/A	612 ± 387	11.5 ± 5.8	100 ± 0	15100 ± 4600	6.78 ± 2.77	N/A
Rifampin	10 µmol/L	99.5 ± 37.5	1.54 ± 0.25	N/A	369 ± 205	7.05 ± 2.13	N/A	15700 ± 4700	7.07 ± 2.93	100 ± 0

Conc = concentration; CYP = cytochrome P450; DMSO = dimethyl sulfoxide; LC/MS/MS = liquid chromatography with tandem mass spectrometry; LDH = lactate dehydrogenase; N/A = not applicable; QD = once daily; v:v = volume-to-volume ratio.

Note: DMSO was the vehicle control. Omeprazole, phenobarbital, and rifampin were the positive controls for CYP1A2, CYP2B6, and CYP3A4/5, respectively.1 ng/mL ixazomib citrate = 1.934 nmol/L (molecular weight of ixazomib citrate is 517.12).

a Values are the mean ± standard deviation (SD) of 3 determinations (human hepatocyte preparations [redacted] and [redacted], unless otherwise indicated.

b Mean ± percent coefficient of variation (%CV).

c n = 2. Data from [redacted] were not included because of the cytotoxicity resulting from the high concentration of ixazomib citrate.

追記:次ページに記載

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib citrate

Report No.：4.2.2.4-10

Type of study: Cytochrome P450 induction
Method: Cultured human hepatocytes were treated QD for 2 to 3 days with vehicle (DMSO), ixazomib citrate [50, 100, 500, 1000, 2000, 3000, or 5000 ng/mL for hepatocytes [redacted] (treated for 3 days); 500, 2000, or 5000 ng/mL for hepatocytes [redacted] and [redacted] (treated for 2 days)], or known human CYP isozyme inducers (omeprazole, phenobarbital, and rifampin). After treatment, cells were harvested and microsomes were isolated and analyzed by LC/MS/MS for markers of CYP isozyme-specific activity (ie, phenacetin *O*-dealkylation, bupropion hydroxylation, and testosterone 6β-hydroxylation). Immunoreactive protein levels for CYP1A2, CYP2B6, and CYP3A4 were also measured, and the potential for ixazomib citrate to cause cytotoxicity was assessed based on the release of LDH into the culture medium (a measure of cell membrane integrity).

Additional information: Treatment of hepatocytes with up to 5000 ng/mL ixazomib citrate resulted in, on average, a 60%, 67%, and 73% decrease in CYP1A2, CYP2B6, and CYP3A4/5 activity, respectively. There was also a corresponding decrease in CYP1A2 (except for [redacted] where there was little to no change in immunoreactive protein levels), CYP2B6, and CYP3A4 immunoreactive protein levels after treatment with up to 5000 ng/mL ixazomib citrate. The cause of these decreases in CYP activity and protein levels are unknown but may be attributed to hepatotoxicity corresponding to prolonged exposure to ixazomib citrate. Under conditions where prototypical inducers caused anticipated increases in CYP isozyme activity, treatment with up to 5000 ng/mL ixazomib citrate did not induce CYP1A2, CYP2B6, and CYP3A4/5 activity or corresponding immunoreactive protein levels. The cause of these decreases in CYP isozyme activity and protein levels are unknown, but may be a result of cytotoxicity corresponding to prolonged exposure to ixazomib citrate in cultured human hepatocytes, as evidenced by an apparent concentration-dependent increase in LDH release resulting from deteriorating cell membrane integrity.

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib

Report No.：4.2.2.4-11

Type of study: Potential reversible inhibition of CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4/5 by ixazomib			
Method: HLMs (0.25 mg/mL with NADPH/$MgCl_2$) were incubated with CYP isozyme-specific substrates and ixazomib (0 to 30 µmol/L). Prototypical CYP isozyme inhibitors were included as positive controls. Supernatants were analyzed by LC/MS/MS for the amount of metabolite formed for each CYP isozyme.			
CYP Isozyme	Substrate	IC_{50} (µmol/L)	
		Ixazomib	Prototypical Inhibitor
1A2	Phenacetin	> 30	> 10 (furafylline)
2B6	Bupropion	> 30	0.34 (ticlopidine)
2C8	Paclitaxel	> 30	2.55 (quercetin)
2C9	Diclofenac	> 30	2.35 (tienilic acid)
2C19	*S*-Mephenytoin	> 30	4.24 (ticlopidine)
2D6	Dextromethorphan	> 30	0.09 (quinidine)
3A4/5	Testosterone	> 30	> 10 (troleandomycin)
3A4/5	Testosterone	> 30	0.12 (ketoconazole)
3A4/5	Midazolam	> 30	> 10 (troleandomycin)
3A4/5	Midazolam	> 30	0.12 (ketoconazole)

CYP = cytochrome P450; IC_{50} = concentration producing 50% inhibition; HLM = human liver microsome; LC/MS/MS = liquid chromatography with tandem mass spectrometry; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate reduced tetrasodium salt hydrate.

Additional information: Ixazomib is not a reversible inhibitor of major human hepatic CYP isozymes.

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib

Report No.：4.2.2.4-6

Type of study: Potential reversible inhibition of CYP1A2, 2C9, 2C19, 2D6, and 3A4/5 by ixazomib

Method: HLMs (0.5 mg/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4) were incubated with CYP isozyme-specific substrates and ixazomib (0 to 30 µmol/L). Prototypical CYP isozyme inhibitors were included as positive controls. Supernatants were analyzed by LC/MS/MS for the amount of metabolite formed for each CYP isozyme.

CYP Isozyme	Substrate	IC_{50} (µmol/L)	
		Ixazomib	Prototypical Inhibitor
1A2	Phenacetin	> 30	1.82 (furafylline)
2C9	Tolbutamide	> 30	0.69 (sulfaphenazole)
2C19	*S*-Mephenytoin	> 30	4.25 (omeprazole)
2D6	Dextromethorphan	> 30	0.135 (quinidine)
3A4/5	Testosterone	> 30	0.191 (ketoconazole)
3A4/5	Midazolam	> 30	0.132 (ketoconazole)

CYP = cytochrome P450; IC_{50} = concentration producing 50% inhibition; HLM = human liver microsomes; LC/MS/MS = liquid chromatography with tandem mass spectrometry.

Additional information: Ixazomib is not a reversible inhibitor of major human hepatic CYP isozymes. The estimated inhibition constant (Ki) was > 15 µmol/L, assuming the inhibition was competitive.

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib citrate

Report No.：4.2.2.4-12

Type of study: Potential reversible inhibition of CYP2B6 and 2C8 by ixazomib citrate		
Method: HLMs (0.1 mg/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4) were incubated with CYP isozyme-specific substrates and ixazomib (0 to 30 µmol/L). Supernatants were analyzed by LC/MS/MS for the amount of metabolite formed for each CYP isozyme.		
CYP Isozyme	Substrate	Ixazomib Citrate IC_{50} (µmol/L)
2B6	Bupropion	> 30
2C8	Amodiaquine	> 30

CYP = cytochrome P450; IC_{50} = concentration producing 50% inhibition; HLM = human liver microsome; LC/MS/MS = liquid chromatography with tandem mass spectrometry.

Additional information: Ixazomib citrate is not a reversible inhibitor of CYP2B6 or 2C8 at concentrations up to 30 µmol/L, the highest concentration tested.

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib

Report No.：4.2.2.4-11

Type of study: Potential time-dependent inhibition of CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, or 3A4/5 by ixazomib

Method 1: No preincubation assay: HLMs (0.25 mg/mL with NADPH/$MgCl_2$) were incubated with CYP isozyme-specific substrates and ixazomib (0 to 30 µmol/L) and 7 major CYP substrates for 5 (CYP2C9, 2D6, and 3A4) or 20 (CYP1A2, 2B6, 2C8, and 2C19) minutes. After the final incubation was complete, the supernatant was analyzed by LC/MS/MS.
Method 2: Preincubation with NADPH assay: HLMs (0.333[a] mg/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4) were preincubated with ixazomib (0 to 40 µmol/L) and NADPH in a total volume of 75 µL for 30 minutes and then mixed with 7 major CYP substrate solutions with a final mixture volume of 100 µL (HLM was 0.25 mg/mL in final mixture solution) for another 5 (CYP2C9, 2D6, and 3A4) or 20 (CYP1A2, 2B6, 2C8, and 2C19) minutes. After the final incubation was complete, the supernatant was analyzed by LC/MS/MS.

CYP Isozyme	Substrate	IC_{50} (µmol/L)				
		Ixazomib		Prototypical Inhibitor[b]		IC_{50} Shift Fold[c]
		No Preincubation	Preincubation with NADPH	No Preincubation	Preincubation with NADPH	
1A2	Phenacetin	> 30	> 30	> 10	3.92	> 2.6
2B6	Bupropion	> 30	> 30	0.34	0.15	2.3
2C8	Paclitaxel	> 30	> 30	2.55	3.76	0.7
P2C9	Diclofenac	> 30	> 30	2.35	0.17	13.8
2C19	*S*-Mephenytoin	> 30	> 30	4.24	1.82	2.3
2D6	Dextromethorphan	> 30	> 30	0.09	0.10	0.9
3A4/5	Testosterone	> 30	> 30	0.12	0.15	0.8
3A4/5	Midazolam	> 30	> 30	0.12	0.15	0.8
3A4/5	Testosterone	> 30	> 30	> 10	0.47	> 21.3
3A4/5	Midazolam	> 30	> 30	> 10	0.95	> 10.5

CYP = cytochrome P450; HLM = human liver microsome; IC_{50} = concentration producing 50% inhibition; LC/MS/MS = liquid chromatography with tandem mass spectrometry; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate reduced tetrasodium salt hydrate.

a Except in the CYP2C8 assay, in which the final concentration was 0.338 mg/mL.

b Prototypical inhibitors for CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, or 3A4/5 with testosterone or midazolam were furafylline, ticlopidine, quercetin, tienilic acid, ticlopidine, quinidine, ketoconazole, and troleandomycin, respectively.

c IC_{50} Shift Fold = (IC_{50} of no preincubation) / (IC_{50} of preincubation with NADPH); if shift fold is > 2, it indicates a time-dependent inhibition. Experiment was conducted in duplicate.

Additional information: Ixazomib is not a time-dependent inhibitor of CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, or 3A4/5.

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib

Report No.：4.2.2.4-6

Type of study: Potential CYP3A4/5 time-dependent inhibition by ixazomib

Method: Ixazomib (0 to 30 µmol/L) was incubated with HLMs (2.5 mg/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4), with or without NADPH for selected time intervals and then diluted 10-fold to incubate with testosterone and NADPH for another 10 minutes. After the final incubation was complete, the supernatant was analyzed by LC/MS/MS for concentrations of 6β-hydroxytestosterone. TAO was used as a positive control for inhibition of CYP3A4/5.

Preincubation Duration (min)	Percentage of CYP3A4/5 Activity Remaining (%)				
	Ixazomib (µmol/L)				TAO (µmol/L)
	0	1	10	30	10
Without NADPH					
0	100	91	89	86	78
15	100	87	112	118	93
30	100	75	84	94	86
With NADPH					
0	100	98	91	102	92
15	100	89	98	86	54
30	100	97	100	98	48

CYP = cytochrome P450; HLM = human liver microsome; LC/MS/MS = liquid chromatography with tandem mass spectrometry; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate reduced tetrasodium salt hydrate; TAO = troleandomycin.

Additional information: Ixazomib is not a time-dependent inhibitor of CYP3A4/5.

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib citrate

Report No.：4.2.2.4-12

Type of study: Potential time-dependent inhibition of CYP1A2, 2B6, 2C8, 2C9, 2C19, or 2D6 by ixazomib citrate

Method: Ixazomib citrate (0 to 30 µmol/L) was incubated with HLMs (2.5 mg/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4), with or without NADPH for selected time intervals and then diluted 10-fold to incubate with CYP isozyme-specific substrates [phenacetin (CYP1A2), bupropion (CYP2B6), amodiaquine (CYP2C8), diclofenac (CYP2C9), *S*-Mephenytoin (CYP2C19), or dextromethorphan (CYP2D6)] and NADPH for another 10 minutes (8 minutes for CYP2C8). After the final incubation was complete, the supernatant was analyzed by LC/MS/MS for concentrations of individual CYP isozyme metabolites. TAO was used as a positive time-dependent control for inhibition, with testosterone as a substrate.

Preincubation Duration (min)	Percentage of CYP1A2 Activity Remaining (%)		
	Ixazomib citrate (µmol/L)		
	0	3	30
Without NADPH			
0	100	85	94
15	100	98	95
30	100	96	96
With NADPH			
0	100	92	94
15	100	91	92
30	100	92	101
Preincubation Duration (min)	Percentage of CYP2B6 Activity Remaining (%)		
	Ixazomib citrate (µmol/L)		
	0	3	30
Without NADPH			
0	100	92	94
15	100	99	101
30	100	93	94
With NADPH			
0	100	101	102
15	100	94	92
30	100	105	100

CYP = cytochrome P450; HLM = human liver microsome; LC/MS/MS = liquid chromatography with tandem mass spectrometry; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate, reduced tetrasodium salt hydrate; TAO = troleandomycin.

Additional information: Ixazomib citrate is not a time-dependent inhibitor of CYP1A2, 2B6, 2C8, 2C9, 2C19, or 2D6.

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib citrate

Report No.：4.2.2.4-12

Type of study: Potential time-dependent inhibition of CYP1A2, 2B6, 2C8, 2C9, 2C19, or 2D6 by ixazomib citrate

Method: Ixazomib citrate (0 to 30 µmol/L) was incubated with HLMs (2.5 mg/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4), with or without NADPH for selected time intervals and then diluted 10-fold to incubate with CYP isozyme-specific substrates [phenacetin (CYP1A2), bupropion (CYP2B6), amodiaquine (CYP2C8), diclofenac (CYP2C9), *S*-Mephenytoin (CYP2C19), or dextromethorphan (CYP2D6)] and NADPH for another 10 minutes (8 minutes for CYP2C8). After the final incubation was complete, the supernatant was analyzed by LC/MS/MS for concentrations of individual CYP isozyme metabolites. TAO was used as a positive time-dependent control for inhibition, with testosterone as a substrate.

Preincubation Duration (min)	Percentage of CYP2C8 Activity Remaining (%)		
	Ixazomib citrate (µmol/L)		
	0	3	30
Without NADPH			
0	100	100	102
15	100	82	82
30	100	106	105
With NADPH			
0	100	116	111
15	100	86	80
30	100	90	99
Preincubation Duration (min)	Percentage of CYP2C9 Activity Remaining (%)		
	Ixazomib citrate (µmol/L)		
	0	3	30
Without NADPH			
0	100	91	87
15	100	95	99
30	100	94	100
With NADPH			
0	100	92	92
15	100	94	97
30	100	104	99

CYP = cytochrome P450; HLM = human liver microsome; LC/MS/MS = liquid chromatography with tandem mass spectrometry; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate, reduced tetrasodium salt hydrate; TAO = troleandomycin.

Additional information: Ixazomib citrate is not a time-dependent inhibitor of CYP1A2, 2B6, 2C8, 2C9, 2C19, or 2D6.

2.6.5.12 薬物動態試験：代謝酵素の誘導/阻害（続き）

2.6.5.12 Pharmacokinetics: Induction/Inhibition of Drug-Metabolizing Enzymes (continued)

Test Article：Ixazomib citrate

Report No.：4.2.2.4-12

Type of study: Potential time-dependent inhibition of CYP1A2, 2B6, 2C8, 2C9, 2C19, or 2D6 by ixazomib citrate

Method: Ixazomib citrate (0 to 30 µmol/L) was incubated with HLMs (2.5 mg/mL in 0.1 mol/L potassium phosphate buffer, pH 7.4), with or without NADPH for selected time intervals and then diluted 10-fold to incubate with CYP isozyme-specific substrates [phenacetin (CYP1A2), bupropion (CYP2B6), amodiaquine (CYP2C8), diclofenac (CYP2C9), *S*-Mephenytoin (CYP2C19), or dextromethorphan (CYP2D6)] and NADPH for another 10 minutes (8 minutes for CYP2C8). After the final incubation was complete, the supernatant was analyzed by LC/MS/MS for concentrations of individual CYP isozyme metabolites. TAO was used as a positive time-dependent control for inhibition, with testosterone as a substrate.

Preincubation Duration (min)	Percentage of CYP2C19 Activity Remaining (%)		
	Ixazomib citrate (µmol/L)		
	0	3	30
Without NADPH			
0	100	95	97
15	100	109	102
30	100	101	94
With NADPH			
0	100	102	96
15	100	106	101
30	100	96	91
Preincubation Duration (min)	Percentage of CYP2D6 Activity Remaining (%)		
	Ixazomib citrate (µmol/L)		
	0	3	30
Without NADPH			
0	100	92	94
15	100	99	98
30	100	106	100
With NADPH			
0	100	89	92
15	100	101	97
30	100	108	98

CYP = cytochrome P450; HLM = human liver microsome; LC/MS/MS = liquid chromatography with tandem mass spectrometry; NADPH = beta-nicotinamide adenine dinucleotide 2′-phosphate, reduced tetrasodium salt hydrate; TAO = troleandomycin.

Additional information: Ixazomib citrate is not a time-dependent inhibitor of CYP1A2, 2B6, 2C8, 2C9, 2C19, or 2D6.

2.6.5.13 薬物動態試験：累積排泄

2.6.5.13 Pharmacokinetics: Excretion

Test Article: [^{14}C]Ixazomib citrate

Species		Sprague-Dawley rats				Beagle Dogs		
Sex (M/F)/Number of Animals		M/3				M/4		
Feeding Condition		Fasted				Fasted		
Administration Article		[^{14}C]Ixazomib citrate				[^{14}C]Ixazomib citrate		
Vehicle/Formulation		1% propylene glycol in [redacted]				1% propylene glycol in [redacted]		
Method of Administration		Oral gavage				Oral gavage		
Dose		0.8 mg/kg as ixazomib citrate				0.15 mg/kg as ixazomib		
		Cumulative Excretion Ratio （% of the Dose）				Cumulative Excretion Ratio （% of the Dose）		
Route of Excretion		Urine	Feces	Cage wash	Total	Urine	Feces	Total
Time	0-6 hr	6.32	----	----	NC	----	----	----
	0-24 hr	14.14	24.10	----	38.23	16.1	13.1	29.2
	0-48 hr	18.24	48.76	----	67.00	22.9	22.2	45.1
	0-72 hr	20.75	53.03	----	73.78	27.1	26.5	53.5
	0-96 hr	22.46	55.07	----	77.52	29.9	29.8	59.6
	0-120 hr	23.63	56.66	----	80.29	32.0	32.6	64.6
	0-144 hr	24.44	57.75	----	82.19	33.6	35.1	68.7
	0-168 hr	25.18	58.55	----	84.22	35.0	36.7	71.7
	0-192 hr	25.63	59.15	----	84.78	----	----	----
	0-216 hr	26.05	59.65	----	85.70	----	----	----
	0-240 hr	26.42	60.22	0.56	87.20	----	----	----
Report No.		4.2.2.3-6				4.2.2.3-7		

----: Not collected. NC: Not calculated.

2.6.5.14 薬物動態試験：胆汁中排泄

2.6.5.14 Pharmacokinetics: Biliary excretion

Test Article: [^{14}C]Ixazomib citrate

Report No.: 4.2.2.3-6

Species		Sprague-Dawley rats				
Sex (M/F)/Number of Animals		M/3				
Feeding Condition		Fasted				
Administration Article		[^{14}C]Ixazomib citrate				
Vehicle/Formulation		1% propylene glycol in [redacted]				
Method of Administration		Oral gavage				
Dose		0.8 mg/kg as ixazomib citrate				
		Cumulative Excretion Ratio （% of the Dose）				
Route of Excretion		Urine	Feces	Bile	Cage wash	Total
Time	0-6 hr	2.03	----	12.76	----	NC
	0-24 hr	9.44	7.72	17.27	----	34.42
	0-48 hr	15.97	24.35	20.46	----	60.78
	0-72 hr	20.40	35.41	22.33	2.57	80.70

----: Not collected. NC: Not calculated.

2.6.5.15 薬物動態試験：薬物相互作用

2.6.5.15 Pharmacokinetics: Drug-Drug Interactions

2.6.5.15.1 Uptake clearance by hepatocytes

Test Article: [^{14}C]Ixazomib citrate

Report No.: 4.2.2.6-1

Type of Study		In vitro uptake clearance by hepatocytes		
Test System		Cryo-preserved human hepatocytes		
Test Article		[^{14}C]Ixazomib citrate		
Concentration of Test Article		1 µmol/L		
Inhibitor (Concentration)		Cyclosporin A (10 µmol/L)		
Method of Detection		LSC		
		Uptake Clearance (µL/30 sec/mg protein)		
Test Compounds	Concentration (µmol/L)	[redacted]	[redacted]	[redacted]
Ixazomib[a]	1	39.0	48.2	60.7
Ixazomib[a] + Cyclosporin A	1 + 10	37.7	47.0	60.4

a Ixazomib citrate rapidly hydrolyzes to form ixazomib in the buffer solution; therefore, all results are expressed in terms of ixazomib.

2.6.5.15 薬物動態試験：薬物相互作用（続き）

2.6.5.15 Pharmacokinetics: Drug-Drug Interactions (continued)

2.6.5.15.2 P-gp inhibition

Test Article: Ixazomib citrate

Report No.: 4.2.2.6-2

Type of Study		In vitro inhibitory effects of ixazomib and quinidine on [^{3}H]digoxin transport across Caco-2 cells			
Test System		Caco-2 cells			
Test Article		Ixazomib citrate			
Concentration of Test Article		1-100 μmol/L			
Substrate (Concentration)		[^{3}H]Digoxin (3 μmol/L)			
Positive Control		Quinidine			
Method of Detection		LSC			
Inhibitor		Papp ($\times 10^{-6}$ cm/sec)		Papp Ratio	% of Control
Compounds	Concentration (μmol/L)	Apical to Basal	Basal to Apical		
Ixazomib[a]	0	0.564	12.8	22.7	100.0
	1	0.602	12.5	20.8	91.2
	3	0.627	12.9	20.6	90.3
	10	0.657	12.8	19.5	85.3
	30	0.694	12.5	18.0	78.3
	100	0.706	12.8	18.1	78.8
Quinidine	30	3.87	5.67	1.5	2.3

Papp: apparent permeability coefficient. All experiments were performed in triplicate.

a Ixazomib citrate rapidly hydrolyzes to ixazomib in the buffer solution; therefore, all results are expressed in terms of ixazomib.

2.6.5.15 薬物動態試験：薬物相互作用（続き）

2.6.5.15 Pharmacokinetics: Drug-Drug Interactions (continued)

2.6.5.15.3 Inhibitory effect on transportes

Test Article: Ixazomib citrate

Report No.: 4.2.2.6-3

Type of Study	In vitro inhibition of BCRP-expressing cells by ixazomib
Test System	BCRP-expressing cells: LLC-PK1 cells
Test Article	Ixazomib citrate
Concentration of Test Article	0-100 μmol/L
Substrate (Concentration)	[^{3}H]Prazosin (0.01 μmol/L)
Positive Control (Concentration)	Ko143 (1 μmol/L)
Method of Detection	LSC

Inhibitor		Control Cells			BCRP-Expressing Cells			Corrected Papp Ratio	% of Control
Compounds	Concentration (μmol/L)	Papp ($\times10^{-6}$ cm/sec)		Papp Ratio	Papp ($\times10^{-6}$ cm/sec)		Papp Ratio		
		Apical to Basal	Basal to Apical		Apical to Basal	Basal to Apical			
Ixazomib[a]	0	49.9	44.2	0.9	8.27	107	12.9	14.3	100.0
	1	47.2	47.8	1.0	9.66	117	12.1	12.1	83.5
	3	49.8	49.5	1.0	9.39	102	10.9	10.9	74.4
	10	46.9	48.0	1.0	9.24	111	12.0	12.0	82.7
	30	47.0	47.1	1.0	9.06	117	12.9	12.9	89.5
	100	45.3	47.7	1.1	10.7	107	10.0	9.1	60.9
Ko143	1	47.6	47.1	1.0	48.6	52.3	1.1	1.1	0.8

Papp: apparent permeability coefficient. All experiments were performed in triplicate.

a Ixazomib citrate rapidly hydrolyzes to form ixazomib in the buffer solution.

2.6.5.15 薬物動態試験：薬物相互作用（続き）

2.6.5.15 Pharmacokinetics: Drug-Drug Interactions (continued)

2.6.5.15.3 Inhibitory effect on transportes (continued)

Test Article: Ixazomib citrate

Report No.: 4.2.2.6-3

Type of Study		In vitro inhibition of OCT, OATs, OATPs, and MATEs-expressing cells by ixazomib						
Test System		OAT1/3-expressing cells: S_2 cells; OCT2, OATP1B1/1B3, and MATE1/2K-expressing cells: HEK293 cells						
Test Article		Ixazomib citrate						
Concentration of Test Article		0-10 μmol/L						
Substrate (Concentration)		OCT2 and MATE1/2K: [^{14}C]Metformin (10 μmol/L), OAT1: [^{3}H]PAH (1 μmol/L), OAT3: [^{3}H] E3S (50 nmol/L), OATP1B1/1B3: [^{3}H]$E_2$17βG (50 nmol/L)						
Positive Control (Concentration)		OCT2: Quinidine (300 μmol/L), OAT1/3: Probenecid (100 μmol/L), OATP1B1/1B3: Rifampicin (10 μmol/L), MATE1: Cimetidine (10 μmol/L), MATE2-K: Cimetidine (100 μmol/L)						
Method of Detection		LSC						
Inhibitor		% of Controls						
Compounds	Concentration (μmol/L)	OCT2	OAT1	OAT3	OATP1B1	OATP1B3	MATE1	MATE2-K
Ixazomib[a]	0	100.0	100.0	100.0	100.0	100.0	100.0	100.0
	0.1	89.9	84.1	84.4	110.0	94.2	103.1	93.5
	0.3	90.6	94.7	80.4	109.8	109.6	101.2	99.2
	1	94.7	92.1	91.9	112.3	96.9	108.4	89.7
	3	88.4	96.0	78.0	99.9	111.2	104.7	72.3
	10	90.0	83.6	83.7	110.7	99.1	101.4	77.9
Positive control	-	3.7	6.4	10.6	8.2	3.8	10.4	0.0[b]

E3S: Estrone3-sulfate. MATE: Multidrug and toxin extrusion transporters. OAT: Organic anion transporter. OATP: Organic anion-transporting polypeptide. OCT: Organic cation transporter. PAH: *p*-Aminohippurate.

a Ixazomib citrate rapidly hydrolyzes to form ixazomib in the buffer solution.

b % control was a negative value, expressed as 0.0.

2.6.5.15 薬物動態試験：薬物相互作用（続き）

2.6.5.15 Pharmacokinetics: Drug-Drug Interactions (continued)

2.6.5.15.4 Effects of carfilzomib on blood cell partitioning

Test Article: Ixazomib citrate

Report No.: 4.2.2.6-4

Type of Study	In vitro evaluation of the effects of carfilzomib on blood cell partitioning		
Test System	Whole blood from human		
Test Article	Ixazomib citrate		
Concentration of Test Article	10, 50, 500 ng/mL as ixazomib citrate		
Additive Article (Concentration)	Carfilzomib (0 to 10 µmol/L)		
Analyte	Ixazomib		
Method of Detection	LC/MS/MS		
Ixazomib Citrate Concentration (ng/mL)	Carfilzomib Concentration (µmol/L)	Incubation Time (min)	Ixazomib Concentration (ng/mL)
10	0-10	30	9.80-15.5
		60	7.61-11.3
50	0-10	30	40.2-61.7
		60	35.9-66.5
500	0-10	30	520-751
		60	427-596

2.6.5.16 薬物動態試験：その他

2.6.5.16 Pharmacokinetics: Other

Not available.