第 2 部（モジュール 2）
CTD の概要

2.6.5 薬物動態試験概要表

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略語

略語	略さない用語	日本語訳
A-B	Apical to basolateral	頂端膜側から基底膜側
Ac-di-sol	Croscarmellose sodium	クロスカルメロースナトリウム
$AUC_{0\text{-}24}$	Area under the plasma concentration vs time curve from 0 to 24 h	0 時間から 24 時間までの血漿中濃度－時間曲線下面積
$AUC_{0\text{-}inf}$	Area under the plasma concentration vs time curve extrapolated to infinity	0 時間から無限大時間まで外挿した血漿中濃度－時間曲線下面積
B-A	Basolateral to apical	基底膜側から頂端膜側
BCRP	Breast cancer resistance protein	乳がん耐性タンパク
BLQ	Below limit of quantification	定量下限未満
CHO	Chinese hamster ovary	チャイニーズハムスター卵巣
CL	Systemic plasma clearance	全身血漿クリアランス
C_{max}	Maximum plasma concentration	最高血漿中濃度
CV	Coefficient of variation	変動係数
CV-1	Green monkey kidney cells	ミドリザル腎細胞
CYP	Cytochrome P450 (isomer)	チトクローム P450（分子種；標準体はタンパク、斜体は遺伝子）
DMSO	Dimethyl sulfoxide	ジメチルスルホキシド

略語	略さない用語	日本語訳
EC_{50}	Concentration that gives half-maximal response	最大効果の 50%の効果をもたらす濃度
EDTA	Ethylenediaminetetraacetic acid	エチレンジアミン四酢酸
E_{max}	Maximal effect	最大誘導作用
f	Female	雌
F	Oral bioavailability	経口バイオアベイラビリティ
GLP	Good Laboratory Practice	医薬品の安全性に関する非臨床試験の実施の基準
HEK	Human embryonic kidney	ヒト胎児腎臓
HLM	Human liver microsomes	ヒト肝ミクロソーム
HP-β-CD	Hydroxypropyl-β-cyclodextrin	ヒドロキシプロピル-β-シクロデキストリン
HPLC	High-performance liquid chromatography	高速液体クロマトグラフィー
IC_{50}	Concentration that causes 50% inhibition	50%阻害濃度
i.v.	Intravenous	静脈内
K_i	Inhibition constant	阻害定数
K_I	Inactivation constant	最大不活性化速度の 50%の速度をもたらす濃度
k_{inact}	Rate constant of enzyme inactivation	最大不活性化速度定数
LC-MS/MS	Liquid chromatography coupled to tandem mass spectrometry	液体クロマトグラフィータンデム質量分析
LLOQ	Lower limit of quantification	定量下限
m	Male	雄
M	Metabolite	代謝物
MATE	Multidrug and toxin extrusion protein	多剤・毒性化合物排出タンパク
MC	Methyl cellulose	メチルセルロース
MDCK	Madin-Darby canine kidney	メイディンーダービーイヌ腎臓
MDR	Multidrug resistance protein	多剤耐性タンパク
mRNA	Messenger ribonucleic acid	メッセンジャーリボ核酸
n	Number	例数
ND	Not determined	確定せず
OAT	Organic anion transporter	有機アニオントランスポーター
OATP	Organic anion transporting polypeptide	有機アニオン輸送ポリペプチド
OCT	Organic cation transporter	有機カチオントランスポーター
P450	Cytochrome P450 (enzyme)	チトクローム P450（酵素）
P_{app}	Apparent absorptive permeability coefficient	みかけの透過係数
PCR	Polymerase chain reaction	ポリメラーゼ連鎖反応
PEG	Polyethylene glycol	ポリエチレングリコール
P-gp	Permeability glycoprotein	P-糖タンパク

略語	略さない用語	日本語訳
PK	Pharmacokinetic(s)	薬物動態
p.o.	Oral	経口
PXR	Pregnane X receptor	プレグナン X 受容体
QCL/M/H	Quality control low/middle/high	低/中/高濃度 QC
RD	Relative deviation	相対標準偏差
RT	Room temperature	室温
SD	Standard deviation	標準偏差
T	Cytotoxic	細胞毒性
$t_{1/2}$	Apparent terminal half-life	みかけの最終相半減期
t_{max}	Time to reach maximum plasma concentration	最高血漿中濃度到達時間
UGT	Uridine 5’-diphospho-glucuronyltransferase	ウリジン 5’-二リン酸-グルクロン酸転移酵（標準体はタンパク、斜体は遺伝子）
V_{ss}	Volume of distribution at steady state	定常状態における分布容積
v/v	Volume/Volume	容量/容量
w/w	Weight/Weight	重量/重量

1. 薬物動態試験：一覧表

Test Articles: ダリドレキサント, ACT-776537, ACT-776063, ACT-1016-3307

Type of Study	Test System	Method of Administration	Testing Facility	Report Number	Location in CTD
Absorption					
Single-dose pharmacokinetics					
Intravenous and oral pharmacokinetics	Rat (Wistar)	i.v. (0.1, 0.3, 1 mg/kg) p.o. (3, 10, 30 mg/kg)	Actelion Pharmaceuticals Ltd	B-13.116	モジュール 4.2.2.2.1
Intravenous and oral pharmacokinetics	Dog (Beagle)	i.v. (0.3, 1, 3 mg/kg) p.o. (1, 3, 10, 30 mg/kg)	Actelion Pharmaceuticals Ltd	B-13.116	モジュール 4.2.2.2.1
Multiple-dose pharmacokinetics					
Oral pharmacokinetics	Rat (Wistar)	p.o. (100, 300, 1000 mg/kg/day)	[redacted]	T-13.221	モジュール 4.2.3.2.5
Oral pharmacokinetics	Dog (Beagle)	p.o. (30, 100, 300/200 mg/kg/day)	[redacted]	T-13.220	モジュール 4.2.3.2.9
Distribution					
Plasma protein binding	Mouse (CD-1), rat (Wistar), rabbit (New Zealand White), dog (Beagle), human	*In vitro* (0.5 - 200 µg/mL)	[redacted]	B-13.202	モジュール 4.2.2.3.1
Plasma protein binding	Human serum albumin and α1-acid glycoprotein	*In vitro* (1.5, 5, 15 µg/mL)	[redacted]	B-13.202	モジュール 4.2.2.3.1
Plasma protein binding of ACT-776537, ACT-776063 and ACT-1016-3307	Mouse (CD-1), rat (Wistar), dog (Beagle), human	*In vitro* (0.5, 1.5, 5 µg/mL human; 1.5, 5, 15 µg/mL mouse, rat and dog)	[redacted]	B-17.061	モジュール 4.2.2.3.2

1. 薬物動態試験：一覧表

Test Articles: ダリドレキサント, ACT-776537, ACT-776063, ACT-1016-3307

Type of Study	Test System	Method of Administration	Testing Facility	Report Number	Location in CTD
Blood/plasma partitioning	Mouse (CD-1), rat (Wistar), rabbit (New Zealand White), dog (Beagle), human	*In vitro* (0.5, 20 µg/mL)	[redacted]	B-13.202	モジュール 4.2.2.3.1
Whole-body autoradiography	Rat (Wistar and Lister-Hooded)	p.o. (10 mg/kg)	[redacted]	B-16.005	モジュール 4.2.2.3.3
Milk excretion	Rat (Wistar)	p.o. (30 mg/kg)	[redacted]	B-20.022	モジュール 4.2.2.3.4
Metabolism					
Metabolic profiling *in vitro*	Mouse, rat, rabbit, dog, monkey and human liver microsomes and hepatocytes	*In vitro*	Actelion Pharmaceuticals Ltd	B-14.019	モジュール 4.2.2.4.2
Metabolic profiling mouse	Mouse (CD-1 and B6C3F1) liver microsomes and hepatocytes	*In vitro*	Idorsia Pharmaceuticals Ltd	B-16.062	モジュール 4.2.2.4.4
	Mouse (CD-1 and B6C3F1)	p.o. (100 mg/kg) for 1 and 14 days	Idorsia Pharmaceuticals Ltd	B-16.062	モジュール 4.2.2.4.4
Metabolic profiling rat	Rat (Wistar)	i.v. (1 mg/kg) p.o. (10 mg/kg)	Actelion Pharmaceuticals Ltd	B-14.054	モジュール 4.2.2.4.3
Metabolizing enzyme identification	Human liver microsomes, recombinant P450	*In vitro*	Actelion Pharmaceuticals Ltd	B-15.073	モジュール 4.2.2.4.5
Metabolite identification	Rat bile and feces	i.v. (1 mg/kg) p.o. (10 mg/kg)	Idorsia Pharmaceuticals Ltd	B-15.042	モジュール 4.2.2.4.1
Metabolite identification	Human plasma	p.o. (25 mg)	Idorsia Pharmaceuticals Ltd	B-15.042	モジュール 4.2.2.4.1

1. 薬物動態試験：一覧表

Test Articles: ダリドレキサント, ACT-776537, ACT-776063, ACT-1016-3307

Type of Study	Test System	Method of Administration	Testing Facility	Report Number	Location in CTD
Metabolite identification	Human hepatocytes	*In vitro*	Idorsia Pharmaceuticals Ltd	B-15.042	モジュール 4.2.2.4.1
Excretion					
Excretion in urine, feces and bile	Rat (Wistar)	i.v. (1 mg/kg) p.o. (10 mg/kg)	Actelion Pharmaceuticals Ltd	B-14.054	モジュール 4.2.2.4.3
Pharmacokinetic Drug Interactions					
P450 inhibition	Human liver microsomes, recombinant CYP2C19	*In vitro*	Actelion Pharmaceuticals Ltd	B-14.018	モジュール 4.2.2.6.1
P450 induction	Human hepatocytes	*In vitro*	Actelion Pharmaceuticals Ltd	B-14.064	モジュール 4.2.2.6.4
P450 induction by ACT-776537, ACT-776063 and ACT-1016-3307	Human hepatocytes	*In vitro*	Idorsia Pharmaceuticals Ltd	B-17.063	モジュール 4.2.2.6.5
PXR activation	CV-1 cells expressing human PXR	*In vitro*	Actelion Pharmaceuticals Ltd	B-14.064	モジュール 4.2.2.6.4
PXR activation by ACT-776537, ACT-776063 and ACT-1016-3307	CV-1 cells expressing human PXR	*In vitro*	Idorsia Pharmaceuticals Ltd	B-17.063	モジュール 4.2.2.6.5

1. 薬物動態試験：一覧表

Test Articles: ダリドレキサント, ACT-776537, ACT-776063, ACT-1016-3307

Type of Study	Test System	Method of Administration	Testing Facility	Report Number	Location in CTD
Drug transporter inhibition	CHO cells expressing OATP1B1, OATP1B3, OAT1 or OCT2; MDCKII cells expressing P-gp/MDR1; HEK293 cells expressing OAT3, OCT1, MATE1 or MATE2-K; Sf9 membrane vesicles expressing BCRP; CHO, HEK293 and MDCKII wild-type cells	*In vitro*	Actelion Pharmaceuticals Ltd	B-14.061	モジュール 4.2.2.6.6
OATP1B1 and OATP1B3 inhibition	CHO cells expressing OATP1B1 or OATP1B3; CHO wild-type cells	*In vitro*	Idorsia Pharmaceuticals Ltd	B-19.069	モジュール 4.2.2.6.8
OATP1B1, OATP1B3, P-gp and BCRP substrate	CHO cells expressing OATP1B1 or OATP1B3; MDCKII cells expressing P-gp/MDR1 or BCRP; CHO and MDCKII wild-type cells	*In vitro*	Idorsia Pharmaceuticals Ltd	B-17.067	モジュール 4.2.2.3.5
P450 inhibition or time-dependent P450 inhibition by daridorexant, ACT-776537, ACT-776063 and ACT-1016-3307	Human liver microsomes, recombinant CYP2C19	*In vitro*	Idorsia Pharmaceuticals Ltd	B-17.055	モジュール 4.2.2.6.2

1. 薬物動態試験：一覧表

Test Articles: ダリドレキサント, ACT-776537, ACT-776063, ACT-1016-3307

Type of Study	Test System	Method of Administration	Testing Facility	Report Number	Location in CTD
Time-dependent CYP1A2 and CYP2B6 inhibition by daridorexant, ACT-776537, ACT-776063 and ACT-1016-3307	Human liver microsomes	*In vitro*	Idorsia Pharmaceuticals Ltd	B-20.034	モジュール 4.2.2.6.3
Drug transporter inhibition by ACT-776537, ACT-776063 and ACT-1016-3307	CHO cells expressing OATP1B1, OATP1B3, OAT1 or OCT2; MDCKII cells expressing P-gp/MDR1, MATE1 or MATE2-K; HEK293 cells expressing OAT3 or OCT1; Sf9 membrane vesicles expressing BCRP; CHO, HEK293 and MDCKII wild-type cells; Sf9 control vesicles	*In vitro*	Idorsia Pharmaceuticals Ltd	B-17.049	モジュール 4.2.2.6.7
P-gp and BCRP substrate study with ACT-776537, ACT-776063 and ACT-1016-3307	MDCKI cells expressing P-gp/MDR1 or BCRP	*In vitro*	[redacted]	B-19.052	モジュール 4.2.2.3.6

1. 薬物動態試験：一覧表

Test Articles: ダリドレキサント, ACT-776537, ACT-776063, ACT-1016-3307

Type of Study	Test System	Method of Administration	Testing Facility	Report Number	Location in CTD
cyp and *ugt* induction in rat and dog 4-week toxicity studies	Rat and dog livers	p.o. (rat: 100, 300, 1000 mg/kg; dog: 30, 100, 300/200 mg/kg)	Actelion Pharmaceuticals Ltd	B-14.065	モジュール 4.2.2.4.6

ACT-541468 = daridorexant; ACT-776537 = M1; ACT-776063 = M3; ACT-1016-3307 = M10.

BCRP = breast cancer resistance protein; CHO = Chinese hamster ovary; CV-1 = green monkey kidney cells; CYP/*CYP* = cytochrome P450 (isomer, protein/*gene*); HEK = human embryonic kidney; i.v. = intravenous; MATE = multidrug and toxin extrusion protein; MDCK = Madin-Darby canine kidney; MDR = multidrug resistance protein; OAT = organic anion transporter; OATP = organic anion transporting polypeptide; OCT = organic cation transporter; P450 = cytochrome P450 (enzyme); P-gp = permeability glycoprotein; p.o. = oral; PXR = pregnane X receptor; *UGT* = uridine 5'-diphospho-glucuronosyltransferase (isomer, *gene*).

2. 分析法及びバリデーション試験

Test Articles: ダリドレキサント, ACT-776537, ACT-776063, ACT-1016-3307

Species	Sample Matrix	Analytes	Calibration Range (ng/mL)	Assay Method	GLP Compliance	Report No.	Location in CTD
Mouse	Plasma	ACT-541468	10.0 - 10000	LC-MS/MS	Yes	BA-17.031	モジュール 4.2.2.1.3
		ACT-776537	10.0 - 10000				
		ACT-776063	10.0 - 10000				
		ACT-1016-3307	10.0 - 10000				
Rat	Plasma	ACT-541468	1.00 - 1000 and 10.0 - 10000	LC-MS/MS	Yes	BA-13.013	モジュール 4.2.2.1.1
Rat	Plasma	ACT-541468	10.0 - 10000	LC-MS/MS	Yes	BA-17.030	モジュール 4.2.2.1.2
		ACT-776537	10.0 - 10000				
		ACT-776063	10.0 - 10000				
		ACT-1016-3307	10.0 - 10000				
Rat	Plasma	ACT-541468	1.00 - 1000 and 10.0 - 10000	LC-MS/MS	Yes	BA-19.100	モジュール 4.2.2.1.8
Rabbit	Plasma	ACT-541468	10.0 - 10000	LC-MS/MS	Yes	BA-14.018	モジュール 4.2.2.1.4
Rabbit	Plasma	ACT-541468	10.0 - 10000	LC-MS/MS	Yes	BA-18.023	モジュール 4.2.2.1.7
		ACT-776537	10.0 - 10000				
		ACT-776063	10.0 - 10000				
		ACT-1016-3307	10.0 - 10000				
Dog	Plasma	ACT-541468	1.00 - 1000 and 10.0 - 10000	LC-MS/MS	Yes	BA-13.014	モジュール 4.2.2.1.5
Dog	Plasma	ACT-541468	10.0 - 10000	LC-MS/MS	Yes	BA-17.029	モジュール 4.2.2.1.6
		ACT-776537	10.0 - 10000				
		ACT-776063	10.0 - 10000				
		ACT-1016-3307	10.0 - 10000				

ACT-541468 = daridorexant; ACT-776537 is also known as metabolite M1; ACT-776063 is also known as metabolite M3; ACT-1016-3307 is also known as metabolite M10.
GLP = Good Laboratory Practice; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry.

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Mouse/ Plasma	ACT-541468	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 4.7% to 11.3%; QCL/M/H: 1.8% to 6.2% Precision (% CV) inter-batch: LLOQ: 9.7%; QCL/M/H: 3.1% to 4.4% Accuracy (% RD) intra-batch: LLOQ: –9.4% to 0.6%; QCL/M/H: –5.4% to 1.8% Accuracy (% RD) inter-batch: LLOQ: –5.8%; QCL/M/H: –3.5% to –2.0%	Yes	BA-17.031	モジュール 4.2.2.1.3

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rat / Plasma	ACT-541468	1.00 - 1000 ng/mL	Protein precipitation	LC-MS/MS [1)]	Precision (% CV) intra-batch: LLOQ: 3.9% to 16.0%; QCL: 3.1% to 9.7%; QCM: 2.5% to 11.7%; QCH: 1.7% to 10.9% Precision (% CV) inter-batch: LLOQ: 15.4%; QCL: 7.6%; QCM: 8.4%; QCH: 7.7% Accuracy (% RD) intra-batch: LLOQ: –8.8% to 13.3%; QCL: –1.2% to 8.2%; QCM: 1.1% to 3.6%; QCH: –9.5% to –0.4% Accuracy (% RD) inter-batch: LLOQ: 5.5%; QCL: 3.1%; QCM: 2.3%; QCH: –5.7%	Yes	BA-13.013	モジュール 4.2.2.1.1

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rat / Plasma	ACT-541468	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS [1)]	Precision (% CV) intra-batch: LLOQ: 2.7% to 4.0%; QCL: 2.3% to 4.3%; QCM: 2.4% to 3.5%; QCH: 1.0% to 4.4% Precision (% CV) inter-batch: LLOQ: 8.3%; QCL: 5.0%; QCM: 4.0%; QCH: 3.7% Accuracy (% RD) intra-batch: LLOQ: –8.8% to 7.7%; QCL: –6.2% to 2.9%; QCM: –2.2% to 3.6%; QCH: –5.3% to –1.9% Accuracy (% RD) inter-batch: LLOQ: –2.1%; QCL: –0.9%; QCM: 1.6%; QCH: –3.3%	Yes	BA-13.013	モジュール 4.2.2.1.1

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rat / Plasma	ACT-541468	1.00 - 1000 ng/mL	Protein precipitation	LC-MS/MS [2)]	Precision (% CV) intra-batch: LLOQ: 3.2% to 6.3%; QCL: 1.8% to 3.9%; QCM: 1.5% to 3.6%; QCH: 1.3% to 3.6% Precision (% CV) inter-batch: LLOQ: 7.3%; QCL: 5.3%; QCM: 2.8%; QCH: 4.7% Accuracy (% RD) intra-batch: LLOQ: –10.2% to 4.0%; QCL: –5.8% to 5.1%; QCM: –3.4% to 2.0%; QCH: –9.9% to 1.3% Accuracy (% RD) inter-batch: LLOQ: –1.0%; QCL: –0.4%; QCM: –1.0%; QCH: –2.1%	Yes	BA-13.013	モジュール 4.2.2.1.1

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rat / Plasma	ACT-541468	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 4.3% to 15.4%; QCL/M/H: 2.4% to 13.8% Precision (% CV) inter-batch: LLOQ: 11.5%; QCL/M/H: 3.5% to 9.6% Accuracy (% RD) intra-batch: LLOQ: –11.2% to 1.9%; QCL/M/H: –9.3% to 4.4% Accuracy (% RD) inter-batch: LLOQ: –3.3%; QCL/M/H: –6.2% to 0.1%	Yes	BA-17.030	モジュール 4.2.2.1.2
Rat / Plasma	ACT-541468	1.00 - 1000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 8.0%; QCL/M/H: 2.3% to 2.4% Accuracy (% RD) intra-batch: LLOQ: 5.6%; QCL/M/H: 4.0% to 4.9%	Yes	BA-19.100	モジュール 4.2.2.1.8

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rat / Plasma	ACT-541468	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 4.4%; QCL/M/H: 2.4% to 8.5% Accuracy (% RD) intra-batch: LLOQ: 3.0%; QCL/M/H: 3.0% to 10.0%	Yes	BA-19.100	モジュール 4.2.2.1.8
Rabbit / Plasma	ACT-541468	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 4.4% to 5.3%; QCL: 1.6% to 2.9%; QCM: 2.0% to 3.3%; QCH: 1.8% to 5.9% Precision (% CV) inter-batch: LLOQ: 5.4%; QCL: 3.1%; QCM: 2.9%; QCH: 3.7% Accuracy (% RD) intra-batch: LLOQ: –5.3% to 0.9%; QCL: –0.5% to 4.7%; QCM: –0.1% to 2.3%; QCH: 0.8% to 2.4% Accuracy (% RD) inter-batch: LLOQ: –1.5%; QCL: 1.7%; QCM: 1.4%; QCH: 1.5%	Yes	BA-14.018	モジュール 4.2.2.1.4

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rabbit/ Plasma	ACT-541468	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 4.7% to 7.6%; QCL/M/H: 3.3% to 9.5% Precision (% CV) inter-batch: LLOQ: 8.5%; QCL/M/H: 5.5% to 10.5% Accuracy (% RD) intra-batch: LLOQ: –8.8% to 5.7%; QCL/M/H: –9.6% to 7.8% Accuracy (% RD) inter-batch: LLOQ: –1.1%; QCL/M/H: –2.0% to 1.3%	Yes	BA-18.023	モジュール 4.2.2.1.7
Dog / Plasma	ACT-541468	1.00 - 1000 ng/mL	Protein precipitation	LC-MS/MS [1)]	Precision (% CV) intra-batch: LLOQ: 7.5%; QCL: 15.7%; QCM: 3.2%; QCH: 4.0% Accuracy (% RD) intra-batch: LLOQ: –12.7%; QCL: 3.6%; QCM: 0.6%; QCH: –7.5%	Yes	BA-13.014	モジュール 4.2.2.1.5

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Dog / Plasma	ACT-541468	1.00 - 1000 ng/mL	Protein precipitation	LC-MS/MS [2)]	Precision (% CV) intra-batch: LLOQ: 4.8% to 6.8%; QCL: 2.5% to 3.7%; QCM: 1.8% to 2.3%; QCH: 1.5% to 1.6% Precision (% CV) inter-batch: LLOQ: 7.1%; QCL: 4.9%; QCM: 2.8%; QCH: 2.6% Accuracy (% RD) intra-batch: LLOQ: –2.0% to 8.5%; QCL: –4.8% to 4.1%; QCM: –1.6% to 2.5%; QCH: –3.9% to 1.2% Accuracy (% RD) inter-batch: LLOQ: 2.7%; QCL: 0.7%; QCM: –0.2%; QCH: –1.4%	Yes	BA-13.014	モジュール 4.2.2.1.5

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Dog / Plasma	ACT-541468	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 3.3% to 5.1%; QCL: 2.0% to 4.2%; QCM: 2.5% to 5.0%; QCH: 1.7% to 2.8% Precision (% CV) inter-batch: LLOQ: 4.8%; QCL: 4.2%; QCM: 4.3%; QCH: 3.6% Accuracy (% RD) intra-batch: LLOQ: –13.1% to –9.0%; QCL: –9.5% to –3.7%; QCM: –2.1% to 3.4%; QCH: –8.8% to –2.7% Accuracy (% RD) inter-batch: LLOQ: –11.5%; QCL: –5.7%; QCM: 1.5%; QCH: –5.9%	Yes	BA-13.014	モジュール 4.2.2.1.5

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Dog / Plasma	ACT-541468	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 3.8% to 8.4%; QCL/M/H: 1.5% to 9.2% Precision (% CV) inter-batch: LLOQ: 6.2%; QCL/M/H: 3.5% to 6.9% Accuracy (% RD) intra-batch: LLOQ: –7.7% to –1.2%; QCL/M/H: –10.9% to 2.6% Accuracy (% RD) inter-batch: LLOQ: –4.9%; QCL/M/H: –8.4% to –2.8%	Yes	BA-17.029	モジュール 4.2.2.1.6

ACT-541468 = daridorexant

1) Method V2
2) Method V3

CV = coefficient of variation; GLP = Good Laboratory Practice; H = high; L = low; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; LLOQ = lower limit of quantification; M = middle; QC = quality control; RD = relative deviation.

2. 分析法及びバリデーション試験

Test article: ACT-776537

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Mouse/ Plasma	ACT-776537	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 1.5% to 9.9%; QCL/M/H: 1.8% to 3.6% Precision (% CV) inter-batch: LLOQ: 7.0%; QCL/M/H: 3.1% to 4.5% Accuracy (% RD) intra-batch: LLOQ: –8.9% to –1.5%; QCL/M/H: –6.5% to 2.7% Accuracy (% RD) inter-batch: LLOQ: –5.9%; QCL/M/H: –4.2% to –1.8%	Yes	BA-17.031	モジュール 4.2.2.1.3

2. 分析法及びバリデーション試験

Test article: ACT-776537

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rat / Plasma	ACT-776537	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 2.6% to 6.1%; QCL/M/H: 2.3% to 8.4% Precision (% CV) inter-batch: LLOQ: 9.0%; QCL/M/H: 3.6% to 5.5% Accuracy (% RD) intra-batch: LLOQ: –11.8% to 5.7%; QCL/M/H: –5.1% to 2.8% Accuracy (% RD) inter-batch: LLOQ: –4.3%; QCL/M/H: –2.9% to 0.2%	Yes	BA-17.030	モジュール 4.2.2.1.2

2. 分析法及びバリデーション試験

Test article: ACT-776537

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rabbit/ Plasma	ACT-776537	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 5.0% to 7.9%; QCL/M/H: 1.2% to 5.9% Precision (% CV) inter-batch: LLOQ: 6.6%; QCL/M/H: 3.0% to 7.0% Accuracy (% RD) intra-batch: LLOQ: 0.5% to 7.2%; QCL/M/H: –7.7% to 5.8% Accuracy (% RD) inter-batch: LLOQ: 4.0%; QCL/M/H: –2.0% to 3.1%	Yes	BA-18.023	モジュール 4.2.2.1.7

2. 分析法及びバリデーション試験

Test article: ACT-776537

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Dog / Plasma	ACT-776537	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 3.0% to 9.0%; QCL/M/H: 1.1% to 5.7% Precision (% CV) inter-batch: LLOQ: 8.0%; QCL/M/H: 4.2% to 6.2% Accuracy (% RD) intra-batch: LLOQ: –6.3% to 5.9%; QCL/M/H: –8.7% to 1.7% Accuracy (% RD) inter-batch: LLOQ: –0.8%; QCL/M/H: –4.2% to –3.1%	Yes	BA-17.029	モジュール 4.2.2.1.6

ACT-776537 is also known as metabolite M1.

CV = coefficient of variation; GLP = Good Laboratory Practice; H = high; L = low; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; LLOQ = lower limit of quantification; M = middle; QC = quality control; RD = relative deviation.

2. 分析法及びバリデーション試験

Test article: ACT-776063

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Mouse/ Plasma	ACT-776063	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 4.9% to 15.2%; QCL/M/H: 1.4% to 7.5% Precision (% CV) inter-batch: LLOQ: 12.0%; QCL/M/H: 2.6% to 6.0% Accuracy (% RD) intra-batch: LLOQ: –13.8% to 0.6%; QCL/M/H: –9.8% to 5.4% Accuracy (% RD) inter-batch: LLOQ: –7.1%; QCL/M/H: –7.4% to 0.8%	Yes	BA-17.031	モジュール 4.2.2.1.3

2. 分析法及びバリデーション試験

Test article: ACT-776063

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rat / Plasma	ACT-776063	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 7.5% to 14.2%; QCL/M/H: 2.4% to 6.2 % Precision (% CV) inter-batch: LLOQ: 9.9%; QCL/M/H: 5.3% to 7.7% Accuracy (% RD) intra-batch: LLOQ: –8.8% to –5.4%; QCL/M/H: –4.0% to 12.4% Accuracy (% RD) inter-batch: LLOQ: –7.3%; QCL/M/H: –0.7% to 4.9%	Yes	BA-17.030	モジュール 4.2.2.1.2

2. 分析法及びバリデーション試験

Test article: ACT-776063

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rabbit/ Plasma	ACT-776063	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 3.2% to 14.8%; QCL/M/H: 1.2% to 11.0% Precision (% CV) inter-batch: LLOQ: 11.3%; QCL/M/H: 4.9% to 9.4% Accuracy (% RD) intra-batch: LLOQ: –3.5% to 11.5%; QCL/M/H: –5.6% to 8.1% Accuracy (% RD) inter-batch: LLOQ: 5.2%; QCL/M/H: –1.9% to 3.1%	Yes	BA-18.023	モジュール 4.2.2.1.7

2. 分析法及びバリデーション試験

Test article: ACT-776063

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Dog / Plasma	ACT-776063	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 6.7% to 10.4%; QCL/M/H: 2.3% to 8.4% Precision (% CV) inter-batch: LLOQ: 8.3%; QCL/M/H: 4.4% to 6.4% Accuracy (% RD) intra-batch: LLOQ: –3.7% to 1.5%; QCL/M/H: –7.9% to 3.9% Accuracy (% RD) inter-batch: LLOQ: –0.9%; QCL/M/H: –3.9% to –1.6%	Yes	BA-17.029	モジュール 4.2.2.1.6

ACT-776063 is also known as metabolite M3.

CV = coefficient of variation; GLP = Good Laboratory Practice; H = high; L = low; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; LLOQ = lower limit of quantification; M = middle; QC = quality control; RD = relative deviation.

2. 分析法及びバリデーション試験

Test article: ACT-1016-3307

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Mouse/ Plasma	ACT-1016-3307	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 3.2% to 8.4%; QCL/M/H: 2.0% to 6.6% Precision (% CV) inter-batch: LLOQ: 8.2%; QCL/M/H: 4.4% to 5.1% Accuracy (% RD) intra-batch: LLOQ: –8.1% to 4.7%; QCL/M/H: –6.6% to 4.7% Accuracy (% RD) inter-batch: LLOQ: –0.5%; QCL/M/H: –2.8% to 2.4%	Yes	BA-17.031	モジュール 4.2.2.1.3

2. 分析法及びバリデーション試験

Test article: ACT-1016-3307

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rat/ Plasma	ACT-1016-3307	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 7.0% to 10.9%; QCL/M/H: 2.1% to 10.1% Precision (% CV) inter-batch: LLOQ: 10.0%; QCL/M/H: 5.6% to 9.1% Accuracy (% RD) intra-batch: LLOQ: –5.8% to 3.6%; QCL/M/H: –6.2% to 8.5% Accuracy (% RD) inter-batch: LLOQ: –1.9%; QCL/M/H: –5.1% to 2.9%	Yes	BA-17.030	モジュール 4.2.2.1.2

2. 分析法及びバリデーション試験

Test article: ACT-1016-3307

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Rabbit/ Plasma	ACT-1016-3307	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 4.3% to 8.9%; QCL/M/H: 3.2% to 14.1% Precision (% CV) inter-batch: LLOQ: 12.5%; QCL/M/H: 5.6% to 9.9% Accuracy (% RD) intra-batch: LLOQ: –14.0% to 10.6%; QCL/M/H: –9.2% to 1.5% Accuracy (% RD) inter-batch: LLOQ: –1.4%; QCL/M/H: –4.7% to –3.4%	Yes	BA-18.023	モジュール 4.2.2.1.7

2. 分析法及びバリデーション試験

Test article: ACT-1016-3307

Species / Sample Matrix	Analytes	Calibration Range	Type of Assay	Method Used	Noteworthy Findings (Precision and Accuracy Validation)	GLP Compliance	Report No.	Location in CTD
Dog/ Plasma	ACT-1016-3307	10.0 - 10000 ng/mL	Protein precipitation	LC-MS/MS	Precision (% CV) intra-batch: LLOQ: 2.1% to 7.6%; QCL/M/H: 1.5% to 6.8% Precision (% CV) inter-batch: LLOQ: 7.2%; QCL/M/H: 4.0% to 6.2% Accuracy (% RD) intra-batch: LLOQ: –12.9% to –0.6%; QCL/M/H: –11.0% to –0.7% Accuracy (% RD) inter-batch: LLOQ: –6.3%; QCL/M/H: –7.1% to –4.3%	Yes	BA-17.029	モジュール 4.2.2.1.6

ACT-1016-3307 is also known as metabolite M10.

CV = coefficient of variation; GLP = Good Laboratory Practice; H = high; L = low; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; LLOQ = lower limit of quantification; M = middle; QC = quality control; RD = relative deviation.

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Mouse	Plasma (EDTA)	4 freeze-and-thaw cycles at –20 °C		25.0	101.5	3	BA-17.031	モジュール 4.2.2.1.3
				8000	93.1	3		
		4 freeze-and-thaw cycles at –80 °C		25.0	93.7	3		
				8000	96.0	3		
		RT	26 hours	25.0	97.5	3		
				8000	99.4	3		
		4 °C	35 hours	25.0	92.7	3		
				8000	97.6	3		
		–20 °C	375 days	25.0	99.9	3		
				8000	93.5	3		
		–80 °C	375 days	25.0	95.7	3		
				8000	94.8	3		
Rat	Plasma (EDTA)	3 freeze-and-thaw cycles at –20 °C		2.00	100.5	3	BA-13.013	モジュール 4.2.2.1.1
				25.0	99.3	3		
				8000	91.5	3		
		3 freeze-and-thaw cycles at –80 °C		2.00	94.7	3		
				25.0	94.1	3		
				8000	92.3	3		
		RT	4 or 5 hours	2.00	99.0	3		
				25.0	97.2	3		
				8000	96.1	3		
		ice	4 hours	25.0	96.5	3		
				8000	93.6	3		

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Rat	Plasma (EDTA)	4 °C	62 hours [1)]	2.00	95.4	3	BA-13.013	モジュール 4.2.2.1.1
			98 hours [2)]		107.9	3		
			41 hours	25.0	101.3	3		
			133 hours [2)]	800	96.0	3		
			41 hours	8000	92.8	3		
		–20 °C	158 days	2.00	98.4	3		
			336 days		106.9	3		
			29 days	25.0	98.8	3		
			160 days		100.0	3		
			378 days		104.5	3		
			29 days	8000	90.7	3		
			160 days		91.7	3		
			378 days		94.6	3		
		–80 °C	158 days	2.00	93.7	3		
			336 days		107.4	3		
			29 days	25.0	99.3	3		
			160 days		97.6	3		
			378 days		107.5	3		
			29 days	8000	92.7	3		
			160 days		88.2	3		
			378 days		91.8	3		

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Rat	Plasma (EDTA)	5 freeze-and-thaw cycles at −20 °C		25.0	103.1	3	BA-17.030	モジュール 4.2.2.1.2
				8000	96.5	3		
		5 freeze-and-thaw cycles at −80 °C		25.0	101.3	3		
				8000	93.3	3		
		RT	24 hours	25.0	105.6	3		
				8000	101.1	3		
		4 °C	41 hours	25.0	101.1	3		
				8000	104.1	3		
		−20 °C	734 days	25.0	111.1	3		
				8000	99.9	3		
		−80 °C	734 days	25.0	114.0	3		
				8000	100.3	3		
Rat	Plasma (EDTA)	RT	24 or 26 hours	2.00	107.7	3	BA-19.100	モジュール 4.2.2.1.8
				8000	96.0	3		
Rabbit	Plasma (EDTA)	3 freeze-and-thaw cycles at –20 °C		25.0	105.6	3	BA-14.018	モジュール 4.2.2.1.4
				8000	99.6	3		
		3 freeze-and-thaw cycles at –80 °C		25.0	102.0	3		
				8000	98.9	3		
		RT	6 hours	25.0	103.3	3		
				8000	98.4	3		
		4 °C	69 hours	25.0	100.5	3		
				8000	90.6	3		

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Rabbit	Plasma (EDTA)	–20 °C	33 days	25.0	105.2	3	BA-14.018	モジュール 4.2.2.1.4
				8000	98.9	3		
		–80 °C	33 days	25.0	107.9	3		
				8000	103.9	3		
Rabbit	Plasma (EDTA)	4 freeze-and-thaw cycles at –20 °C		25.0	93.9	3	BA-18.023	モジュール 4.2.2.1.7
				8000	87.5	3		
		4 freeze-and-thaw cycles at –80 °C		25.0	95.7	3		
				8000	93.9	3		
		RT	73 hours	25.0	90.3	3		
				8000	98.0	3		
		4 °C	89 hours	25.0	103.6	3		
				8000	92.9	3		
		–20 °C	386 days	25.0	91.2	3		
				8000	91.3	3		
		–80 °C	386 days	25.0	90.5	3		
				8000	91.4	3		
Dog	Plasma (EDTA)	3 freeze-and-thaw cycles at –20 °C		2.00	105.3	3	BA-13.014	モジュール 4.2.2.1.5
				25.0	95.9	3		
				8000	95.1	3		
		3 freeze-and-thaw cycles at –80 °C		2.00	102.3	3		
				25.0	96.1	3		
				8000	93.7	3		

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	RT	5 hours	2.00	99.2	3	BA-13.014	モジュール 4.2.2.1.5
				25.0	101.2	3		
				8000	91.0	3		
		4 °C	33 hours [2)]	2.00	103.4	3		
			89 hours	25.0	100.4	3		
			33 hours [2)]	800	97.8	3		
			89 hours	8000	97.1	3		
		–20 °C	152 days	2.00	96.4	3		
			322 days		102.5	3		
			134 days	25.0	86.8	3		
			351 days		104.4	3		
			134 days	8000	86.5	3		
			351 days		95.8	3		
		–80 °C	152 days	2.00	103.2	3		
			322 days		106.0	3		
			134 days	25.0	100.0	3		
			351 days		103.6	3		
			134 days	8000	87.0	3		
			351 days		94.6	3		

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	5 freeze-and-thaw cycles at –20 °C		25.0	86.8	3	BA-17.029	モジュール 4.2.2.1.6
				8000	97.1	3		
				50000 [3)]	100.1	3		
		5 freeze-and-thaw cycles at –80 °C		25.0	90.1	3		
				8000	98.1	3		
				50000 [3)]	106.9	3		
		RT	70 hours	25.0	88.1	3		
				8000	96.8	3		
				50000 [3)]	97.0	3		
		4 °C	164 hours	25.0	95.2	3		
				8000	109.5	3		
		–20 °C	28 days	25.0	97.1	3		
			83 days		94.5	3		
			363 days		83.9 [4)]	3		
			598 days		80.4 [4)]	3		
			28 days	8000	80.8 [4)]	3		
			83 days		108.3	3		
			363 days		94.9	3		
			598 days		91.5	3		
			28 days	50000 [3)]	78.5 [4)]	3		
			83 days		99.7	3		
			363 days		95.9	3		
			598 days		86.5	3		

2. 分析法及びバリデーション試験

Test article: ダリドレキサント

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	−80 °C	28 days	25.0	101.5	3	BA-17.029	モジュール 4.2.2.1.6
			83 days		90.5	3		
			363 days		90.1	3		
			598 days		92.4	3		
			28 days	8000	98.4	3		
			83 days		97.3	3		
			363 days		94.7	3		
			598 days		93.2	3		
			28 days	50000 [3)]	92.7	3		
			83 days		93.9	3		
			363 days		92.6	3		
			598 days		86.5	3		

1) Method V2
2) Method V3
3) Dilution QC
4) Outside acceptance criteria

EDTA = ethylenediaminetetraacetic acid; n = number; QC = quality control; RT = room temperature.

2. 分析法及びバリデーション試験

Test article: ACT-776537

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Mouse	Plasma (EDTA)	4 freeze-and-thaw cycles at –20 °C		25.0	101.9	3	BA-17.031	モジュール 4.2.2.1.3
				8000	91.9	3		
		4 freeze-and-thaw cycles at –80 °C		25.0	98.0	3		
				8000	98.1	3		
		RT	26 hours	25.0	90.5	3		
				8000	89.3	3		
		4 °C	35 hours	25.0	92.8	3		
				8000	97.1	3		
		–20 °C	375 days	25.0	85.6	3		
				8000	80.0 [2)]	3		
		–80 °C	375 days	25.0	102.8	3		
				8000	101.6	3		
Rat	Plasma (EDTA)	5 freeze-and-thaw cycles at –20 °C		25.0	95.2	3	BA-17.030	モジュール 4.2.2.1.2
				8000	91.7	3		
		5 freeze-and-thaw cycles at –80 °C		25.0	98.0	3		
				8000	100.7	3		
		RT	24 hours	25.0	97.2	3		
				8000	106.5	3		
		4 °C	41 hours	25.0	94.5	3		
				8000	102.4	3		
		–20 °C	734 days	25.0	72.4 [2)]	3		
				8000	66.7 [2)]	3		

2. 分析法及びバリデーション試験

Test article: ACT-776537

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Rat	Plasma (EDTA)	–80 °C	734 days	25.0	93.7	3	BA-17.030	モジュール 4.2.2.1.2
				8000	96.8	3		
Rabbit	Plasma (EDTA)	4 freeze-and-thaw cycles at –20 °C		25.0	96.7	3	BA-18.023	モジュール 4.2.2.1.7
				8000	87.3	3		
		4 freeze-and-thaw cycles at –80 °C		25.0	96.7	3		
				8000	91.4	3		
		RT	73 hours	25.0	104.8	3		
				8000	99.8	3		
		4 °C	89 hours	25.0	97.2	3		
				8000	98.2	3		
		–20 °C	386 days	25.0	71.5 [2)]	3		
				8000	51.5 [2)]	3		
		–80 °C	386 days	25.0	91.5	3		
				8000	94.6	3		
Dog	Plasma (EDTA)	5 freeze-and-thaw cycles at –20 °C		25.0	81.7 [2)]	3	BA-17.029	モジュール 4.2.2.1.6
				8000	89.5	3		
				50000 [1)]	88.3	3		
		6 freeze-and-thaw cycles at –20 °C		25.0	84.7 [2)]	3		
		1 freeze-and-thaw cycle at –20 °C and 28 days storage at –20 °C		25.0	87.3	3		
		5 freeze-and-thaw cycles at –80 °C		25.0	88.8	3		
				8000	89.6	3		
				50000 [1)]	95.1	3		

2. 分析法及びバリデーション試験

Test article: ACT-776537

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	RT	70 hours	25.0	93.2	3	BA-17.029	モジュール 4.2.2.1.6
				8000	94.1	3		
				50000 [1)]	97.5	3		
		4 °C	164 hours	25.0	95.2	3		
			159 hours	8000	108.2	3		
		–20 °C	28 days	25.0	87.3	3		
			83 days		84.5 [2)]	3		
			363 days		76.4 [2)]	3		
			598 days		77.7 [2)]	3		
			28 days	8000	82.9 [2)]	3		
			83 days		93.3	3		
			363 days		85.4	3		
			598 days		77.2 [2)]	3		
			28 days	50000 [1)]	89.0	3		
			83 days		91.8	3		
			363 days		80.9 [2)]	3		
			598 days		77.8 [2)]	3		
		–80 °C	28 days	25.0	91.1	3		
			83 days		88.9	3		
			363 days		96.9	3		
			598 days		80.7 [2)]	3		

2. 分析法及びバリデーション試験

Test article: ACT-776537

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	–80 °C	28 days	8000	92.1	3	BA-17.029	モジュール 4.2.2.1.6
			83 days		93.0	3		
			363 days		103.4	3		
			598 days		86.0	3		
			28 days	50000 [1)]	95.5	3		
			83 days		93.7	3		
			363 days		91.2	3		
			598 days		85.9	3		

ACT-776537 is also known as metabolite M1.
1) Dilution QC
2) Outside acceptance criteria
EDTA = ethylenediaminetetraacetic acid; n = number; QC = quality control; RT = room temperature.

2. 分析法及びバリデーション試験

Test article: ACT-776063

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Mouse	Plasma (EDTA)	4 freeze-and-thaw cycles at –20 °C		25.0	96.4	3	BA-17.031	モジュール 4.2.2.1.3
				8000	95.0	3		
		4 freeze-and-thaw cycles at –80 °C		25.0	90.3	3		
				8000	100.6	3		
		RT	26 hours	25.0	91.5	3		
				8000	101.2	3		
		4 °C	35 hours	25.0	94.8	3		
				8000	100.5	3		
		–20 °C	375 days	25.0	88.1	3		
				8000	90.4	3		
		–80 °C	375 days	25.0	93.7	3		
				8000	94.0	3		
Rat	Plasma (EDTA)	5 freeze-and-thaw cycles at –20 °C		25.0	102.3	3	BA-17.030	モジュール 4.2.2.1.2
				8000	97.2	3		
		5 freeze-and-thaw cycles at –80 °C		25.0	99.1	3		
				8000	96.3	3		
		RT	24 hours	25.0	98.7	3		
				8000	106.6	3		
		4 °C	41 hours	25.0	104.3	3		
				8000	97.0	3		
		–20°C	734 days	25.0	105.1	3		
				8000	101.9	3		

2. 分析法及びバリデーション試験

Test article: ACT-776063

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Rat	Plasma (EDTA)	–80 °C	734 days	25.0	101.2	3	BA-17.030	モジュール 4.2.2.1.2
				8000	104.3	3		
Rabbit	Plasma (EDTA)	4 freeze-and-thaw cycles at –20 °C		25.0	86.7	3	BA-18.023	モジュール 4.2.2.1.7
				8000	94.8	3		
		4 freeze-and-thaw cycles at –80 °C		25.0	91.1	3		
				8000	94.0	3		
		RT	73 hours	25.0	99.1	3		
				8000	90.5	3		
		4 °C	89 hours	25.0	106.3	3		
				8000	89.7	3		
		–20 °C	386 days	25.0	102.4	3		
				8000	94.4	3		
		–80 °C	386 days	25.0	106.1	3		
				8000	91.0	3		
Dog	Plasma (EDTA)	5 freeze-and-thaw cycles at –20 °C		25.0	85.1	3	BA-17.029	モジュール 4.2.2.1.6
				8000	94.8	3		
				50000 [1)]	86.9	3		
		5 freeze-and-thaw cycles at –80 °C		25.0	89.5	3		
				8000	96.3	3		
				50000 [1)]	95.3	3		
		RT	70 hours	25.0	91.3	3		
				8000	91.2	3		
				50000 [1)]	100.1	3		

2. 分析法及びバリデーション試験

Test article: ACT-776063

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	4 °C	164 hours	25.0	104.9	3	BA-17.029	モジュール 4.2.2.1.6
				8000	102.8	3		
		–20 °C	28 days	25.0	95.1	3		
			83 days		94.4	3		
			363 days		75.5 [2)]	3		
			598 days		72.0 [2)]	3		
			28 days	8000	91.3	3		
			83 days		100.1	3		
			363 days		97.3	3		
			598 days		85.7	3		
			28 days	50000 [1)]	89.3	3		
			83 days		96.7	3		
			363 days		93.3	3		
			598 days		89.8	3		
		–80 °C	28 days	25.0	100.0	3		
			83 days		87.7	3		
			363 days		86.1	3		
			598 days		80.0 [2)]	3		
			28 days	8000	94.3	3		
			83 days		99.7	3		
			363 days		96.6	3		
			598 days		92.2	3		

2. 分析法及びバリデーション試験

Test article: ACT-776063

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	–80 °C	28 days	50000 [1)]	93.7	3	BA-17.029	モジュール 4.2.2.1.6
			83 days		96.2	3		
			363 days		93.7	3		
			598 days		89.5	3		

ACT-776063 is also known as metabolite M3.

1) Dilution QC

2) Outside acceptance criteria

EDTA = ethylenediaminetetraacetic acid; n = number; QC = quality control; RT = room temperature.

2. 分析法及びバリデーション試験

Test article: ACT-1016-3307

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Mouse	Plasma (EDTA)	4 freeze-and-thaw cycles at –20 °C		25.0	81.7 [2)]	3	BA-17.031	モジュール 4.2.2.1.3
				8000	91.1	3		
		4 freeze-and-thaw cycles at –80 °C		25.0	83.1 [2)]	3		
				8000	95.0	3		
		RT	26 hours	25.0	91.6	3		
				8000	95.7	3		
		4 °C	35 hours	25.0	96.7	3		
				8000	100.8	3		
		–20 °C	375 days	25.0	80.9 [2)]	3		
				8000	76.4 [2)]	3		
		–80 °C	375 days	25.0	99.3	3		
				8000	100.8	3		
Rat	Plasma (EDTA)	5 freeze-and-thaw cycles at –20 °C		25.0	116.1 [3)]	3	BA-17.030	モジュール 4.2.2.1.2
				8000	91.1	3		
		5 freeze-and-thaw cycles at –80 °C		25.0	97.5	3		
				8000	91.2	3		
		RT	24 hours	25.0	108.1	3		
				8000	104.1	3		
		4 °C	41 hours	25.0	106.5	3		
				8000	97.4	3		
		–20 °C	734 days	25.0	112.0	3		
				8000	110.0	3		

2. 分析法及びバリデーション試験

Test article: ACT-1016-3307

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Rat	Plasma (EDTA)	–80 °C	734 days	25.0	109.1	3	BA-17.030	モジュール 4.2.2.1.2
				8000	107.6	3		
Rabbit	Plasma (EDTA)	4 freeze-and-thaw cycles at –20 °C		25.0	108.5	3	BA-18.023	モジュール 4.2.2.1.7
				8000	95.8	3		
		4 freeze-and-thaw cycles at –80 °C		25.0	108.5	3		
				8000	96.8	3		
		RT	73 hours	25.0	100.7	3		
				8000	94.0	3		
		4 °C	89 hours	25.0	99.2	3		
				8000	93.9	3		
		–20 °C	386 days	25.0	98.9	3		
				8000	96.0	3		
		–80 °C	386 days	25.0	101.1	3		
				8000	97.1	3		
Dog	Plasma (EDTA)	5 freeze-and-thaw cycles at –20 °C		25.0	82.1 [2)]	3	BA-17.029	モジュール 4.2.2.1.6
				8000	90.3	3		
				50000 [1)]	91.6	3		
		6 freeze-and-thaw cycles at –20 °C		25.0	92.1	3		
		5 freeze-and-thaw cycles at –80 °C		25.0	84.3 [2)]	3		
				8000	90.3	3		
				50000 [1)]	88.5	3		
		6 freeze-and-thaw cycles at –80 °C		25.0	90.8	3		

2. 分析法及びバリデーション試験

Test article: ACT-1016-3307

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	RT	70 hours	25.0	100.4	3	BA-17.029	モジュール 4.2.2.1.6
				8000	97.0	3		
				50000 [1)]	99.0	3		
		4 °C	164 hours	25.0	94.7	3		
				8000	103.5	3		
		–20 °C	28 days	25.0	97.5	3		
			83 days		94.1	3		
			363 days		87.7	3		
			598 days		82.9 [2)]	3		
			28 days	8000	91.8	3		
			83 days		101.7	3		
			363 days		98.4	3		
			598 days		93.8	3		
			28 days	50000 [1)]	93.2	3		
			83 days		96.6	3		
			363 days		95.8	3		
			598 days		88.9	3		
		–80 °C	28 days	25.0	102.3	3		
			83 days		95.7	3		
			363 days		95.1	3		
			598 days		87.3	3		

2. 分析法及びバリデーション試験

Test article: ACT-1016-3307

Species	Matrix (Anticoagulant)	Condition		Concentration Tested (ng/mL)	% Remaining	n	Report No.	Location in CTD
		Temperature	Time					
Dog	Plasma (EDTA)	–80 °C	28 days	8000	96.1	3	BA-17.029	モジュール 4.2.2.1.6
			83 days		101.6	3		
			363 days		104.5	3		
			598 days		89.5	3		
			28 days	50000 [1)]	94.1	3		
			83 days		100.1	3		
			363 days		99.1	3		
			598 days		91.5	3		

ACT-1016-3307 is also known as metabolite M10.

1) Dilution QC
2) Outside acceptance criteria
3) Results for QC dilution after three cycles out of acceptance criteria, preparation error was suspected.

EDTA = ethylenediaminetetraacetic acid; n = number; QC = quality control; RT = room temperature.

3. 薬物動態試験：吸収：単回投与

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.2.1

Report no.: B-13.116

Species (Strain):	Rat (Wistar)	Rat (Wistar)	Rat (Wistar)	Rat (Wistar)
Sex / Number of animals:	m/5	m/5	f/5	m/5
Feeding condition:	Fed	Fed	Fed	Fed
Vehicle/Formulation:	Aqueous solution in HP-β-CD 30%, adjusted to pH 4.3 - 4.6	Aqueous solution in HP-β-CD 30%, adjusted to pH 4.3 - 4.6	Aqueous solution in HP-β-CD 30%, adjusted to pH 4.3 - 4.6	Aqueous solution in HP-β-CD 30%, adjusted to pH 4.3 - 4.6
Method of administration:	i.v.	i.v.	i.v.	i.v.
Dose (mg/kg):	0.1	0.3	0.3	1.0
Sample:	Plasma	Plasma	Plasma	Plasma
Analyte:	ACT-541468	ACT-541468	ACT-541468	ACT-541468
Assay:	LC-MS/MS	LC-MS/MS	LC-MS/MS	LC-MS/MS
PK parameters:				
$AUC_{0\text{-}inf}$ (ng·h/mL)	23.2	77.5	102	226
CL (mL/min/kg)	72	65	49	74
V_{ss} (L/kg)	1.8	3.0	2.5	5.4
$t_{1/2}$ (h)	0.5	1.5	1.4	2.1

ACT-541468 = daridorexant

$AUC_{0\text{-}inf}$ = area under the plasma concentration vs time curve extrapolated to infinity; CL = systemic plasma clearance; f = female; HP-β-CD = hydroxypropyl-β-cyclodextrin; i.v. = intravenous; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; m = male; PK = pharmacokinetic(s); $t_{1/2}$ = apparent terminal half-life; V_{ss} = volume of distribution at steady state.

3. 薬物動態試験：吸収：単回投与

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.2.1

Report no.: B-13.116

Species (Strain):	Rat (Wistar)	Rat (Wistar)	Rat (Wistar)	Rat (Wistar)	Rat (Wistar)
Sex/ Number of animals:	m/5	m/5	m/5	f/5	m/5
Feeding condition:	Fed	Fed	Fed	Fed	Fed
Vehicle/Formulation:	Suspension in 0.5% MC	Suspension in 0.5% MC	Suspension in 0.5% MC	Suspension in 0.5% MC	Suspension in 0.5% MC
Method of administration:	p.o.	p.o.	p.o.	p.o.	p.o.
Dose (mg/kg):	3	10	10 [1)]	10	30
Sample:	Plasma	Plasma	Plasma	Plasma	Plasma
Analyte:	ACT-541468	ACT-541468	ACT-541468	ACT-541468	ACT-541468
Assay:	LC-MS/MS	LC-MS/MS	LC-MS/MS	LC-MS/MS	LC-MS/MS
PK parameters:					
t_{max} (h) [2)]	0.3	0.3	0.3	0.3	0.3
C_{max} (ng/mL)	61.4	387	147	606	721
$AUC_{0\text{-}inf}$ (ng·h/mL)	69.5	338	264	1030	1140
F (%)	10	15	12	30	17

ACT-541468 = daridorexant; daridorexant was administered as hydrochloride salt unless stated otherwise (doses are calculated for the free base).

1) Administered as free base.

2) Median

$AUC_{0\text{-}inf}$ = area under the plasma concentration vs time curve extrapolated to infinity; C_{max} = maximum plasma concentration; f = female; F = oral bioavailability; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; m = male; MC = methyl cellulose; PK = pharmacokinetic(s); p.o. = oral; t_{max} = time to reach maximum plasma concentration.

3. 薬物動態試験：吸収：単回投与

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.2.1

Report no.: B-13.116

Species (Strain):	Dog (Beagle)	Dog (Beagle)	Dog (Beagle)
Sex / Number of animals:	m/4	m/4	m/4
Feeding condition:	Fasted	Fasted	Fasted
Vehicle/Formulation:	Aqueous solution in HP-β-CD 30%, adjusted to pH 4.2 - 4.3	Aqueous solution in HP-β-CD 30%, adjusted to pH 4.2 - 4.3	Aqueous solution in HP-β-CD 30%, adjusted to pH 4.2 - 4.3
Method of administration:	i.v.	i.v.	i.v.
Dose (mg/kg):	0.3	1.0	3.0
Sample:	Plasma	Plasma	Plasma
Analyte:	ACT-541468	ACT-541468	ACT-541468
Assay:	LC-MS/MS	LC-MS/MS	LC-MS/MS
PK parameters:			
$AUC_{0\text{-}inf}$ (ng·h/mL)	523	1740	5050
CL (mL/min/kg)	9.6	9.6	9.9
V_{ss} (L/kg)	1.4	1.7	2.5
$t_{1/2}$ (h)	2.5	3.0	4.1

ACT-541468 = daridorexant

$AUC_{0\text{-}inf}$ = area under the plasma concentration vs time curve extrapolated to infinity; CL = systemic plasma clearance; HP-β-CD = hydroxypropyl-β-cyclodextrin; i.v. = intravenous; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; m = male; PK = pharmacokinetic(s); $t_{1/2}$ = apparent terminal half-life; V_{ss} = volume of distribution at steady state.

3. 薬物動態試験：吸収：単回投与

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.2.1

Report no.: B-13.116

Species (Strain):	Dog (Beagle)	Dog (Beagle)	Dog (Beagle)	Dog (Beagle)
Sex / Number of animals:	m/4	m/4	m/4	m/4
Feeding condition:	Fasted	Fasted	Fasted	Fed
Vehicle/Formulation:	Milled powder blend with Ac-di-sol	Milled powder blend with Ac-di-sol	Milled powder blend with Ac-di-sol	Milled powder blend with Ac-di-sol
Method of administration:	p.o.	p.o.	p.o.	p.o.
Dose (mg/kg):	1.0	3.0	10	10
Sample:	Plasma	Plasma	Plasma	Plasma
Analyte:	ACT-541468	ACT-541468	ACT-541468	ACT-541468
Assay:	LC-MS/MS	LC-MS/MS	LC-MS/MS	LC-MS/MS
PK parameters:				
t_{max} (h) [1)]	1.1	0.8	2.0	2.5
C_{max} (ng/mL)	169	486	1060	1410
$AUC_{0\text{-}inf}$ (ng·h/mL)	547	1770	7650	9710
F (%)	31 [2)]	34 [3)]	45 [3)]	58 [3)]

ACT-541468 = daridorexant
Fasted animals received 6 µg/kg pentagastrin via the intramuscular route 20 minutes before and 30 minutes after an oral dose of ACT-541468.

1) Median
2) Bioavailability calculated using data from the 1 mg/kg i.v. dose group.
3) Bioavailability calculated using data from the 3 mg/kg i.v. dose group.

Ac-di-sol = croscarmellose sodium; $AUC_{0\text{-}inf}$ = area under the plasma concentration vs time curve extrapolated to infinity; C_{max} = maximum plasma concentration; F = oral bioavailability; i.v. = intravenous; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; m = male; PK = pharmacokinetic(s); p.o. = oral; t_{max} = time to reach maximum plasma concentration.

3. 薬物動態試験：吸収：単回投与

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.2.1

Report no.: B-13.116

Species (Strain):	Dog (Beagle)	Dog (Beagle)	Dog (Beagle)	Dog (Beagle)	Dog (Beagle)	Dog (Beagle)
Sex / Number of animals:	m/4	m/4	m/4	m/4	m/4	m/4
Feeding condition:	Fasted	Fasted	Fasted	Fasted	Fasted	Fasted
Vehicle/Formulation:	Amorphous free base dispersion	Non-milled, salt, poloxamer with Ac-di-sol	Lipid-based solution	Non-milled, powder blend with Ac-di-sol	Salt suspension in 0.5% MC and 10% PEG400	Crystalline free base suspension in 0.5% MC and 10% PEG400
Method of administration:	p.o.	p.o.	p.o.	p.o.	p.o.	p.o.
Dose (mg/kg):	2.5 [1),3)]	2.5 [1),3)]	2.6 [1),3)]	10 [1)]	30 [4)]	30 [4)]
Sample:	Plasma	Plasma	Plasma	Plasma	Plasma	Plasma
Analyte:	ACT-541468	ACT-541468	ACT-541468	ACT-541468	ACT-541468	ACT-541468
Assay:	LC-MS/MS	LC-MS/MS	LC-MS/MS	LC-MS/MS	LC-MS/MS	LC-MS/MS
PK parameters:						
t_{max} (h) [2)]	2.0	1.5	0.5	3.0	0.6	1.0
C_{max} (ng/mL)	475	246	623	652	3380	3140
$AUC_{0\text{-}inf}$ (ng·h/mL)	1990	1810	1380	6940	28800	22600
F (%)	75	42	32	41	57	45

ACT-541468 = daridorexant
Fasted animals received 6 µg/kg pentagastrin via the intramuscular route 20 minutes before and 30 minutes after an oral dose of ACT-541468.

1) Capsule
2) Median
3) Total dose: 30 mg
4) Gavage

Ac-di-sol = croscarmellose sodium; $AUC_{0\text{-}inf}$ = area under the plasma concentration vs time curve extrapolated to infinity; C_{max} = maximum plasma concentration; F = oral bioavailability; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; m = male; MC = methyl cellulose; PEG = polyethylene glycol; PK = pharmacokinetic(s); p.o. = oral; t_{max} = time to reach maximum plasma concentration.

4. 薬物動態試験：吸収：反復投与

Test article: ダリドレキサント
Location in CTD: モジュール 4.2.3.2.5
Report no.: T-13.221

Species (Strain):	Rat (Wistar)
Feeding condition:	Fed
Vehicle/Formulation:	0.5% (w/w) methylcellulose suspension in purified water
Method of administration:	p.o. (gavage)
Doses (mg/kg/day):	100, 300, 1000
Duration of administration:	4 weeks
Sample:	Plasma
Analyte:	ACT-541468
Assay:	Liquid chromatography coupled to tandem mass spectrometry

Dose		C_{max} (µg/mL)		AUC_{0-24} (µg·h/mL)	
(mg/kg/day)	Sex / Number	Day 1	Week 4	Day 1	Week 4
100	m/6	2.46	2.33	10.9	12.2
	f/6	3.62	2.93	18.1	17.5
300	m/6	8.03	4.56	61.1	32.1
	f/6	12.4	7.86	90.7	54.6
1000	m/6	12.6	9.82	171	78.1
	f/4	16.9	13.2	222	128

AUC_{0-24} = area under the plasma concentration vs time curve from 0 to 24 h; C_{max} = maximum plasma concentration; f = female; m = male; p.o. = oral; w/w = weight/weight.

4. 薬物動態試験：吸収：反復投与

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.3.2.9

Report no.: T-13.220

Species (Strain):	Dog (Beagle)
Feeding condition:	Fed
Vehicle/Formulation:	0.5% (w/w) methylcellulose suspension in purified water
Method of administration	p.o.
Doses (mg/kg/day):	30, 100, 300/200 [1)]
Duration of administration:	4 weeks
Sample:	Plasma
Analyte:	ACT-541468
Assay:	Liquid chromatography coupled to tandem mass spectrometry

Dose		**C_{max} (µg/mL)**		**AUC_{0-24} (µg·h/mL)**	
(mg/kg/day)	**Sex / Number**	**Day 1**	**Week 4**	**Day 1**	**Week 4**
30	m/3	3.56	7.27	20.6	41.5
	f/3	2.93	6.11	18.9	36.7
100	m/3	8.92	22.6	95.1	184
	f/3	8.12	13.7	57.1	83.5
300/200 [1)]	m/5	11.4	25.7	170	301
	f/5	15.5	36.1	241	331

1) Dose = 300 mg/kg/day (Day 1 to Day 6), no treatment (Day 7 to Day 10), 200 mg/kg/day (Day 11 to Day 28)

AUC_{0-24} = area under the plasma concentration vs time curve from 0 to 24 h; C_{max} = maximum plasma concentration; f = female; m = male; p.o. = oral; w/w = weight/weight.

5. 薬物動態試験：分布

Test article: ダリドレキサント
Location in CTD: モジュール 4.2.2.3.3
Report no. B-16.005

Species (Strain):	Albino rat (Wistar), pigmented rat (Lister Hooded)
Sex / Number of animals:	Male/1 per time point
Feeding condition:	Fed
Vehicle/Formulation:	0.5% methylcellulose in water
Method of administration:	p.o.
Dose (mg/kg):	10
Radionuclide:	^{14}C
Specific activity:	342×10^6 - 344×10^6 Bq/mg

	Concentrations of Radioactivity (µg equivalents of ACT-541468/g of tissue) albino / pigmented			
	Sampling Time			
Tissues/Organs	**0.5 hours**	**4 hours**	**24 hours**	**3 days**
Adrenal cortex	5.96	0.808	BLQ / BLQ	BLQ / BLQ
Adrenal medulla	5.16	0.558	BLQ / BLQ	BLQ / BLQ
Aortic wall	1.70	0.413	BLQ / BLQ	BLQ / BLQ
Bile ducts	73.4	6.43	BLQ / BLQ	BLQ / BLQ
Blood	1.43	0.461	BLQ / BLQ	BLQ / BLQ
Bone Marrow	1.09	BLQ	BLQ / BLQ	BLQ / BLQ
Bone surface	0.260	BLQ	BLQ / BLQ	BLQ / BLQ
Brain	0.487	BLQ	BLQ / BLQ	BLQ / BLQ
Brown fat	3.48	0.568	BLQ / BLQ	BLQ / BLQ
Bulbo-urethral gland	2.17	0.545	BLQ / BLQ	BLQ / BLQ
Cecum mucosa	1.37	1.81	3.85 / BLQ	BLQ / BLQ
Choroid plexus	0.234	BLQ	BLQ / BLQ	BLQ / BLQ

5. 薬物動態試験：分布

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.3.3

Report no. B-16.005

	Concentrations of Radioactivity (µg equivalents of ACT-541468/g of tissue) albino / pigmented			
	Sampling Time			
Tissues/Organs	**0.5 hours**	**4 hours**	**24 hours**	**3 days**
Epididymis	0.735	0.318	BLQ / BLQ	BLQ / BLQ
Ex-orbital lachrymal gland	2.10	0.439	BLQ / BLQ	BLQ / BLQ
Harderian gland	2.23	0.537	BLQ / BLQ	BLQ / BLQ
Intra-orbital lachrymal gland	2.03	0.370	BLQ / BLQ	BLQ / BLQ
Kidney cortex	5.26	1.42	BLQ / BLQ	BLQ / BLQ
Kidney medulla	5.04	1.49	BLQ / BLQ	BLQ / BLQ
Large intestinal mucosa	0.903	BLQ	2.67 / 0.462	BLQ / 0.295
Lens	BLQ	BLQ	BLQ / ND	BLQ / BLQ
Liver	22.5	6.75	0.532 / 0.459	BLQ / BLQ
Lung	1.66	0.429	BLQ / BLQ	BLQ / BLQ
Mandibular lymph node	0.748	0.403	BLQ / BLQ	BLQ / BLQ
Meninges	0.475	BLQ	BLQ / BLQ	BLQ / BLQ
Muscle	0.975	0.300	BLQ / BLQ	BLQ / BLQ
Myocardium	3.13	0.452	BLQ / BLQ	BLQ / BLQ
Nasal mucosa	0.368	BLQ	BLQ / BLQ	BLQ / BLQ
Oesophageal wall	1.81	BLQ	BLQ / BLQ	BLQ / BLQ
Pancreas	2.73	0.499	BLQ / BLQ	BLQ / BLQ
Periodontal membrane	0.818	0.241	BLQ / BLQ	BLQ / BLQ
Pineal body	1.83	0.332	BLQ / BLQ	BLQ / BLQ
Pituitary	2.05	0.337	BLQ / BLQ	BLQ / BLQ

5. 薬物動態試験：分布

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.3.3

Report no. B-16.005

	Concentrations of Radioactivity (µg equivalents of ACT-541468/g of tissue) albino / pigmented			
	Sampling Time			
Tissues/Organs	**0.5 hours**	**4 hours**	**24 hours**	**3 days**
Preputial gland	1.76	0.619	BLQ / BLQ	BLQ / BLQ
Prostrate	1.37	0.432	BLQ / BLQ	BLQ / BLQ
Rectum mucosa	0.837	0.317	BLQ / 0.685	BLQ / BLQ
Salivary glands	1.78	0.337	BLQ / BLQ	BLQ / BLQ
Seminal vesicle	0.457	0.240	BLQ / BLQ	BLQ / BLQ
Skin (non-pigmented)	1.03	0.336	BLQ / BLQ	BLQ / BLQ
Small intestinal mucosa	6.33	0.922	0.238 / BLQ	BLQ / BLQ
Spinal cord	0.634	BLQ	BLQ / BLQ	BLQ / BLQ
Spleen	2.04	0.367	BLQ / BLQ	BLQ / BLQ
Stomach mucosa (fundus)	3.98	0.756	0.233 / BLQ	BLQ / BLQ
Stomach mucosa (non-fundic)	8.91	1.18	BLQ / BLQ	0.223 / BLQ
Testis	0.516	BLQ	BLQ / BLQ	BLQ / BLQ
Thymus	1.20	0.222	BLQ / BLQ	BLQ / BLQ
Thyroid	2.32	2.08	BLQ / BLQ	BLQ / BLQ
Tongue	2.09	0.407	BLQ / BLQ	BLQ / BLQ
Tooth pulp	1.32	0.317	BLQ / BLQ	BLQ / BLQ
Trachea	0.434	BLQ	BLQ / BLQ	BLQ / BLQ
Urinary bladder wall	2.79	0.444	BLQ / BLQ	BLQ / BLQ
Urine	10.2	19.3	BLQ / 0.460	BLQ / BLQ
Uveal tract/retina	0.256	BLQ	BLQ / BLQ	BLQ / BLQ

5. 薬物動態試験：分布

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.3.3

Report no. B-16.005

	Concentrations of Radioactivity (µg equivalents of ACT-541468/g of tissue) albino / pigmented			
	Sampling Time			
Tissues/Organs	**0.5 hours**	**4 hours**	**24 hours**	**3 days**
White fat	0.375	0.448	BLQ / BLQ	BLQ / BLQ
Upper limit of quantification	331	333	333	333
Lower limit of quantification	0.219	0.221	0.221	0.221

No 0.5 or 4 hours samples were collected for pigmented animals. All 7 days samples were BLQ for both albino and pigmented animals. Day 21 samples were collected for pigmented rats only and were all BLQ. Day 7 and 21 samples had lower limits of quantification of 0.219 and 0.221 µg equivalents of ACT-541468/g of tissue, respectively.

BLQ = below limit of quantification; ND = not determined; p.o. = oral.

6. 薬物動態試験：タンパク結合及び血球移行

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.3.1

Report no. B-13.202

Type of study: *In vitro* (^{14}C-labelled) binding to plasma proteins

Method: Equilibrium dialysis in duplicate for 6 hours at 37 °C

Species (Strain)	Concentrations Tested	% Bound
Mouse (CD-1)	0.5 - 200 µg/mL	95.8 - 97.5
Rat (Wistar)	0.5 - 200 µg/mL	95.2 - 97.2
Rabbit (New Zealand White)	0.5 - 200 µg/mL	95.6 - 99.5
Dog (Beagle)	0.5 - 200 µg/mL	94.6 - 97.6
Human	0.5 - 200 µg/mL	97.1 - 99.7
Human serum albumin	1.5, 5, 15 µg/mL	93.9 - 94.9
α1-acid glycoprotein	1.5, 5, 15 µg/mL	70.2 - 98.3

Daridorexant is highly bound to plasma proteins in all species (> 97% at 0.5 µg/mL). Rabbit and human plasma exhibited lower unbound fractions compared to mouse, rat and dog plasma. Concentration-dependent binding of plasma proteins was observed in all species with the non-linear human plasma protein binding, likely to be a result of saturable binding to α1-acid glycoprotein.

6. 薬物動態試験：タンパク結合及び血球移行

Test article: ACT-776537

Location in CTD: モジュール **4.2.2.3.2**

Report no. B-17.061

Type of study: *In vitro* binding to plasma proteins

Method: Equilibrium dialysis in triplicate for 3.5 hours at 37 °C

Species (Strain)	Concentrations Tested	% Mean Bound	Overall % Mean
Mouse (CD-1)	1.5 µg/mL	98.7	98.8
	5 µg/mL	98.7	
	15 µg/mL	98.9	
Rat (Wistar)	1.5 µg/mL	99.5	99.5
	5 µg/mL	99.6	
	15 µg/mL	99.4	
Dog (Beagle)	1.5 µg/mL	98.8	98.6
	5 µg/mL	98.6	
	15 µg/mL	98.5	
Human	0.5 µg/mL	99.9	99.8
	1.5 µg/mL	99.9	
	5 µg/mL	99.7	

ACT-776537 = M1

Plasma protein binding of ACT-776537 remained unchanged up to 5 µg/mL in human and 15 µg/mL in mouse, rat and dog.

6. 薬物動態試験：タンパク結合及び血球移行

Test article: ACT-776063
Location in CTD: モジュール 4.2.2.3.2
Report no. B-17.061

Type of study: *In vitro* binding to plasma proteins
Method: Equilibrium dialysis in triplicate for 3.5 hours at 37 °C

Species (Strain)	Concentrations Tested	% Mean Bound	Overall % Mean
Mouse (CD-1)	1.5 µg/mL	93.1	ND
	5 µg/mL	91.1	
	15 µg/mL	88.9	
Rat (Wistar)	1.5 µg/mL	93.8	ND
	5 µg/mL	91.8	
	15 µg/mL	88.0	
Dog (Beagle)	1.5 µg/mL	94.2	ND
	5 µg/mL	88.6	
	15 µg/mL	83.0	
Human	0.5 µg/mL	99.6	ND
	1.5 µg/mL	99.5	
	5 µg/mL	98.6	

ACT-776063 = M3.
Plasma protein binding of ACT-776063 was concentration-dependent in mouse, rat, dog and human. Therefore, overall mean protein binding values were not determined (ND).

6. 薬物動態試験：タンパク結合及び血球移行

Test article: ACT-1016-3307

Location in CTD: モジュール **4.2.2.3.2**

Report no. B-17.061

Type of study: *In vitro* binding to plasma proteins

Method: Equilibrium dialysis in triplicate for 3.5 hours at 37 °C

Species (Strain)	Concentrations Tested	% Mean Bound	Overall % Mean
Mouse (CD-1)	1.5 µg/mL	79.0	79.4
	5 µg/mL	78.5	
	15 µg/mL	80.6	
Rat (Wistar)	1.5 µg/mL	69.4	68.8
	5 µg/mL	68.5	
	15 µg/mL	68.6	
Dog (Beagle)	1.5 µg/mL	79.0	ND
	5 µg/mL	71.0	
	15 µg/mL	60.9	
Human	0.5 µg/mL	96.0	ND
	1.5 µg/mL	95.6	
	5 µg/mL	92.2	

ACT-1016-3307 = M10

Plasma protein binding of ACT-1016-3307 remained unchanged up to 15 µg/mL in mouse and rat. In dog and human, plasma protein binding was concentration-dependent, therefore overall mean plasma protein binding values were not determined (ND) in these species.

6. 薬物動態試験：タンパク結合及び血球移行

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.3.1

Report no. B-13.202

Type of study: *In vitro* (^{14}C-labelled) blood/plasma partitioning

Method: Incubation (0.5 - 20 µg/mL) on a blood-roller for 30 minutes at 37 °C. Blood and plasma samples analyzed by liquid scintillation counting.

Species (Strain)	Blood/Plasma Ratio
Mouse (CD-1)	1.33 - 1.67
Rat (Wistar)	2.07 - 2.32
Rabbit (New Zealand White)	0.82 - 1.15
Dog (Beagle)	0.67 - 0.96
Human	0.54 - 1.06

The blood/plasma partitioning ratio was concentration-dependent in all species.

7. 薬物動態試験：妊娠又は授乳動物における試験

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.3.4

Report no. B-20.022

Species (Strain):	Rat (Wistar)
Sex / Number of animals:	Female/5
Feeding condition:	Fed
Vehicle/Formulation:	Aqueous suspension containing 0.5% methylcellulose
Method of administration:	p.o.
Dose (mg/kg):	30

Time	Daridorexant			M1			M3			M10		
(h)	Plasma	Milk	Ratio	Plasma	Milk	Ratio	Plasma	Milk	Ratio	Plasma	Milk	Ratio
1	1450 ± 310	20800 ± 3600	14	209 ± 63	318 ± 180	1.5	947 ± 297	1590 ± 700	1.7	77.1 ± 22.8	56.9 ± 25.0	0.7
2 [1)]	921	17800	19	544	747	1.4	1210	1810	1.5	88.1	67.9	0.8
24 [1)]	BLQ	6.74 [2)]	--	94.0	14.8 [2)]	0.2	BLQ	BLQ	--	BLQ	BLQ	--

M1 = ACT-776537; M3 = ACT-776063; M10 = ACT-1016-3307. All concentration data are given in ng/mL.

1) n = 2, no SD calculated

2) n = 1

BLQ = below limit of quantification; p.o. = oral; SD = standard deviation.

8. 薬物動態試験：その他の分布試験

その他の分布試験は実施しなかった

9. 薬物動態試験：代謝：*In Vivo*

Test article: ダリドレキサント
Location in CTD: モジュール 4.2.2.4.4
Report no. B-16.062

Species (Strain):	Mouse (CD-1 and B6C3F1)
Sex / Number of animals:	Female/3
Feeding condition:	Fed
Vehicle/Formulation:	Aqueous suspension containing 0.5% methylcellulose
Method of administration:	p.o.
Dose (mg/kg):	100, administered for 1 or 14 days
Radionuclide:	^{14}C
Specific activity:	3.7×10^{6} Bq/kg

			ACT-541468 or Metabolite in Plasma (ng equivalents/mL)																	
Time		**Σ M**	**Parent**	**M 1**	**M 2**	**M 3**	**M 4**	**M 5**	**M 6**	**M 7**	**M 8**	**M 9**	**M 10**	**M 17**	**M 21**	**M 33**	**M 35**	**M 37**	**M 41**	**M 42**	**Σ All**
CD-1																				
Day 1	1 h	10724	1376	459	2092	738	492	526	554	823		744	772	296	514		1544	280	280	610	12100
	2 h	8438	662	420	3009	275	308			320		972	473		580		1643			438	9099
Day 14	1 h	10347	353		399		629				996	2194		347			4153		1630		10700
	2 h	8070	130		299		444				824	1703		325			3543		932		8200
B6C3F1																				
Day 1	1 h	13301	1597	796	2128	751	529	563	1597	375	414	2035	474		547	414	1168	870	316	323	14899
	2 h	8122	678	393	1769	404	479	304	964	277		855	356		500	178	462	373	346	462	8800
Day 14	1 h	17725	576		985		481				1819	3100		1177			7368		2796		18302
	2 h	6693	407		1093		390				465	974				373	2440		959		7100

9. 薬物動態試験：代謝：*In Vivo*

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.4

Report no. B-16.062

		ACT-541468 or Metabolite in Liver (% of administered dose)																		
Time	Σ M	Parent	M 1	M 2	M 3	M 4	M 5	M 6	M 7	M 8	M 9	M 7/9	M 10	M 11	M 17	M 20	M 21	M 31	M 35	M 37	Σ All
CD-1																					
Day 1	9.07	0.63	0.09	1.12	0.42	0.69	0.19	0.30	1.11	0.23	2.18		0.54		0.36		0.95		0.72	0.29	9.71
Day 14	7.00	0.29		0.20	0.09	0.99	0.06	0.11	0.79		2.80		0.45		0.22	0.23	0.28		0.59	0.20	7.30
B6C3F1																					
Day 1	3.91	0.49		0.23	0.17	1.19	0.13					1.12	0.15	0.37	0.20		0.15			0.19	4.40
Day 14	7.83	0.47		0.32		3.13		0.09		0.08		2.15	0.45	0.28	0.19		0.14	0.24	0.44	0.32	8.30

		ACT-541468 or Metabolite in Gall Bladder (% of administered dose)												
Time	Σ M	Parent	M2	M4	M6	M8	M9	M17	M20	M33	M35	M38	M41	Σ All
CD-1														
Day 1	15.3	0.0	0.38	0.22	1.13	0.91	8.71	0.71	0.69	0.23	0.94	0.69	0.69	15.3
Day 14	1.70	0.0			0.06	0.09	1.03	0.07	0.15		0.12		0.18	1.70
B6C3F1														
Day 1	5.62	0.0			0.79	0.28	3.24	0.22	0.52		0.12	0.24	0.20	5.62
Day 14	5.20	0.0			0.24	0.40	2.86	0.33	0.73	0.07	0.07	0.07	0.44	5.20

Liver samples from 1 to 8 hours were pooled in CD-1 and B6C3F1 mice. Gall bladder samples from 1 to 4 hours and from 1 to 8 hours were pooled in CD-1 and B6C3F1 mice, respectively. Empty cells indicate levels below the limit of quantification, which was less than 0.01% of the dose.

M = metabolite; p.o. = oral.

9. 薬物動態試験：代謝：*In Vivo*

Test article: ダリドレキサント
Location in CTD: モジュール 4.2.2.4.3
Report no. B-14.054

Species (Strain):	Rat (Wistar)	
Sex / Number of animals:	Male/2	
Feeding condition:	Fed	
Vehicle/Formulation:	Aqueous solution in 30% hydroxypropyl- β-cyclodextrin containing 5% (v/v) DMSO	Aqueous suspension containing 0.5% methylcellulose
Method of administration:	i.v.	p.o.
Dose (mg/kg):	1	10
Radionuclide:	^{14}C	
Specific activity:	3.7×10^{6} Bq/kg	

ACT-541468 or Metabolite in Plasma (ng equivalents/mL), Urine, Bile and Feces (% of administered dose)

Sample (Time: h)	**Σ M**	**Parent**	**M 1**	**M 2**	**M 3**	**M 4**	**M 5**	**M 6**	**M 7**	**M 8**	**M 9**	**M 10**	**M 11**	**M 12**	**M 13**	**M 14**	**M 17**	**M 18**	**M 20**	**M 21**	**M 22**	**M 23**	**M 24**	**M 28**
i.v.																								
Plasma (0.5)	135	80	12	40	18	3.4	9.0	4.1				6.0	10		5.5	4.5	4.8					5.4	13	
Plasma (2)	105	16	11	31	3.5		6.6	3.1				2.2			15	2.0	1.9						3.1	
Plasma (6)	36	2.6	13	12												11								
Urine (0 - 8)	1.0	0.1					0.3		0.1			0.3								0.1		0.2		
Bile (0 - 24)	87			4.4	2.2	2.1		27		3.0	19	2.7	5.4		3.1	2.1	2.4	3.2	5.0	3.6	1.8			0.2
Feces (0 - 48)	1.0			0.2				0.1	0.3			0.2												0.1

9. 薬物動態試験：代謝：*In Vivo*

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.3

Report no. B-14.054

		ACT-541468 or Metabolite in Plasma (ng equivalents/mL), Urine, Bile and Feces (% of administered dose)																				
Sample (Time: h)	**Σ M**	**Parent**	**M 1**	**M 2**	**M 3**	**M 4**	**M 5**	**M 6**	**M 7**	**M 8**	**M 9**	**M 10**	**M 11**	**M 12**	**M 13**	**M 14**	**M 17**	**M 18**	**M 20**	**M 21**	**M 22**	**M 23**	**M 24**	**M 28**
p.o.																								
Plasma (0.5)	1204	298	83	207	249	53	145	24				95	51		98	67	24					44	69	
Plasma (2)	951	146	84	222	152	11	99	40	19			71	33		84	51	25						63	
Plasma (6)	411	101	95	88	90	22	20	38					16			81								
Urine (0 - 8)	0.8	0.1					0.2		0.1			0.2					-			0.1	-	0.2		
Bile (0 - 24)	80			7.5	0.5	2.2		17	1.4	1.8	12	1.3	6.2	5.9	5.3	4.1	5.5	1.3	4.2	3.7				
Feces (0 - 48)	10	0.6		1.7		1.8		0.3	2.5			0.6	0.6	0.6	1.5			0.2			0.3			

Empty cells indicate absence of the metabolite.

- = trace level, accounting for less than 0.1% of dose; DMSO = dimethyl sulfoxide; i.v. = intravenous; M = metabolite; p.o. = oral.

10. 薬物動態試験：代謝：*In Vitro*

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.2

Report no. B-14.019

Type of study: Identification of ^{14}C-labelled metabolites produced *in vitro*

Test system: Mouse, rat (male and female), rabbit, dog, monkey and human liver microsomes and hepatocytes

Concentration: 10 µmol/L

Method: HPLC with on-line radio-detection

Species		**Incubation**	**% of Total Chromatogram Radioactivity**											
(Strain)	**Sample**	**Time**	**Parent**	**M1**	**M2**	**M3**	**M4**	**M5**	**M6**	**M7**	**M8**	**M9**	**M10**	**Σ M**
Mouse	Liver microsomes	5 min	55			10	21	13						45
(CD-1)	Hepatocytes	1 h	30				22			3.7	5.5	36		70
Male Rat	Liver microsomes	10 min	68			13	9.7	5.9						29
(Wistar)		30 min	52			17	13	5.6						36
	Hepatocytes	1 h	63	1.4		12	3.0	4.5	4.7			4.3		30
		4 h	7.1			8.4		5.6	29		1.9	23	3.2	71
Female Rat	Liver microsomes	10 min	82			5.0	7.0							12
(Wistar)		30 min	67			6.7	12	2.2						21
	Hepatocytes	1 h	76			9.2	2.2	4.8	3.1			2.6		22
		4 h	13			9.8		4.6	28					42
Rabbit	Liver microsomes	10 min	67			6.6	9.6	15						31
(New Zealand White)	Hepatocytes	1 h	79			4.3	5.6	2.0				8.6		21

10. 薬物動態試験：代謝：*In Vitro*

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.2

Report no. B-14.019

Species (Strain)	Sample	Incubation Time	% of Total Chromatogram Radioactivity											
			Parent	M1	M2	M3	M4	M5	M6	M7	M8	M9	M10	Σ M
Dog (Beagle)	Liver microsomes	10 min	63			12	11	15						37
	Hepatocytes	1 h	66	1.4		11	5.4	14		1.0		0.9	0.7	34
		4 h	23	3.8	4.1	17		28				21	3.4	77
Monkey (Cynomolgus)	Liver microsomes	10 min	55			11	11	23						45
	Hepatocytes	1 h	74			5.5	7.6	10				2.8		26
		4 h	69			4.7	9.1	8.4	2.1			6.7		31
Human	Liver microsomes	10 min	81			7.0	4.3	7.6						19
	Hepatocytes, Batch 1	1 h	63			9.9	3.9	11	3.0			7.7	1.6	37
	Hepatocytes, Batch 2	4 h	72			12	5.8					10		28
	Hepatocytes, Batch 3	1 h	40	4.4	6.9	10	6.7	5.4		3.8	7.2	8.8	6.2	60

Empty cells indicate absence of the metabolite.

HPLC = high-performance liquid chromatography; M = metabolite.

10. 薬物動態試験：代謝：*In Vitro*

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.4

Report no. B-16.062

Type of study: Identification of ^{14}C-labelled metabolites produced *in vitro*

Test system: CD-1 and B6C3F1 mouse liver microsomes and hepatocytes

Concentration: 10 µmol/L

Method: HPLC with on-line radio-detection

Species (Strain)	Sample	Incubation Time	% of Total Chromatogram Radioactivity												
			Parent	M3	M4	M5	M7	M8	M9	M10	M12	M17	M20	M38	Σ M
Mouse (CD-1)	Liver microsomes	5 min	51	9.4	20	12	2.9			2.0	2.5				49
	Hepatocytes	60 min	28	2.6	22		3.6	5.3	35	3.1					72
Mouse (B6C3F1)	Liver microsomes	5 min	45	8.0	30	9.8	2.6			1.5	3.1				55
	Hepatocytes	60 min	13	5.5	7.4		4.0	6.6	50	5.0		3.6	1.6	2.9	87

Empty cells indicate absence of the metabolite.

HPLC = high-performance liquid chromatography; M = metabolite.

10. 薬物動態試験：代謝：*In Vitro*

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.5

Report no. B-15.073

Type of study: Identification of cytochrome P450 isomers responsible for the metabolism of daridorexant *in vitro*

Test system: Incubations with recombinant human cytochrome P450

Concentration: 10 µmol/L

Method: LC-MS/MS coupled with ^{14}C-radio-detection

	% of Total Chromatogram Radioactivity								
Cytochrome P450	**M1**	**M3**	**M4**	**M5**	**M7**	**M10**	**M12**	**M13**	**Daridorexant**
CYP1A2			4.6						95
CYP2B6									100
CYP2C8			23						77
CYP2C9			18						82
CYP2C19		3.3	44		8.2		26	1.5	14
CYP2D6			59				9.6		32
CYP3A4	5.6	18	2.5	36		4.9	9.4	5.8	18

Empty cells indicate absence of the metabolite.

CYP = cytochrome P450 (isomer); LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; M = metabolite.

10. 薬物動態試験：代謝：*In Vitro*

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.5

Report no. B-15.073

Type of study: Identification of cytochrome P450 isomers responsible for the metabolism of daridorexant *in vitro*

Test system: Incubations with human liver microsomes in the presence and absence of chemical cytochrome P450 inhibitors

Concentration: 10 µmol/L

Method: LC-MS/MS coupled with ^{14}C-radio-detection

Cytochrome P450	Inhibitor	Inhibitor Concentration (µmol/L)	% of Total Chromatogram Radioactivity			
			M3	M4	M5	Daridorexant
CYP1A2	Furafylline	20	5.5	5.4	8.8	80
CYP2B6	Ticlopidine	30	3.2	4.2	7.5	85
CYP2C8	Montelukast	3	4.4	4.6	11	80
CYP2C9	Sulfaphenazole	3	2.8	5.1	13	79
CYP2C19	*S*-(+)-*N*-3-benzylnirvanol	5	4.1	4.3	11	81
CYP2D6	Quinidine	1	5.6	4.9	13	77
CYP3A4	Ketoconazole	1				100
	Azamulin	5				100

Empty cells indicate absence of the metabolite.

CYP = cytochrome P450 (isomer); LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; M = metabolite.

10. 薬物動態試験：代謝：*In Vitro*

Test article: ACT-776537, ACT-776353, ACT-776063, ACT-774170, ACT-1006-2069

Location in CTD: モジュール **4.2.2.4.5**

Report no. B-15.073

Type of study: Identification of metabolites observed in incubations of chemical references of metabolites *in vitro*

Test system: Incubations with human liver microsomes or recombinant human cytochrome P450

Concentration: 10 µmol/L

Method: LC-MS/MS

	Incubation System		
Reference (Metabolite Code)	**HLM**	**CYP2C19**	**CYP3A4**
ACT-776537 (M1)	M2	M2, M13, M24	M2, [m/z 467] [1)]
ACT-776353 (M2)			
ACT-776063 (M3)	M1	M12	M1, M12
ACT-774170 (M4)	M12	M7, M12	M12
ACT-1006-2069 (M5)	M7, M10, [m/z 481] [1)]	M7, M10	M7, M10, [m/z 481] [1)]

Empty cells indicate absence of a metabolite. Molecular mass given as [M+H+].

1) Additional metabolites of unknown nature.

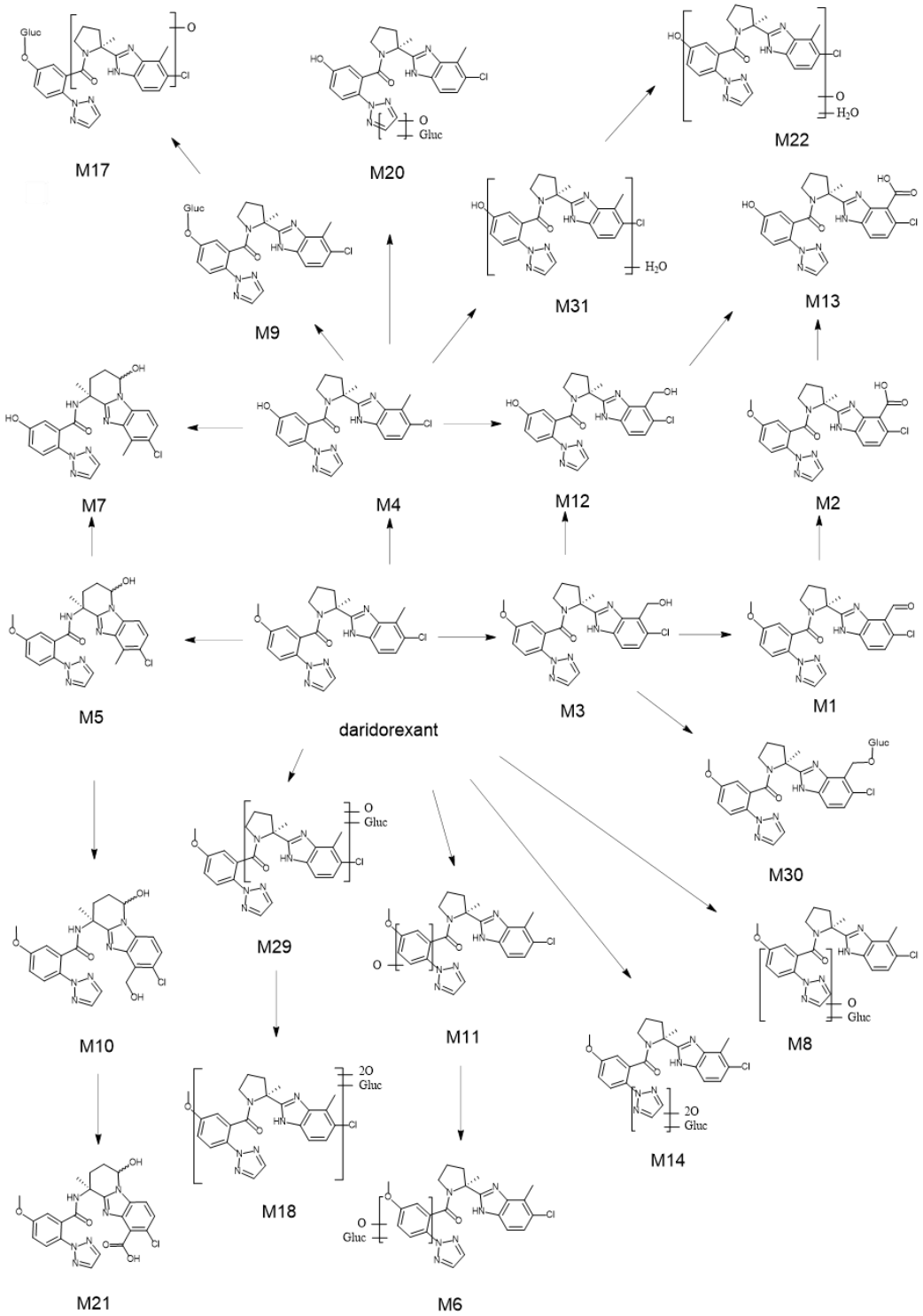
CYP = cytochrome P450 (isomer); HLM = human liver microsomes; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; M = metabolite.

11. 薬物動態試験：推定代謝経路

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.1

Report no. B-15.042



12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ダリドレキサント, ACT-776537, ACT-776063, ACT-1016-3307
Location in CTD: モジュール 4.2.2.6.4, モジュール 4.2.2.6.5
Report no. B-14.064, B-17.063

Type of study: Human PXR activation *in vitro*
Test system: CV-1 cells
Method: Transcriptional activation assay, measurement of luciferase activity

Concentration (µmol/L)	Fold induction (mean ± SD)						
	Daridorexant	Rifampicin [1)]	ACT-776537	ACT-776063	ACT-1016-3307	Rifampicin [1)]	
						Experiment 1	Experiment 2
0.03	1.04 ± 0.07	0.99 ± 0.05	1.17 ± 0.14	1.01 ± 0.11	0.97 ± 0.15	0.96 ± 0.14	1.15 ± 0.16
0.09	1.13 ± 0.11	-	-	-	-	-	-
0.1	-	1.35 ± 0.06	1.26 ± 0.15	1.03 ± 0.14	0.87 ± 0.05	1.25 ± 0.06	1.60 ± 0.11
0.28	1.22 ± 0.17	-	-	-	-	-	-
0.3	-	1.80 ± 0.13	2.09 ± 0.18	1.30 ± 0.23	0.87 ± 0.20	2.16 ± 0.46	2.82 ± 0.06
0.93	2.12 ± 0.25	-	-	-	-	-	-
1	-	3.83 ± 0.51	3.99 ± 0.33	2.15 ± 0.25	0.96 ± 0.20	4.00 ± 0.42	4.82 ± 0.14
2.78	3.61 ± 0.78	-	-	-	-	-	-
3	-	5.85 ± 0.51	5.16 ± 0.18	3.99 ± 0.09	1.12 ± 0.08	5.84 ± 0.78	6.50 ± 0.17
9.25	5.98 ± 0.04	-	-	-	-	-	-
10	-	6.78 ± 0.43	T	5.56 ± 0.19	1.21 ± 0.16	6.60 ± 0.94	6.99 ± 0.11
27.8	5.68 ± 0.91	-	-	-	-	-	-
30	-	6.84 ± 0.29	T	5.52 ± 0.07	2.35 ± 0.05	6.21 ± 1.22	6.30 ± 0.08
EC_{50} (µmol/L)	3.0	1.2	ND	2.4	ND	0.8	0.6
E_{max} (fold vs. control)	6.6	7.3	ND	6.2	ND	6.8	7.0

ACT-776537 = M1; ACT-776063 = M3; ACT-1016-3307 = M10

1) Positive control

- = no data; CV-1 = green monkey kidney cells; EC_{50} = concentration that gives half-maximal response; E_{max} = maximal effect; ND = not determined; PXR = pregnane X receptor; SD = standard deviation; T = cytotoxic.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.6.1

Report no. B-14.018

Type of study: Competitive cytochrome P450 inhibition *in vitro*

Test system: Human liver microsomes and recombinant CYP2C19

Method: Incubation of test substances with human liver microsomes or recombinant cytochrome P450 with daridorexant concentrations up to 100 µmol/L for up to 40 min at 37 °C. Analysis of samples by LC-MS/MS.

Cytochrome P450	Reaction	IC_{50} (µmol/L)	K_i (µmol/L)
CYP1A2	Phenacetin *O*-deethylation	84	ND
CYP2A6	Coumarin 7-hydroxylation	> 100	ND
CYP2B6	Bupropion hydroxylation	67	ND
CYP2C8	Paclitaxel 6α-hydroxylation	18	ND
CYP2C9	Diclofenac 4'-hydroxylation	19	ND
CYP2C19	(*S*)-mephenytoin 4'-hydoxylation	8.2	ND
CYP2D6	Dextromethorphan *N*-demethylation	46	ND
CYP3A4	Midazolam 1'-hydroxylation	15	4.8
CYP3A4	Testosterone 6β-hydroxylation	7.3	4.6

CYP = cytochrome P450 (isomer); IC_{50} = concentration that causes 50% inhibition; K_i = inhibition constant; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; ND = not determined.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.6.1, モジュール 4.2.2.6.2, モジュール 4.2.2.6.3

Report no. B-14.018, B-17.055, B-20.034

Type of study: Time-dependent cytochrome P450 inhibition *in vitro*

Test system: Human liver microsomes and recombinant CYP2C19

Method: Analysis of samples by LC-MS/MS

Cytochrome P450	IC_{50} without Pre-incubation (µmol/L)	IC_{50} with Pre-incubation (µmol/L)	IC_{50} Shift
CYP1A2	> 100	> 100	ND
CYP2B6	> 100	> 100	ND
CYP2C8	32	41	0.8
CYP2C9	56	64	0.9
CYP2C19	33	21	1.6
CYP2D6	72	77	0.9
CYP3A4	22	6.4	3.4
CYP3A4	K_I = 44 µmol/L	k_{inact} = 0.036 min^{-1}	k_{inact}/K_I = 0.8 L/mmol/min

CYP = cytochrome P450 (isomer); IC_{50} = concentration that causes 50% inhibition; K_I = inactivation constant; k_{inact} = rate constant of enzyme inactivation; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ACT-776537, ACT-776063, ACT-1016-3307
Location in CTD: モジュール 4.2.2.6.2, モジュール 4.2.2.6.3
Report no. B-17.055, B-20.034

Type of study: Time-dependent cytochrome P450 inhibition *in vitro*
Test system: Human liver microsomes and recombinant CYP2C19
Method: Analysis of samples by LC-MS/MS

Cytochrome P450	Compound	IC_{50} without Pre-incubation (µmol/L)	IC_{50} with Pre-incubation (µmol/L)	IC_{50} Shift
CYP1A2	ACT-776537	> 100	> 100	ND
CYP2B6	ACT-776537	> 100	> 100	ND
CYP2C8	ACT-776537	42	43	1.0
CYP2C9	ACT-776537	69	89	0.8
CYP2C19	ACT-776537	48	26	1.8
CYP2D6	ACT-776537	99	> 100	< 1
CYP3A4	ACT-776537	95	21	4.5
CYP3A4	ACT-776537	K_I = 333 µmol/L	k_{inact} = 0.071 min^{-1}	k_{inact}/K_I = 0.2 L/mmol/min
CYP1A2	ACT-776063	> 100	> 100	ND
CYP2B6	ACT-776063	> 100	> 100	ND
CYP2C8	ACT-776063	> 100	80	> 1.3
CYP2C9	ACT-776063	> 100	> 100	ND
CYP2C19	ACT-776063	> 100	> 100	ND
CYP2D6	ACT-776063	> 100	> 100	ND
CYP3A4	ACT-776063	> 100	76	> 1.3

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ACT-776537, ACT-776063, ACT-1016-3307

Location in CTD: モジュール 4.2.2.6.2, モジュール 4.2.2.6.3

Report no. B-17.055, B-20.034

Cytochrome P450	Compound	IC_{50} without Pre-incubation (µmol/L)	IC_{50} with Pre-incubation (µmol/L)	IC_{50} Shift
CYP1A2	ACT-1016-3307	> 100	> 100	ND
CYP2B6	ACT-1016-3307	> 100	> 100	ND
CYP2C8	ACT-1016-3307	> 100	> 100	ND
CYP2C9	ACT-1016-3307	> 100	> 100	ND
CYP2C19	ACT-1016-3307	> 100	> 100	ND
CYP2D6	ACT-1016-3307	> 100	> 100	ND
CYP3A4	ACT-1016-3307	> 100	> 100	ND

ACT-776537 = M1; ACT-776063 = M3; ACT-1016-3307 = M10.

CYP = cytochrome P450 (isomer); IC_{50} = concentration that causes 50% inhibition; K_I = inactivation constant; k_{inact} = rate constant of enzyme inactivation; LC-MS/MS = liquid chromatography coupled to tandem mass spectrometry; ND = not determined.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.6.4

Report no. B-14.064

Type of study: Cytochrome P450 induction *in vitro*

Test system: Human hepatocytes

Method: Real-time PCR

	CYP1A2 mRNA Fold Induction (mean ± SD)					
	Daridorexant			Omeprazole [1)]		
Concentration (µmol/L)	Batch: YOW	Batch: NON	Batch: EJW	Batch: YOW	Batch: NON	Batch: EJW
0.3	0.78 ± 0.07	0.76 ± 0.09	0.72 ± 0.02	-	-	-
1	1.00 ± 0.09	0.79 ± 0.04	0.84 ± 0.01	3.52 ± 0.71	2.23 ± 0.25	1.51 ± 0.26
3	1.15 ± 0.24	0.98 ± 0.24	0.94 ± 0.15	-	-	-
10	1.31 ± 0.10	1.66 ± 0.40	1.07 ± 0.17	20.4 ± 3.86	14.6 ± 0.89	9.06 ± 0.25
25	-	-	-	27.5 ± 4.98	24.6 ± 4.75	18.3 ± 4.66
30	3.24 ± 0.21	1.98 ± 0.09	2.18 ± 0.25	-	-	-
50	-	-	-	42.4 ± 6.80	21.7 ± 5.38	26.8 ± 6.98
100	-	-	-	44.0 ± 8.17	27.7 ± 2.11	49.3 ± 4.01

1) Positive control

- = no data; CYP = cytochrome P450 (isomer); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ダリドレキサント
Location in CTD: モジュール 4.2.2.6.4
Report no. B-14.064

Type of study: Cytochrome P450 induction *in vitro*
Test system: Human hepatocytes
Method: Real-time PCR

	CYP2B6 mRNA Fold Induction (mean ± SD)							
	Daridorexant				Phenobarbital [1)]			
Concentration (µmol/L)	Batch: YOW	Batch: NON	Batch: HEP220786	Batch: EJW	Batch: YOW	Batch: NON	Batch: HEP220786	Batch: EJW
0.3	0.90 ± 0.10	0.72 ± 0.03	1.17 ± 0.11	0.74 ± 0.07	-	-	-	-
1	1.38 ± 0.10	1.15 ± 0.08	2.26 ± 0.09	1.08 ± 0.08	-	-	-	-
3	2.41 ± 0.25	1.75 ± 0.50	3.99 ± 0.99	1.38 ± 0.29	-	-	-	-
10	2.45 ± 0.39	3.45 ± 0.60	9.30 ± 1.94	1.57 ± 0.27	-	-	-	-
30	2.60 ± 0.16	1.57 ± 0.07	13.1 ± 1.02	0.81 ± 0.08	-	-	-	-
100	-	-	-	-	1.13 ± 0.14	1.73 ± 0.25	2.75 ± 0.12	1.29 ± 0.08
300	-	-	-	-	2.96 ± 0.20	3.09 ± 0.96	6.30 ± 1.21	3.09 ± 0.80
500	-	-	-	-	5.69 ± 0.81	5.68 ± 2.09	7.60 ± 0.85	2.57 ± 0.45
1000	-	-	-	-	10.3 ± 0.67	7.25 ± 0.98	17.2 ± 3.00	3.63 ± 0.54
3000	-	-	-	-	8.88 ± 1.17	7.73 ± 1.01	24.0 ± 1.20	5.22 ± 1.17

1) Positive control
- = no data; CYP = cytochrome P450 (isomer); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ダリドレキサント
Location in CTD: モジュール 4.2.2.6.4
Report no. B-14.064

Type of study: Cytochrome P450 induction *in vitro*
Test system: Human hepatocytes
Method: Real-time PCR

	CYP2C9 mRNA Fold Induction (mean ± SD)					
	Daridorexant			Rifampicin [1)]		
Concentration (µmol/L)	Batch: NON	Batch: HEP220786	Batch: EJW	Batch: NON	Batch: HEP220786	Batch: EJW
0.1	-	-	-	1.29 ± 0.20	1.47 ± 0.23	0.75 ± 0.04
0.3	0.67 ± 0.09	1.13 ± 0.16	0.61 ± 0.01	2.48 ± 0.06	1.43 ± 0.08	0.98 ± 0.20
1	0.94 ± 0.09	1.25 ± 0.19	0.79 ± 0.04	3.10 ± 0.74	1.98 ± 0.33	1.35 ± 0.14
3	1.37 ± 0.36	1.64 ± 0.11	1.03 ± 0.13	-	-	-
10	2.17 ± 0.30	1.85 ± 0.10	1.23 ± 0.08	4.28 ± 0.57	1.93 ± 0.05	2.42 ± 0.34
20	-	-	-	4.75 ± 0.58	2.23 ± 0.36	2.03 ± 0.20
30	1.08 ± 0.14	2.27 ± 0.15	0.87 ± 0.04	-	-	-
50	-	-	-	5.26 ± 0.92	2.13 ± 0.06	2.14 ± 0.22
EC_{50} (µmol/L)	-	1.2	-	0.8	0.3	1.6
E_{max} (fold vs. control)	-	2.3	-	4.9	2.1	2.3

1) Positive control

- = no data; CYP = cytochrome P450 (isomer); EC_{50} = concentration that gives half-maximal response; E_{max} = maximal effect; mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ダリドレキサント
Location in CTD: モジュール 4.2.2.6.4
Report no. B-14.064

Type of study: Cytochrome P450 induction *in vitro*
Test system: Human hepatocytes
Method: Real-time PCR

	CYP3A4 mRNA Fold Induction (mean ± SD)					
	Daridorexant			Rifampicin [1)]		
Concentration (µmol/L)	Batch: NON	Batch: HEP220786	Batch: EJW	Batch: NON	Batch: HEP220786	Batch: EJW
0.1	-	-	-	3.75 ± 0.92	6.94 ± 1.83	2.43 ± 0.20
0.3	0.94 ± 0.04	1.38 ± 0.09	1.15 ± 0.38	9.98 ± 0.86	13.0 ± 0.51	6.45 ± 1.16
1	1.75 ± 0.15	2.88 ± 0.39	2.49 ± 0.36	14.7 ± 3.60	33.4 ± 12.4	13.3 ± 1.36
3	4.93 ± 1.13	8.91 ± 1.95	6.56 ± 0.58	-	-	-
10	11.9 ± 2.20	23.4 ± 0.82	10.1 ± 0.84	26.5 ± 1.91	40.7 ± 7.05	31.7 ± 3.45
20	-	-	-	29.4 ± 3.28	45.6 ± 4.59	32.1 ± 0.95
30	5.07 ± 0.81	33.1 ± 0.46	3.69 ± 0.44	-	-	-
50	-	-	-	29.9 ± 6.33	49.9 ± 2.35	27.3 ± 5.55
EC_{50} (µmol/L)	-	2.4	-	1.0	0.6	1.3
E_{max} (fold vs. control)	-	37.9	-	30.1	47.4	32.3

1) Positive control

- = no data; CYP = cytochrome P450 (isomer); EC_{50} = concentration that gives half-maximal response; E_{max} = maximal effect; mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ACT-776537

Location in CTD: モジュール 4.2.2.6.5

Report no. B-17.063

Type of study: Cytochrome P450 induction *in vitro*

Test system: Human hepatocytes

Method: Real-time PCR

	CYP1A2 mRNA Fold Induction (mean ± SD)					
	ACT-776537			Omeprazole [1)]		
Concentration (µmol/L)	Batch: YOW	Batch: YYX	Batch: AKB	Batch: YOW	Batch: YYX	Batch: AKB
0.3	0.89 ± 0.08	0.91 ± 0.11	1.54 ± 0.17	-	-	-
1	0.95 ± 0.04	0.85 ± 0.11	1.78 ± 0.36	1.92 ± 0.26	2.01 ± 0.18	3.13 ± 0.40
3	1.08 ± 0.15	1.1 ± 0.1	1.50 ± 0.16	-	-	-
10	1.10 ± 0.17	0.8 ± 0.2	1.93 ± 0.48	12.9 ± 2.33	9.62 ± 0.80	16.4 ± 3.99
25	-	-	-	23.4 ± 3.77	15.4 ± 2.32	31.3 ± 4.34
50	-	-	-	25.5 ± 2.25	18.1 ± 3.60	38.2 ± 6.87
100	-	-	-	28.8 ± 6.83	33.7 ± 4.46	43.4 ± 0.67

ACT-776537 = M1

1) Positive control

- = no data; CYP = cytochrome P450 (isomer); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ACT-776537
Location in CTD: モジュール 4.2.2.6.5
Report no. B-17.063

Type of study: Cytochrome P450 induction *in vitro*
Test system: Human hepatocytes
Method: Real-time PCR

	CYP2B6 mRNA Fold Induction (mean ± SD)					
	ACT-776537			Phenobarbital [1)]		
Concentration (µmol/L)	Batch: YOW	Batch: YYX	Batch: AKB	Batch: YOW	Batch: YYX	Batch: AKB
0.3	0.79 ± 0.22	0.88 ± 0.12	1.47 ± 0.25	-	-	-
1	1.33 ± 0.14	0.74 ± 0.15	1.98 ± 0.17	-	-	-
3	1.99 ± 0.70	1.46 ± 0.35	3.04 ± 0.11	-	-	-
10	1.76 ± 0.68	2.22 ± 0.49	5.57 ± 1.58	-	-	-
100	-	-	-	0.90 ± 0.17	0.93 ± 0.17	2.13 ± 0.22
300	-	-	-	2.28 ± 0.12	2.93 ± 0.05	3.95 ± 0.91
500	-	-	-	2.33 ± 0.60	3.43 ± 1.07	9.01 ± 2.40
1000	-	-	-	4.87 ± 0.46	10.0 ± 1.95	14.5 ± 2.30
3000	-	-	-	7.33 ± 0.21	18.0 ± 6.21	23.5 ± 5.77

ACT-776537 = M1
1) Positive control
- = no data; CYP = cytochrome P450 (isomer); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ACT-776063
Location in CTD: モジュール 4.2.2.6.5
Report no. B-17.063

Type of study: Cytochrome P450 induction *in vitro*
Test system: Human hepatocytes
Method: Real-time PCR

	CYP1A2 mRNA Fold Induction (mean ± SD)					
	ACT-776063			Omeprazole [1)]		
Concentration (µmol/L)	Batch: YOW	Batch: YYX	Batch: AKB	Batch: YOW	Batch: YYX	Batch: AKB
0.3	0.74 ± 0.10	0.69 ± 0.10	1.84 ± 0.16	-	-	-
1	0.72 ± 0.04	0.61 ± 0.12	1.81 ± 0.38	1.92 ± 0.26	2.01 ± 0.18	3.13 ± 0.40
3	0.70 ± 0.12	0.87 ± 0.14	2.03 ± 0.17	-	-	-
10	0.83 ± 0.20	1.30 ± 0.22	3.22 ± 0.96	12.9 ± 2.33	9.62 ± 0.80	16.4 ± 3.99
25	-	-	-	23.4 ± 3.77	15.4 ± 2.32	31.3 ± 4.34
50	-	-	-	25.5 ± 2.25	18.1 ± 3.60	38.2 ± 6.87
100	-	-	-	28.8 ± 6.83	33.7 ± 4.46	43.4 ± 0.67

ACT-776063 = M3
1) Positive control
- = no data; CYP = cytochrome P450 (isomer); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ACT-776063
Location in CTD: モジュール 4.2.2.6.5
Report no. B-17.063

Type of study: Cytochrome P450 induction *in vitro*
Test system: Human hepatocytes
Method: Real-time PCR

	CYP2B6 mRNA Fold Induction (mean ± SD)					
	ACT-776063			Phenobarbital [1)]		
Concentration (µmol/L)	Batch: YOW	Batch: YYX	Batch: AKB	Batch: YOW	Batch: YYX	Batch: AKB
0.3	0.87 ± 0.10	0.42 ± 0.18	1.68 ± 0.29	-	-	-
1	0.74 ± 0.12	0.61 ± 0.13	1.65 ± 0.31	-	-	-
3	1.08 ± 0.03	0.89 ± 0.27	2.84 ± 0.19	-	-	-
10	1.93 ± 0.43	3.38 ± 0.82	8.92 ± 1.52	-	-	-
100	-	-	-	0.90 ± 0.17	0.93 ± 0.17	2.13 ± 0.22
300	-	-	-	2.28 ± 0.12	2.93 ± 0.05	3.95 ± 0.91
500	-	-	-	2.33 ± 0.60	3.43 ± 1.07	9.01 ± 2.40
1000	-	-	-	4.87 ± 0.46	10.0 ± 1.95	14.5 ± 2.30
3000	-	-	-	7.33 ± 0.21	18.0 ± 6.21	23.5 ± 5.77

ACT-776063 = M3
1) Positive control
- = no data; CYP = cytochrome P450 (isomer); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ACT-1016-3307

Location in CTD: モジュール 4.2.2.6.5

Report no. B-17.063

Type of study: Cytochrome P450 induction *in vitro*

Test system: Human hepatocytes

Method: Real-time PCR

	CYP1A2 mRNA Fold Induction (mean ± SD)					
	ACT-1016-3307			Omeprazole [1)]		
Concentration (µmol/L)	Batch: YOW	Batch: YYX	Batch: AKB	Batch: YOW	Batch: YYX	Batch: AKB
0.3	0.55 ± 0.05	0.65 ± 0.24	2.41 ± 0.84	-	-	-
1	0.52 ± 0.11	0.62 ± 0.10	2.16 ± 0.46	1.92 ± 0.26	2.01 ± 0.18	3.13 ± 0.40
3	0.61 ± 0.20	0.76 ± 0.09	2.06 ± 0.32	-	-	-
10	0.67 ± 0.09	0.56 ± 0.14	2.67 ± 1.53	12.9 ± 2.33	9.62 ± 0.80	16.4 ± 3.99
25	-	-	-	23.4 ± 3.77	15.4 ± 2.32	31.3 ± 4.34
50	-	-	-	25.5 ± 2.25	18.1 ± 3.60	38.2 ± 6.87
100	-	-	-	28.8 ± 6.83	33.7 ± 4.46	43.4 ± 0.67

ACT-1016-3307 = M10

1) Positive control

- = no data; CYP = cytochrome P450 (isomer); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ACT-1016-3307
Location in CTD: モジュール 4.2.2.6.5
Report no. B-17.063

Type of study: Cytochrome P450 induction *in vitro*
Test system: Human hepatocytes
Method: Real-time PCR

	CYP2B6 mRNA Fold Induction (mean ± SD)					
	ACT-1016-3307			Phenobarbital [1)]		
Concentration (µmol/L)	Batch: YOW	Batch: YYX	Batch: AKB	Batch: YOW	Batch: YYX	Batch: AKB
0.3	0.42 ± 0.12	0.56 ± 0.06	1.70 ± 0.53	-	-	-
1	0.58 ± 0.03	0.57 ± 0.04	1.37 ± 0.33	-	-	-
3	0.75 ± 0.14	0.72 ± 0.09	1.82 ± 0.16	-	-	-
10	0.86 ± 0.26	0.71 ± 0.12	2.62 ± 0.93	-	-	-
100	-	-	-	0.90 ± 0.17	0.93 ± 0.17	2.13 ± 0.22
300	-	-	-	2.28 ± 0.12	2.93 ± 0.05	3.95 ± 0.91
500	-	-	-	2.33 ± 0.60	3.43 ± 1.07	9.01 ± 2.40
1000	-	-	-	4.87 ± 0.46	10.0 ± 1.95	14.5 ± 2.30
3000	-	-	-	7.33 ± 0.21	18.0 ± 6.21	23.5 ± 5.77

ACT-1016-3307 = M10
1) Positive control
- = no data; CYP = cytochrome P450 (isomer); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation.

12. 薬物動態試験：薬物代謝酵素の誘導／阻害

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.6

Report no. B-14.065

Type of study: Up-regulation of hepatic cyp mRNA in rats and dogs, and ugt mRNA in rats from liver specimen of both 4-week toxicity studies

Test system: Rat and dog livers

Method: Real-time PCR

	cyp and ugt mRNA Fold Induction (mean ± SD)					
	Male			**Female**		
Rat	**100 mg/kg/day**	**300 mg/kg/day**	**1000 mg/kg/day**	**100 mg/kg/day**	**300 mg/kg/day**	**1000 mg/kg/day**
cyp3a1	6.3 ± 3.4	17 ± 6.3	38 ± 15	12 ± 2.8	57 ± 10	141 ± 25
cyp2b1	3.6 ± 2.9	10 ± 7.6	56 ± 34	19 ± 7.4	239 ± 127	7720 ± 4187
cyp2b2	49 ± 4.0	8.1 ± 5.3	22 ± 10	2.2 ± 0.3	9.2 ± 4.2	46 ± 18
ugt1a1	1.8 ± 0.9	2.3 ± 1.1	3.8 ± 1.6	1.8 ± 0.3	3.6 ± 1.0	8.4 ± 3.0
ugt1a6	0.9 ± 0.5	0.8 ± 0.4	2.6 ± 1.1	1.6 ± 0.4	5.7 ± 2.3	6.5 ± 3.3
ugt2b1	2.8 ± 1.9	3.1 ± 1.5	7.0 ± 3.5	1.6 ± 0.5	7.3 ± 3.0	8.9 ± 3.1
	Male			**Female**		
Dog	**30 mg/kg/day**	**100 mg/kg/day**	**300/200 mg/kg/day**	**30 mg/kg/day**	**100 mg/kg/day**	**300/200 mg/kg/day**
cyp3a12	1.5 ± 0.8	2.5 ± 1.5	3.1 ± 2.5	3.1 ± 0.5	2.6 ± 1.3	5.3 ± 1.6

cyp/*cyp* = cytochrome P450 (isomer protein/*gene*); mRNA = messenger ribonucleic acid; PCR = polymerase chain reaction; SD = standard deviation; ugt/*ugt* = uridine 5'-diphospho-glucuronosyltransferase (isomer protein/*gene*).

13. 薬物動態試験：排泄

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.4.3

Report no. B-14.054

Species (Strain):	Rat (Wistar)	Rat (Wistar)
Sex / Number of animals:	Male/2	Male/2
Feeding condition:	Fed	Fed
Vehicle/Formulation:	Aqueous solution in 30% hydroxypropyl-β-cyclodextrin containing 5% (v/v) DMSO	Aqueous suspension containing 0.5% methylcellulose
Method of administration:	i.v.	p.o.
Dose (mg/kg):	1	10
Analyte:	^{14}C	^{14}C
Assay:	Liquid scintillation counting	Liquid scintillation counting

	% of Dose				% of Dose			
Time	**Urine**	**Feces**	**Bile**	**Total**	**Urine**	**Feces**	**Bile**	**Total**
0 - 4 h	0.5 - 1.3	-	76 - 82		0.4 - 0.6	0.01	47 - 58	
4 - 8 h	0.3 - 0.6	0.01	4.1 - 7.1		0.5 - 0.6	0.01	14 - 23	
8 - 24 h	0.2 - 0.3	0.9 - 1.0	1.7 - 3.6		0.2 - 0.6	4.9 - 6.1	9.5 - 19	
24 - 48 h	0.04	0.3 - 0.6	0.3		0.03 - 0.1	1.5 - 4.7	0.6 - 1.6	
48 - 72 h	0.002 - 0.03	0.02 - 0.6	0.05 - 0.08		0.02 - 0.03	0.3 - 0.5	0.1 - 0.8	
0 - 72 h	**1.3 - 1.8**	**1.1 - 1.7**	**87 - 88**	**90 - 91**	**1.5 - 1.6**	**6.7 - 11**	**81.2 - 92.5**	**94 - 101**

- = no sample; DMSO = dimethyl sulfoxide; i.v. = intravenous; p.o. = oral; v/v = volume/volume.

14. 薬物動態試験：排泄：胆汁中

単独での試験は実施していない

13 項のラット胆汁排泄試験結果を参照

15. 薬物動態試験：薬物相互作用

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.6.6

Report no. B-14.061

Type of study: Drug transporter inhibition *in vitro*

Test system: CHO cells expressing OATP1B1, OATP1B3, OAT1 or OCT2; MDCKII cells expressing P-gp/MDR1; Sf9 membrane vesicles expressing BCRP; HEK293 cells expressing OAT3, OCT1, MATE1 or MATE2-K; CHO, MDCKII and HEK293 wild-type cells

Transporter	Test System	Substrate	IC_{50} (µmol/L)
P-gp/MDR1	MDCKII cells	1 µmol/L Digoxin	24
		5 µmol/L Colchicine	21
BCRP	Sf9 vesicles	100 µmol/L Methotrexate	3.0
OATP1B1	CHO cells	0.5 µmol/L Atorvastatin acid	18
OATP1B3	CHO cells	5 µmol/L Taurocholic acid	32
OAT1	CHO cells	1 µmol/L p-aminohippuric acid	> 100
OAT3	HEK293 cells	0.5 µmol/L Estrone-3-sulfate	39
OCT1	HEK293 cells	0.5 µmol/L 1-methyl-4-phenylpyridinium (iodide)	71
OCT2	CHO cells	0.5 µmol/L 1-methyl-4-phenylpyridinium (iodide)	> 100
MATE1	HEK293 cells	100 µmol/L Metformin	9.7
MATE2-K	HEK293 cells	100 µmol/L Metformin	8.4

BCRP = breast cancer resistance protein; CHO = Chinese hamster ovary; HEK = human embryonic kidney; IC_{50} = concentration that causes 50% inhibition; MATE = multidrug and toxin extrusion protein; MDCK = Madin-Darby canine kidney; MDR = multidrug resistance protein; OAT = organic anion transporter; OATP = organic anion transporting polypeptide; OCT = organic cation transporter; P-gp = permeability glycoprotein.

15. 薬物動態試験：薬物相互作用

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.6.8

Report no. B-19.069

Type of study: OATP1B1 and OATP1B3 inhibition with a pre-incubation period *in vitro*

Test system: CHO cells expressing OATP1B1 or OATP1B3; CHO wild-type cells

Transporter	Test System	Test Article (Concentration)	Inhibitor (Concentration)	IC_{50} (µmol/L)
OATP1B1	CHO cells	Daridorexant (0.1 - 100 µmol/L)	Cyclosporin A (0.1 - 10 µmol/L)	11
OATP1B3	CHO cells	Daridorexant (0.1 - 100 µmol/L)	Cyclosporin A (0.1 - 10 µmol/L)	16

CHO = Chinese hamster ovary; IC_{50} = concentration that causes 50% inhibition; OATP = organic anion transporting polypeptide.

15. 薬物動態試験：薬物相互作用

Test article: ダリドレキサント

Location in CTD: モジュール 4.2.2.3.5

Report no. B-17.067

Type of study: Drug transporter substrate *in vitro*

Test system: CHO cells expressing OATP1B1 or OATP1B3; MDCKII cells expressing P-gp/MDR1 or BCRP; CHO and MDCKII wild-type cells

Transporter	Test System	Test Article (Concentration)	Inhibitor (Concentration)	Substrate (Yes/No)
P-gp/MDR1	MDCKII cells	Daridorexant (0.1, 0.3, 1, 3, 10 µmol/L)	Zosuquidar (1 µmol/L)	No
BCRP	MDCKII cells	Daridorexant (0.1, 0.3, 1, 3, 10 µmol/L)	Ko143 (1 µmol/L)	No
OATP1B1	CHO cells	Daridorexant (0.1, 1, 10 µmol/L)	Cyclosporin A (10 µmol/L)	No
OATP1B3	CHO cells	Daridorexant (0.1, 1, 10 µmol/L)	Cyclosporin A (10 µmol/L)	No

BCRP = breast cancer resistance protein; CHO = Chinese hamster ovary; MDCK = Madin-Darby canine kidney; MDR = multidrug resistance protein; OATP = organic anion transporting polypeptide; P-gp = permeability glycoprotein.

15. 薬物動態試験：薬物相互作用

Test article: ACT-776537, ACT-776063, ACT-1016-3307

Location in CTD: モジュール 4.2.2.6.7

Report no. B-17.049

Type of study: Drug transport inhibition *in vitro*

Test system: CHO cells expressing OATP1B1, OATP1B3, OAT1 or OCT2; MDCKII cells expressing P-gp/MDR1, MATE1 or MATE2-K; Sf9 membrane vesicles expressing BCRP; HEK293 cells expressing OAT3 or OCT1; CHO, MDCKII and HEK293 wild-type cells; Sf9 control vesicles

			IC_{50} (µmol/L)		
Transporter	**Test System**	**Substrate**	**ACT-776537**	**ACT-776063**	**ACT-1016-3307**
P-gp/MDR1	MDCKII cells	Digoxin	31	94	> 100
		Colchicine	32	> 100	> 100
BCRP	Sf9 vesicles	Methotrexate	10	21	> 100
OATP1B1	CHO cells	Atorvastatin acid	4.5	> 30	> 50
OATP1B3	CHO cells	Atorvastatin acid	> 30	> 30	> 50
OAT1	CHO cells	p-aminohippuric acid	> 30	> 30	> 50
OAT3	HEK293 cells	Estrone-3-sulfate	> 22.5	> 90	> 300
OCT1	HEK293 cells	Metformin	0.5	8.0	180
OCT2	CHO cells	1-methyl-4-phenylpyrinium (iodide)	> 30	> 30	> 50
MATE1	MDCKII cells	Metformin	16	23	230
MATE2-K	MDCKII cells	Metformin	> 22.5	100	> 300

ACT-776537 = M1; ACT-776063 = M3; ACT-1016-3307 = M10.

BCRP = breast cancer resistance protein; CHO = Chinese hamster ovary; HEK = human embryonic kidney; IC_{50} = concentration that causes 50% inhibition; MATE = multidrug and toxin extrusion protein; MDCK = Madin-Darby canine kidney; MDR = multidrug resistance protein; OAT = organic anion transporter; OATP = organic anion transporting polypeptide; OCT = organic cation transporter; P-gp = permeability glycoprotein.

15. 薬物動態試験：薬物相互作用

Test article: ACT-776537, ACT-776063, ACT-1016-3307
Location in CTD: モジュール 4.2.2.3.6
Report no. B-19.052

Type of study: Drug transporter substrate *in vitro*
Test system: MDCKI cells expressing P-gp/MDR1 or BCRP; MDCKI wild-type cells

		Average P_{app} (× 10^{-6} cm/s)		
Test Article	**Inhibitor**	**A-B**	**B-A**	**Efflux Ratio**
P-gp/MDR1				
1 µmol/L ACT-776537	-	17.9	46.2	2.6
1 µmol/L ACT-776537	2 µmol/L Elacridar	51.0	26.5	0.5
1 µmol/L ACT-776063	-	3.0	59.5	19.4
1 µmol/L ACT-776063	2 µmol/L Elacridar	40.1	30.0	0.7
1 µmol/L ACT-1016-3307	-	0.2	27.3	129
1 µmol/L ACT-1016-3307	2 µmol/L Elacridar	2.0	2.0	1.0
BCRP				
1 µmol/L ACT-776537	-	40.0	32.5	0.8
1 µmol/L ACT-776537	0.5 µmol/L Ko143	42.9	37.4	0.9
1 µmol/L ACT-776063	-	18.5	46.2	2.5
1 µmol/L ACT-776063	0.5 µmol/L Ko143	22.5	51.7	2.3
1 µmol/L ACT-1016-3307	-	0.4	14.3	33.9
1 µmol/L ACT-1016-3307	0.5 µmol/L Ko143	0.4	14.1	35.4

ACT-776537 = M1; ACT-776063 = M3; ACT-1016-3307 = M10.

A-B = apical to basolateral; B-A = basolateral to apical; BCRP = breast cancer resistance protein; MDCK = Madin-Darby canine kidney; MDR = multidrug resistance protein; P_{app} = apparent absorptive permeability coefficient; P-gp = permeability glycoprotein.

16. 薬物動態試験：その他

その他の試験は実施しなかった