Discovery of the Novel, Orally Active $S1P_1$ Receptor Agonist ACT-128800 (Ponesimod)

Martin H. Bolli, Actelion Pharmaceuticals Ltd

3. Wädenswiler Chemie-Tag, 16. Juni 2011

ACTELION

Discovery of the Novel, Orally Active $S1P_1$ Receptor Agonist ACT-128800 (Ponesimod)

- Drug Discovery Chemistry at Actelion
- $S1P_1$ Agonist Biology & Pharmacology
- The Discovery of Ponesimod
  - Synthesis of iminothiazolidinones
  - SAR leading to ponesimod
- Ponesimod Properties
- Summary

Actelion 1997

- founded in Allschwil, BL by
  - Jean-Paul Clozel
  - Martine Clozel
  - Thomas Widmann
  - André J. Mueller
  - Walter Fischli

- Vision: A company based on two pillars
  - develop and sell bosentan (in-licensed from Roche)
  - discover and develop own novel drugs

Actelion Earns Money by Sellings Drugs to Treat

- Pulmonary Arterial Hypertension
  - oral
  - inhaled
  - intravenous
- Gaucher Type I and Niemann-Pick C disease

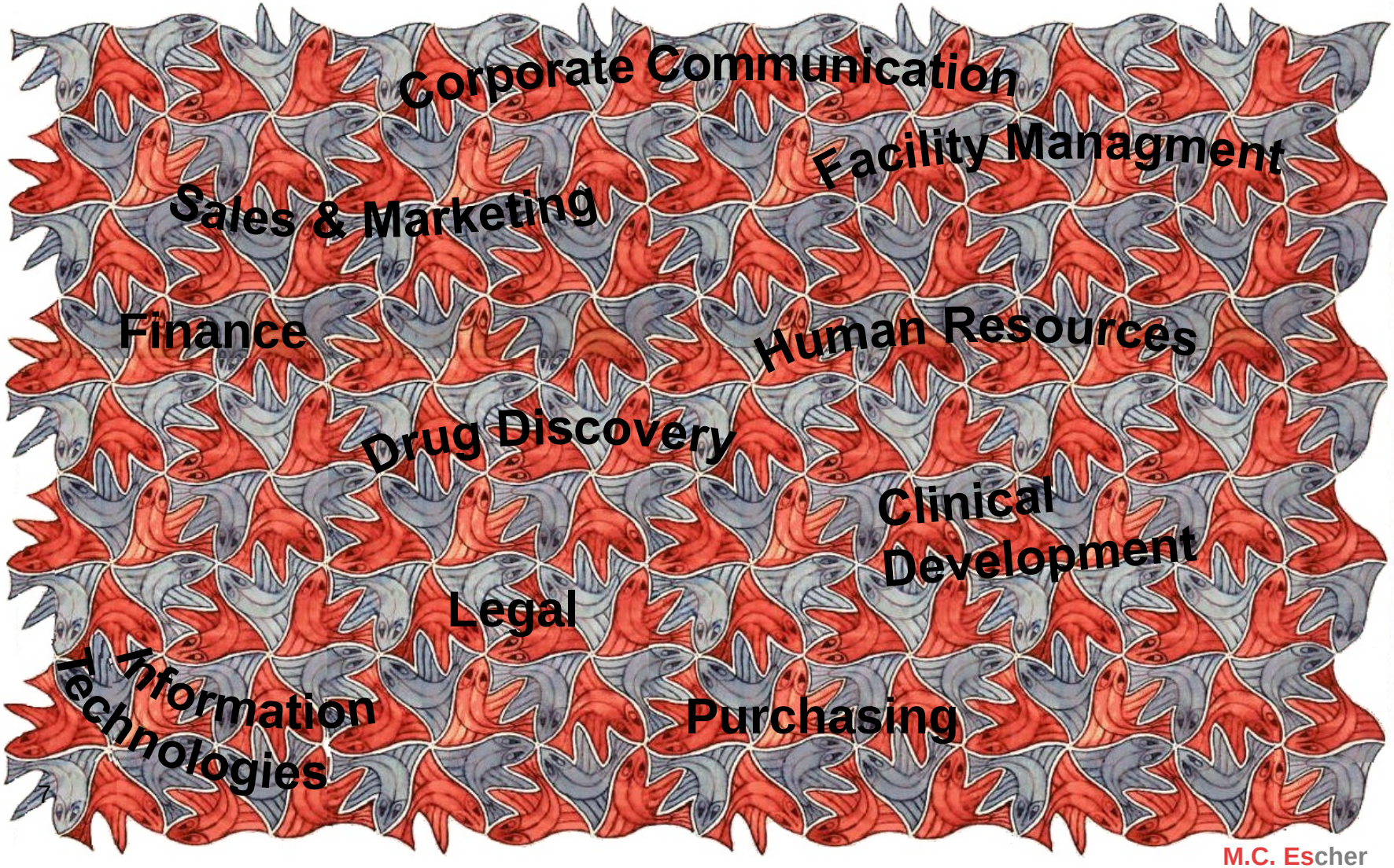
… and Invests Money into the Discovery and Development of Novel Drugs

- Endothelin Antagonists
- Orexin Antagonists
- $CRTH_2$ Antagonists
- Sphingosine 1-Phosphate Agonists
- Renin Inhibitors
- Antibiotics
- …

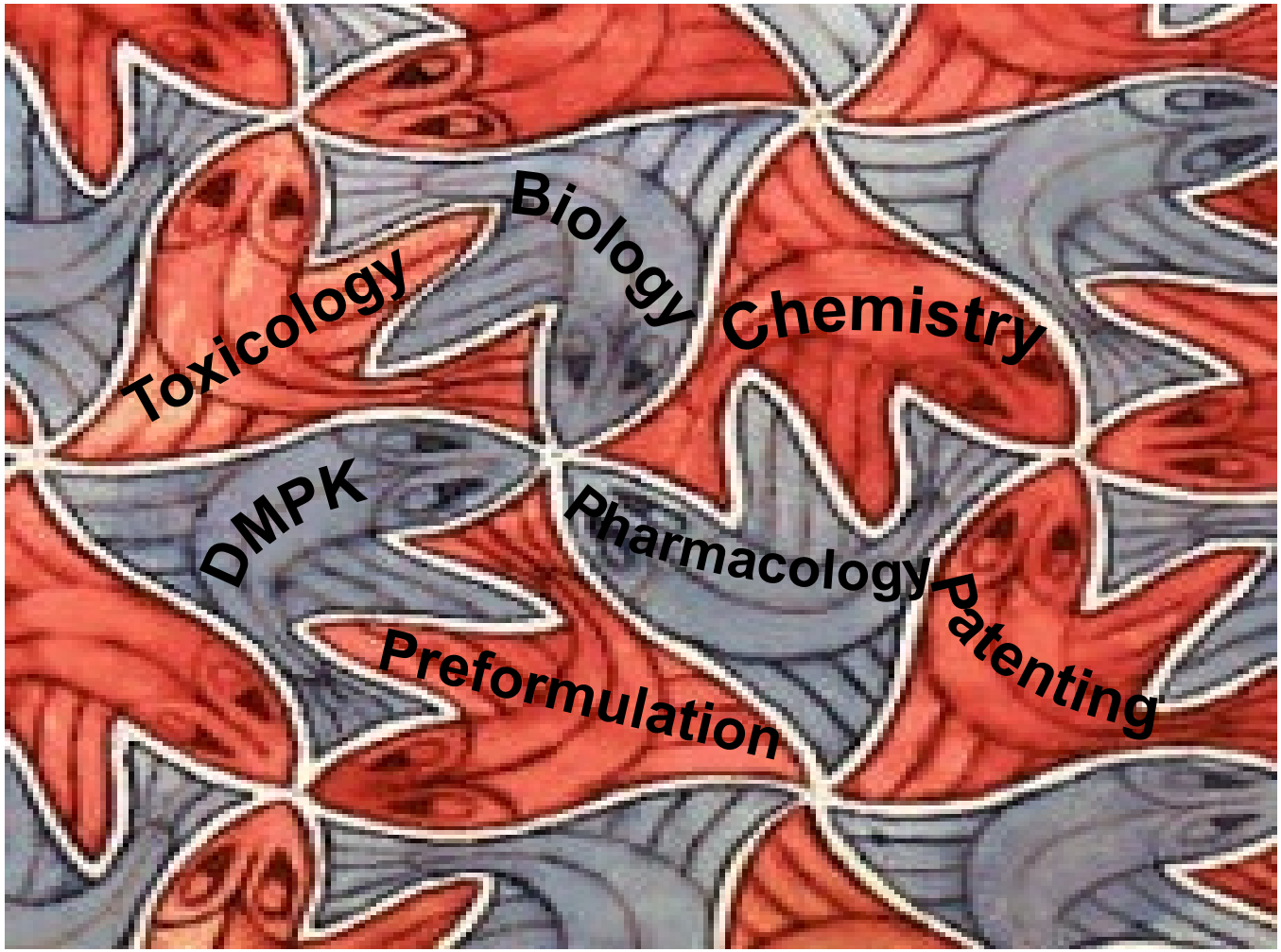
Actelion 2011
Strong Commitment to Research and Development

	Employees	
Drug Discovery	401	Allschwil
Clinical Develoment	643	
Support Functions	420	
Marketing & Sales	1029	Allschwil + subsidiaries in >25 countries

Actelion
Network of Departments



Drug Discovery
Network of Disciplines



Drug Discovery Chemistry

Medicinal Chemistry

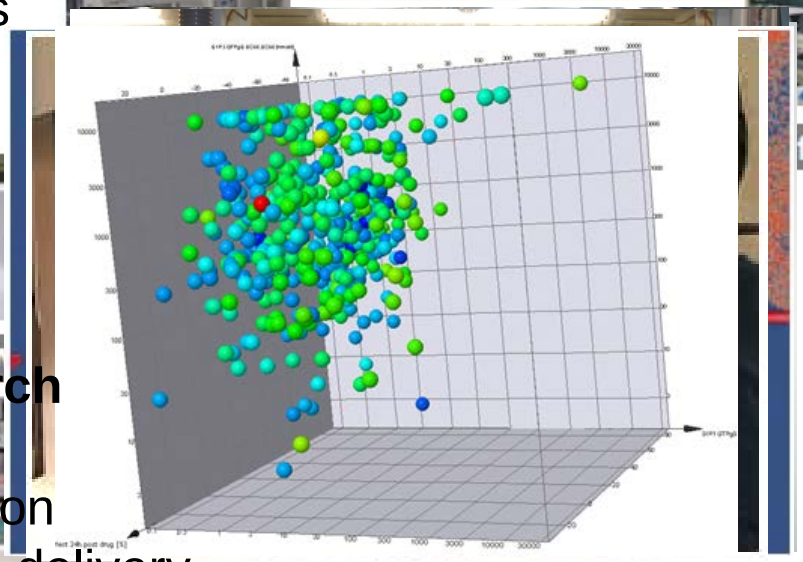
- ‚one by one‘
- small libraries

Open Access Instrumentation

- anal. LC-MS
- prep. HPLC
- NMR
- *chiral separation*
- *SFC*

Drug Discovery Chemistry

Medicinal Chemistry
- ‚one by one'
- small libraries



Open Access Instrumentation
- anal. LC-MS
- prep. HPLC
- NMR
- *chiral separation*
- *SFC*

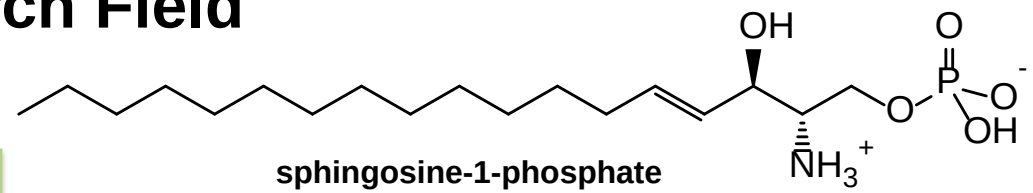
Process Research
- route finding
- route optimisation
- drug substance delivery

Computational Chemistry
- crystallography
- molecular modeling
- property calculation
- HTS hit clustering

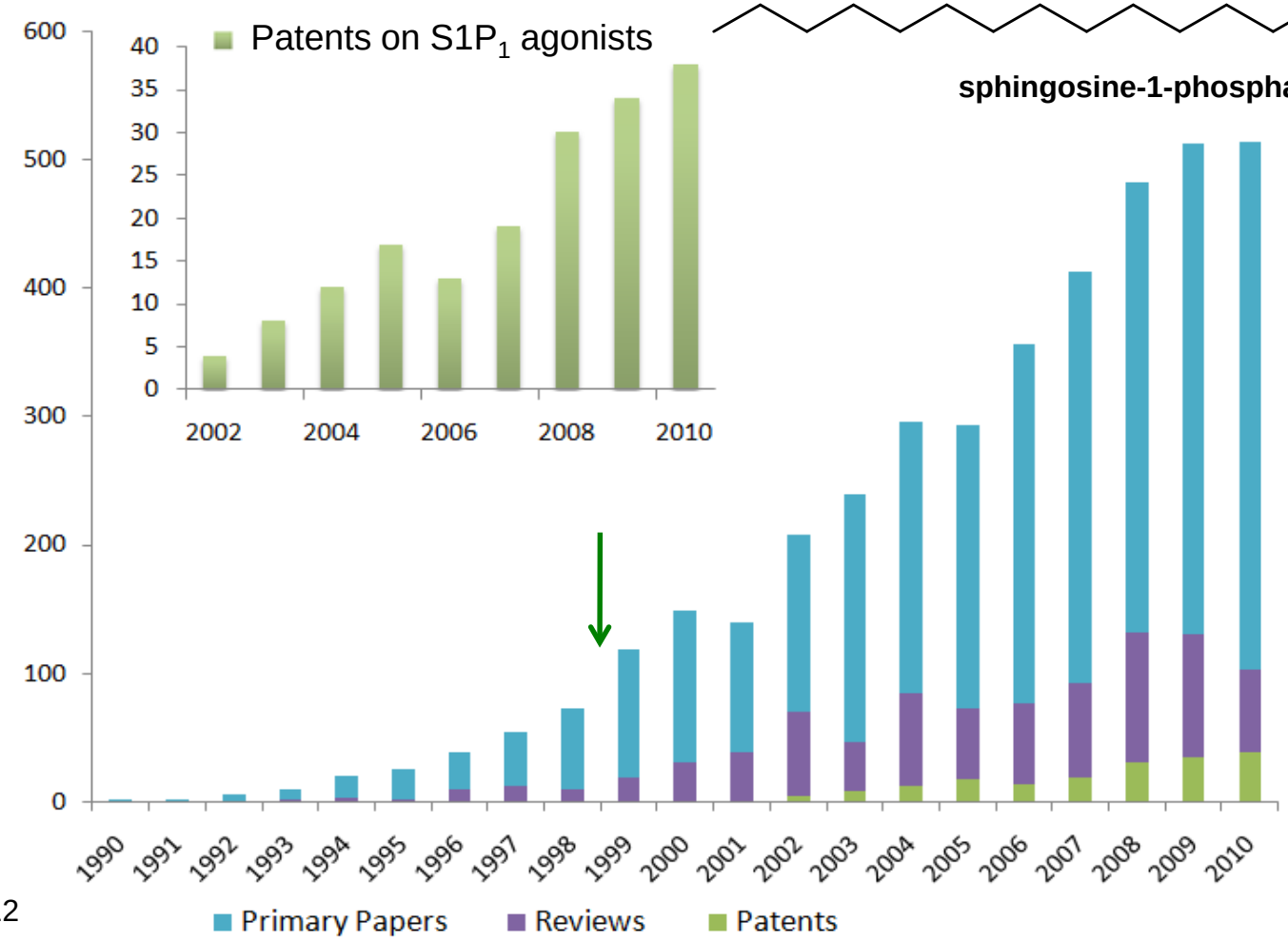
High Throughput Medicinal Chemistry
- large compound libraries

Now Its Time for Science!

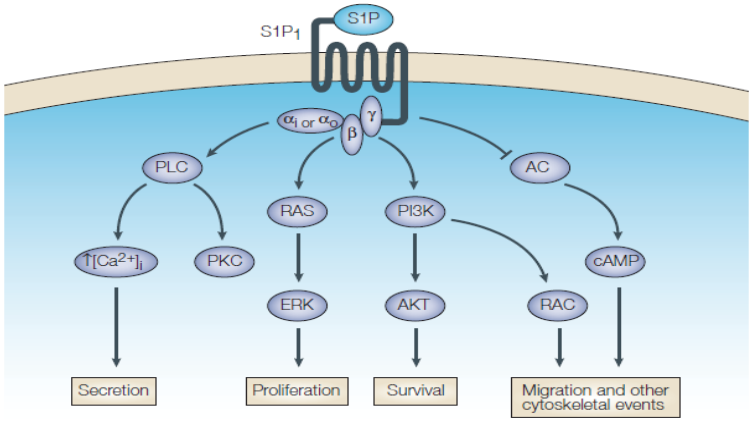
Sphingosine-1-Phosphate
A Highly Active Research Field

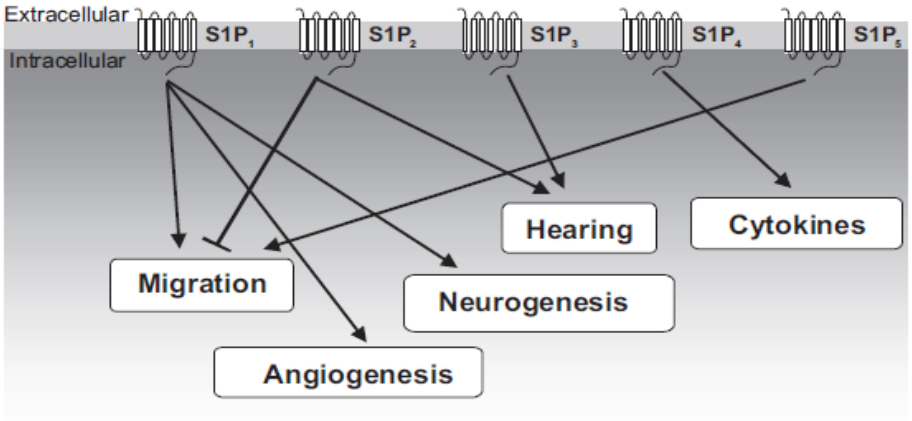


sphingosine-1-phosphate



S1P Receptor Biology & Pharmacology



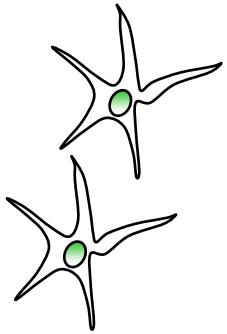


$S1P_1$	$S1P_2$	$S1P_3$	$S1P_4$	$S1P_5$
ubiquitous	ubiquitous	ubiquitous	lymphoid and haematopoietic tissue	brain: astrocytes, oligodendrocytes; skin,
cell migration↑, proliferation, survival, angiogenesis, **lymphocyte trafficking**, endothelial barrier enhancement, vascular maturation and tone, astrocyte mediated neuroprotection, neurogenesis	cell migration↓, vascular development	**heart rate↓, vasoconstriction**, vascular development, macrophage and monocyte migration, B cell tolerance induction and maturation,	cell migration ↑↓, proliferation and cytokine secretion in T cells ↓,	proliferation↓, survival of mature oligodendrocytes, astrocyte stimulation, remyelination,

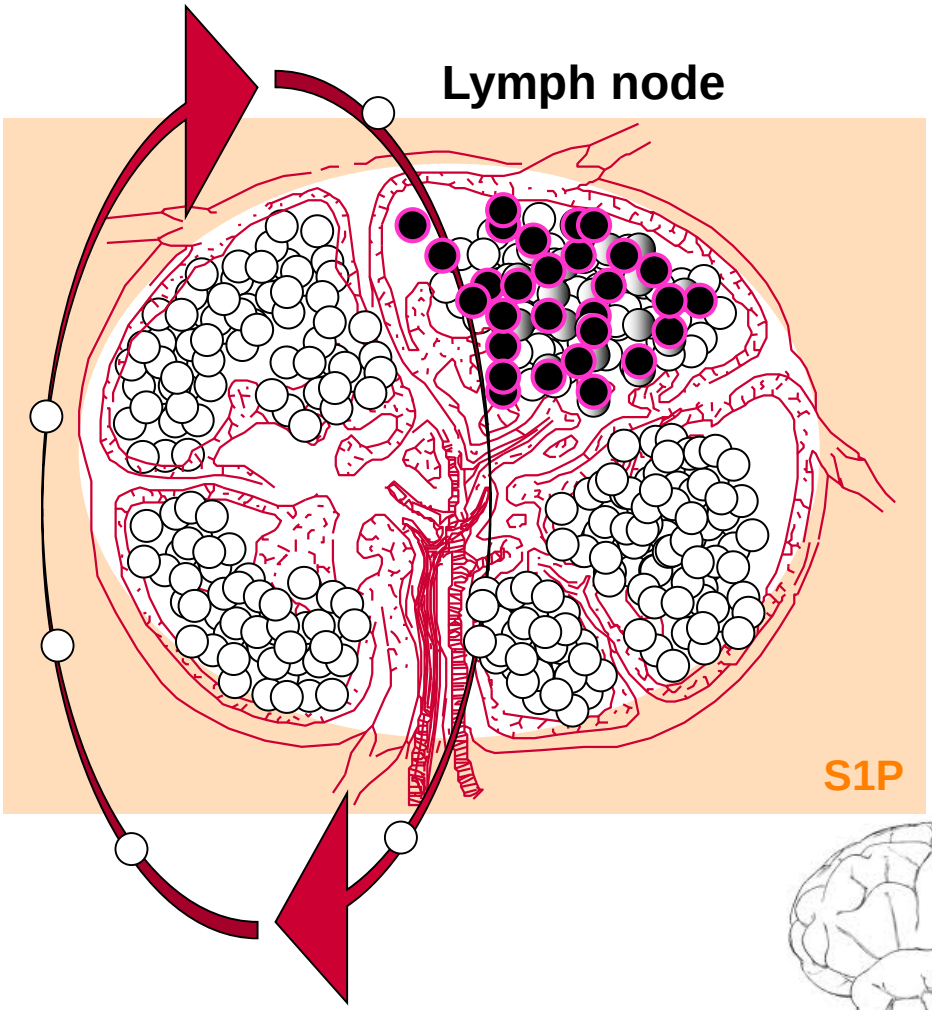
Rosen et. al. Nat. Rev. Immunol. **5** (2005) 560-570
K. Takabe et al. *Pharmacol. Rev.* **60**(2008) 181-195

T Cells Circulate Through the Body

Antigen-presenting cell



Lymph node



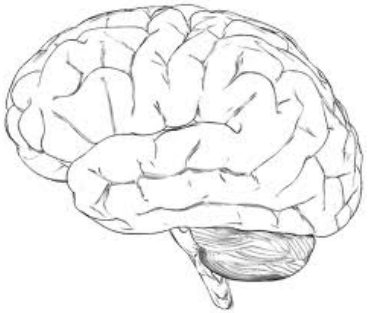
Lymph system

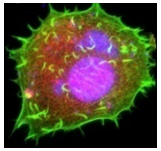
Naïve T cell

Activated T cell

Effector T cell

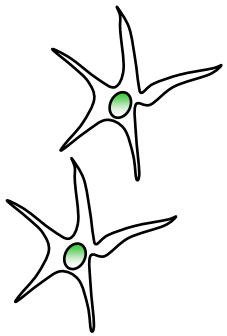
Infected Cell
Target organ



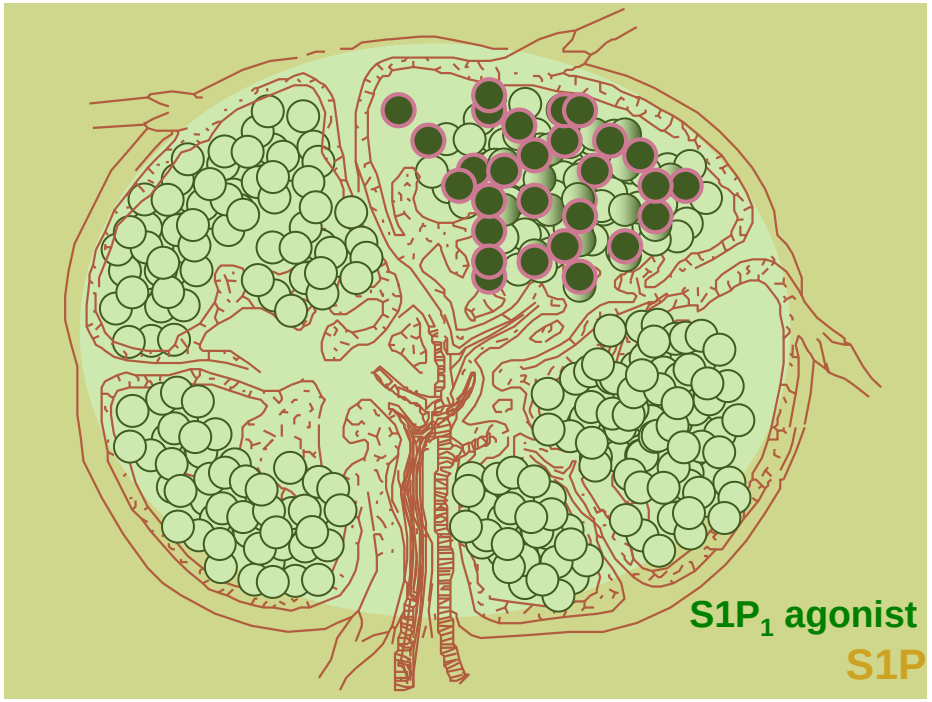


$S1P_1$ Agonists Sequester T Cells in Lymphnodes

Antigen-presenting cell



Lymph node



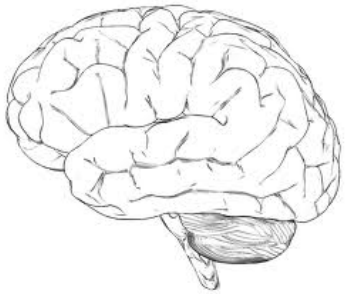
Naïve T cell

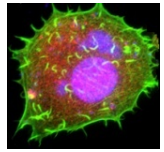
Activated T cell

Effector T cell

Lymph system

Infected Cell
Target organ





The NEW ENGLAND JOURNAL of MEDICINE

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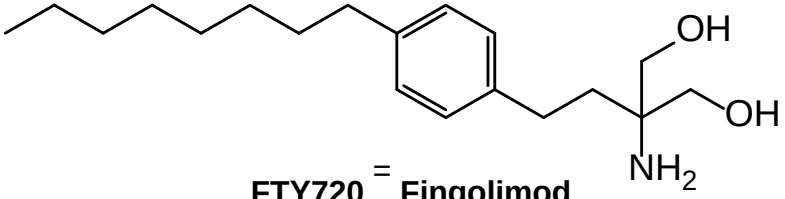
A Placebo-Controlled Trial of Oral Fingolimod in Relapsing Multiple Sclerosis

Ludwig Kappos, M.D., Ernst-Wilhelm Radue, M.D., Paul O'Connor, M.D., Chris Polman, M.D., Reinhard Hohlfeld, M.D., Peter Calabresi, M.D., Krzysztof Selmaj, M.D., Catherine Agoropoulou, Ph.D., Malgorzata Leyk, Ph.D., Lixin Zhang-Auberson, M.D., Ph.D., and Pascale Burtin, M.D., Ph.D., for the FREEDOMS Study Group*

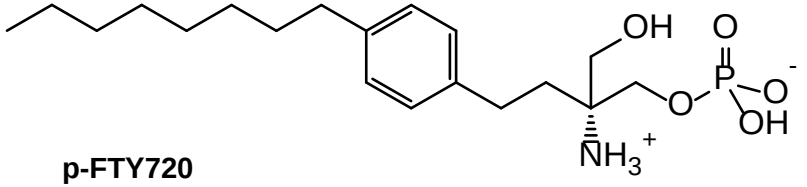
Oral Fingolimod or Intramuscular Interferon for Relapsing Multiple Sclerosis

Jeffrey A. Cohen, M.D., Frederik Barkhof, M.D., Giancarlo Comi, M.D., Hans-Peter Hartung, M.D., Bhupendra O. Khatri, M.D., Xavier Montalban, M.D., Jean Pelletier, M.D., Ruggero Capra, M.D., Paolo Gallo, M.D., Guillermo Izquierdo, M.D., Klaus Tiel-Wilck, M.D., Ana de Vera, M.D., James Jin, Ph.D., Tracy Stites, Ph.D., Stacy Wu, M.D., Shreeram Aradhye, M.D., and Ludwig Kappos, M.D., for the TRANSFORMS Study Group*

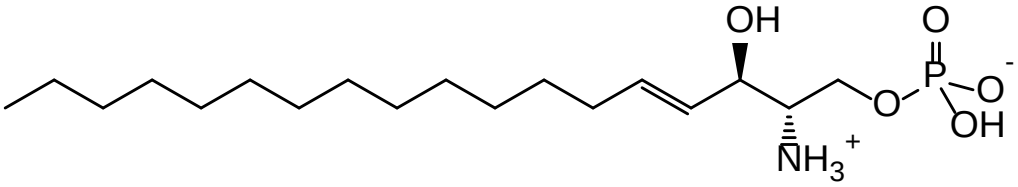
FTY720 (Fingolimod, Gilenya™)
A First Non-selective $S1P_1$ Agonist on the Market



FTY720 = Fingolimod



p-FTY720



Sphingosine 1-phosphate

- FTY720 is phosphorylated *in vivo*
- p-FTY720 is a potent agonist of $S1P_{1,\ 3,\ 4,\ and\ 5}$
- FTY720 has a long half-life in vivo

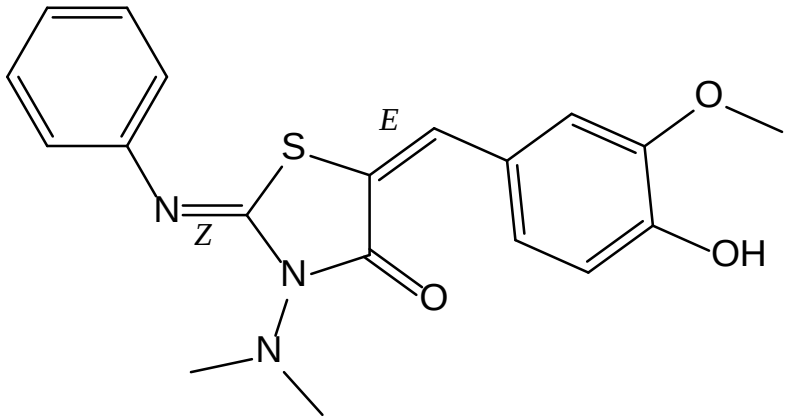
S1P Project Goal

- potent, orally active $S1P_1$ receptor agonist
- selective against $S1P_3$
- rapid on-set of action and rapid reversibility *in vivo*
- use blood lymphocyte counts as biomarker

The Discovery of Ponesimod

HTS, Synthesis & SAR Studies

High Throuhput Screening Hit



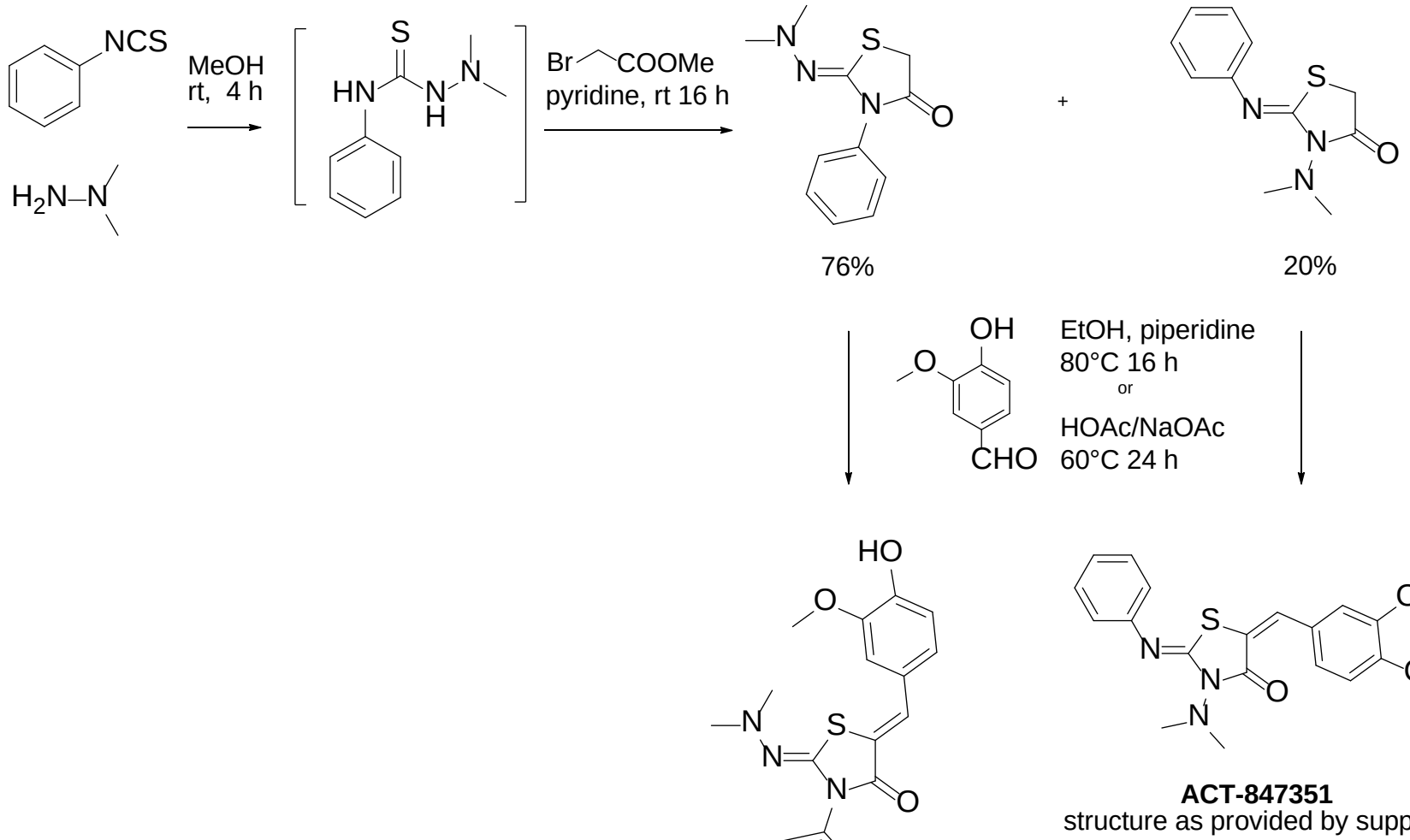
ACT-847351

structure as provided by supplier

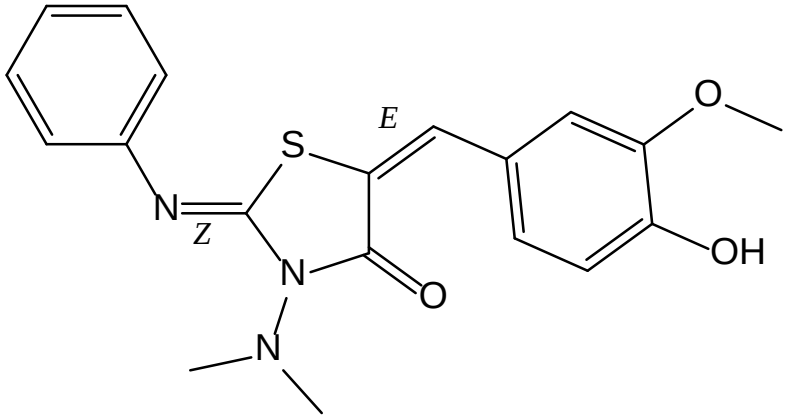
NMR shows presence of only one compound

EC_{50} hS1P$_1$ GTPγS 144 nM

Synthesis of ACT-847351



High Throuhput Screening Hit
A First Surprise



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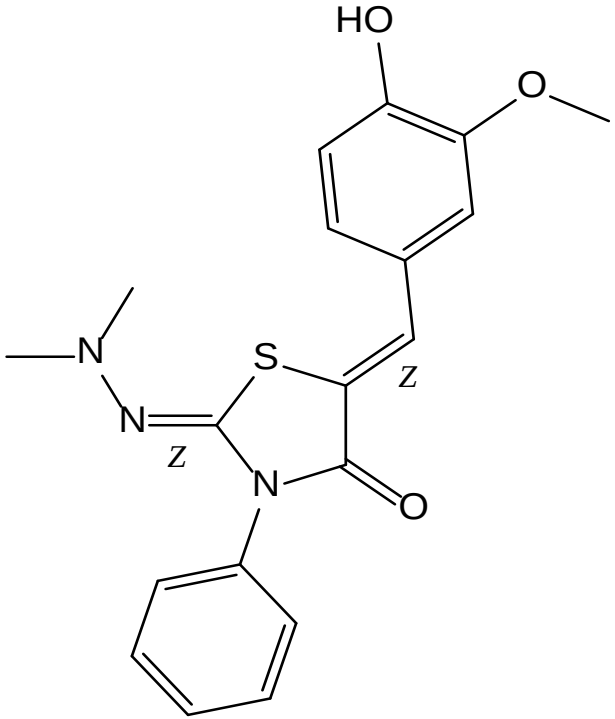
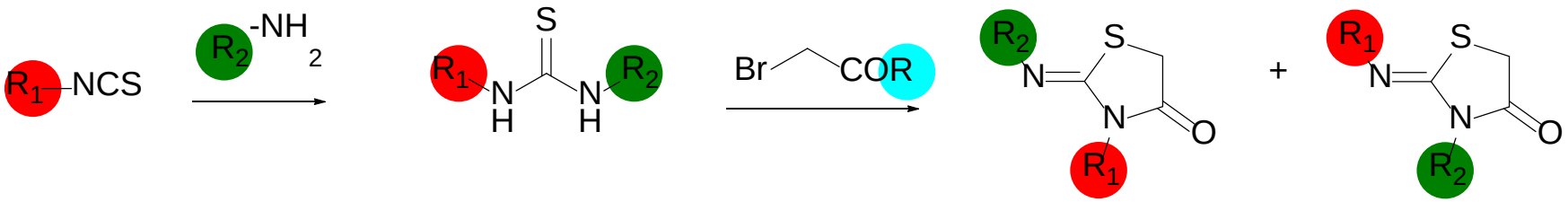


ACT-847351

the correct structure

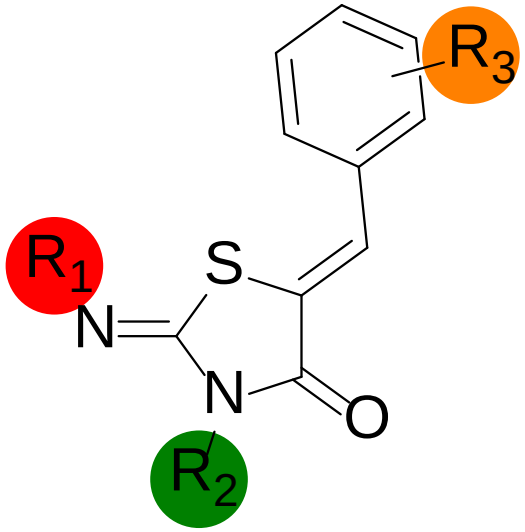
Iminothiazolidinone Synthesis
Summary Regioselectivity



R	R_1	R_2	A		B
OCH_3	non- or 3- or 4-substituted phenyl	branched	10 - 20	:	1
OCH_3	2-substituted phenyl	branched	3-4	:	1
OCH_3	2,6-disubstituted phenyl	branched	1	:	~13
OCH_3	any substituted phenyl	straight	1	:	45 - >150
Br	any substituted phenyl	straight or branched	>20	:	1

→ bromo acetylbromide reactant of choice

SAR of Iminothiazolidinone Derivatives as $S1P_1$ Receptor Agonists

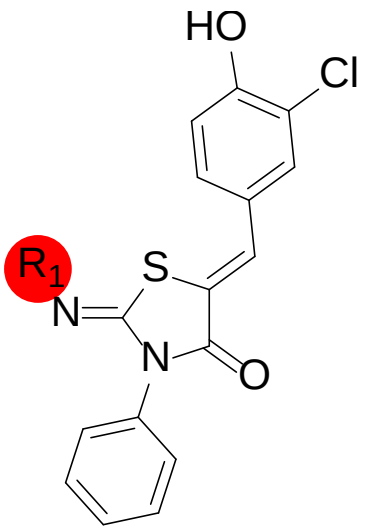


GTPγS assay

- potency on $S1P_1$
- selectivity against $S1P_3$

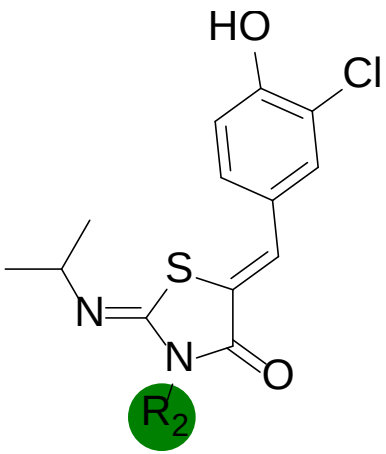
often flat SAR

SAR of Iminothiazolidinone Derivatives
Influence of R_1



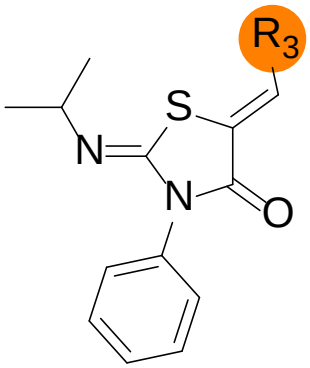
R_1	$S1P_1$ EC_{50} [nM]	$S1P_3$ EC_{50} [nM]
H	>10000	>10000
methyl	990	8810
ethyl	186	1229
n-propyl	67	189
n-butyl	112	264
iso-propyl	47	120
tert.-butyl	147	96
cyclopropyl	103	114
cyclobutyl	202	128
cyclopentyl	347	302

SAR of Iminothiazolidinone Derivatives
Influence of R_2



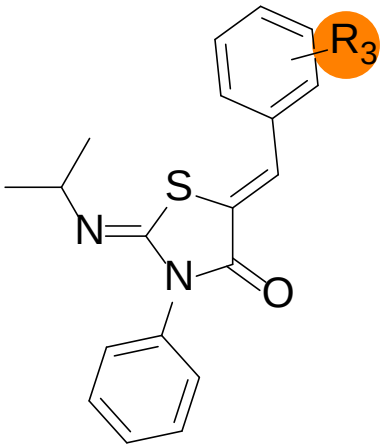
R_2	$S1P_1$ EC_{50} [nM]	$S1P_3$ EC_{50} [nM]
phenyl	47	120
2-methyl-phenyl	34	139
2-chloro-phenyl	54	425
2-methoxy-phenyl	106	428
2,6-dimethyl-phenyl	154	307
3-methyl-phenyl	110	200
3-chloro-phenyl	35	129
2-methyl-3-chloro-phenyl	31	246
4-methyl-phenyl	78	88
benzyl	630	674
phenethyl	925	183

SAR of Iminothiazolidinone Derivatives
Influence of R_3



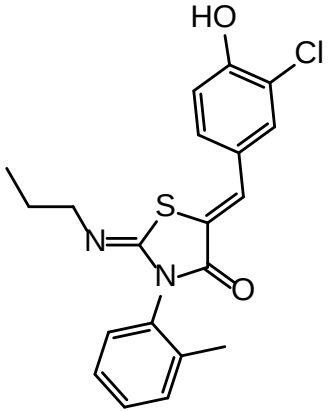
R_3	$S1P_1$ EC_{50} [nM]	$S1P_3$ EC_{50} [nM]
phenyl	1237	317
2-pyridinyl	863	n.d.
3-pyridinyl	6746	n.d.
4-pyridinyl	1520	n.d.
4-quinolinyl	1749	n.d.
3-indolyl	5046	n.d.
phenethyl	541	976
cyclohexyl	>10000	n.d.
ethyl	>10000	n.d.
cyclopropyl	>10000	n.d.

SAR of Iminothiazolidinone Derivatives
Influence of R_3

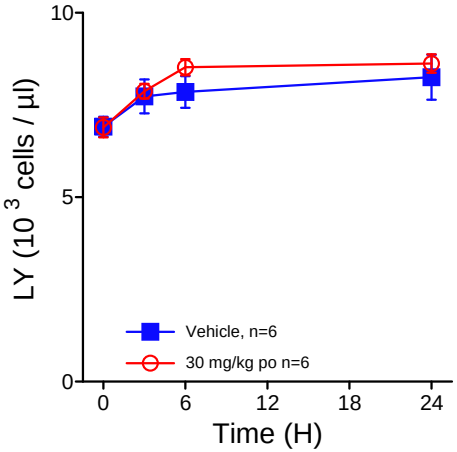


R_3	$S1P_1$ EC_{50} [nM]	$S1P_3$ EC_{50} [nM]
H	1237	317
2-methoxy	>10000	2583
3-methoxy	185	35
3-hydroxy	752	103
4-methoxy	201	106
4-hydroxy	122	95
4-hydroxy-3-fluoro	225	99
4-hydroxy-3-chloro	47	120
4-hydroxy-3-methyl	37	50
4-hydroxy-3-methoxy	200	335

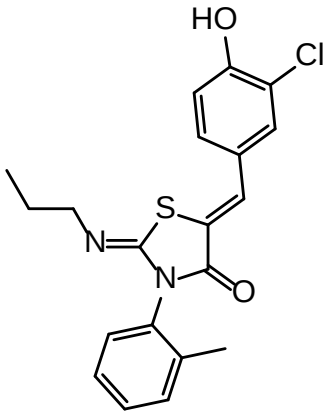
Combining Best Elements

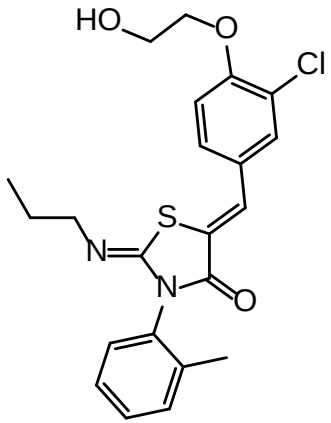


EC_{50} GTP γS	$S1P_1$ [nM]	19
	$S1P_3$ [nM]	81
		21

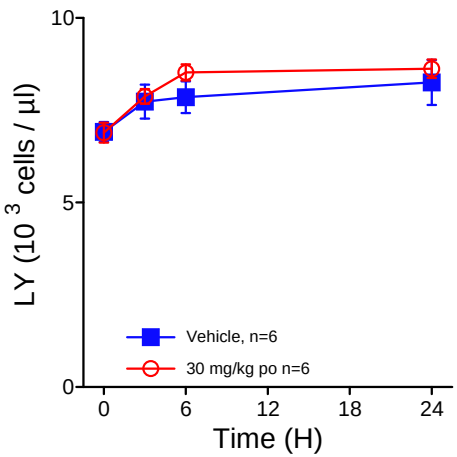


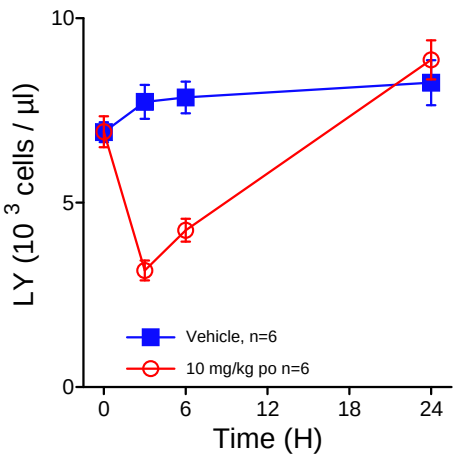
Combining Best Elements

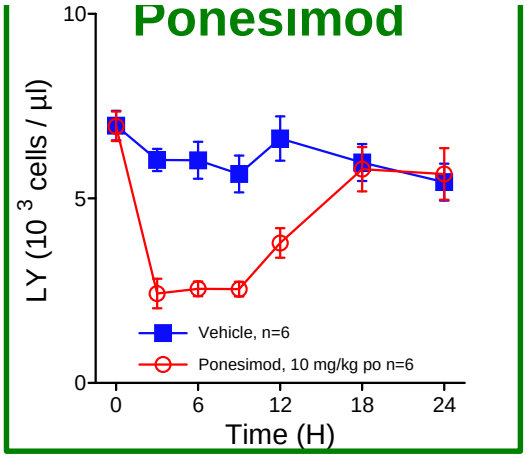




GTP γS	EC_{50} S1P	[nM]	19	11
	EC_{50} $S1P_1$	[nM]	81	124
	EC_{50} $S1P_3$	[nM]	21	299
PK rat (dog) (3 mg/kg)	c_{max}	[h]	0.2	
	$t_{1/2}$	[mL/min/kg]	92	
	CL	[%]	<1	
	F			







Ponesimod Properties

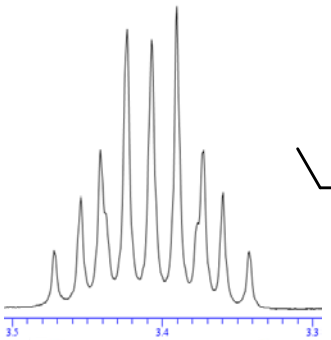
Chemistry Biology Pharmacology

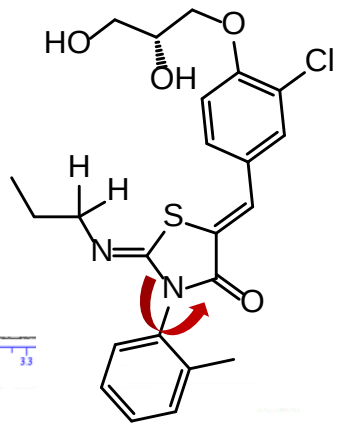
Ponesimod Chemistry

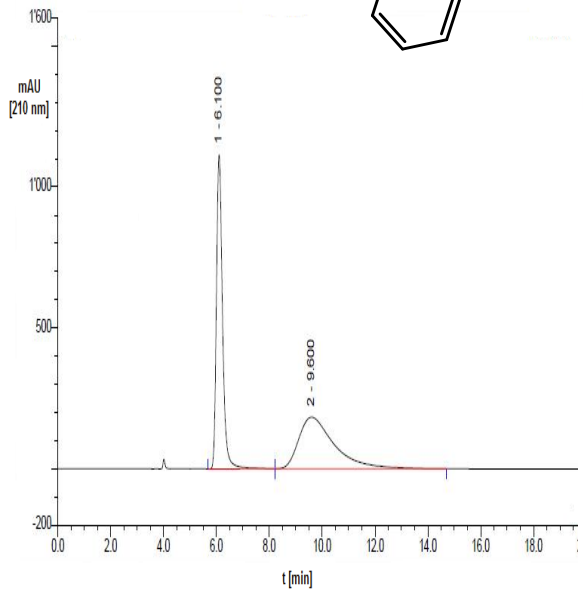
Isomers, Isomers, Isomers

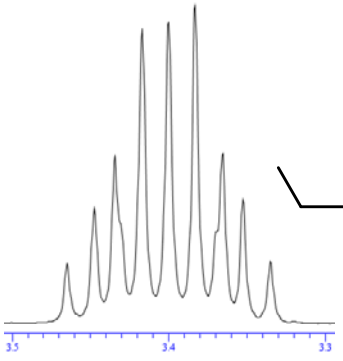
Ponesimod

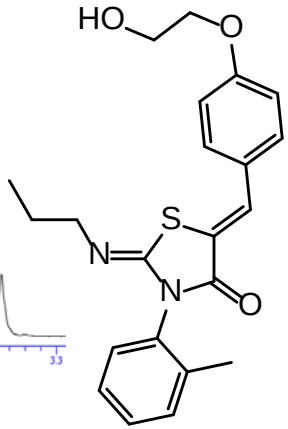
^{1}H NMR and Chiral Chromatography

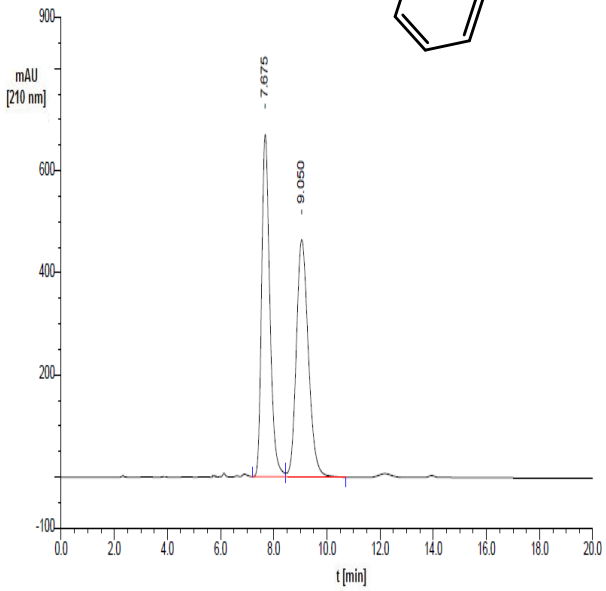


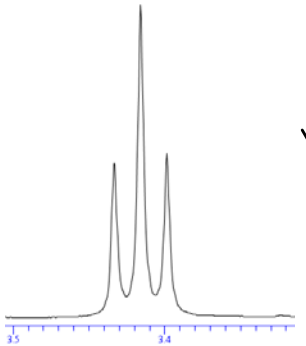


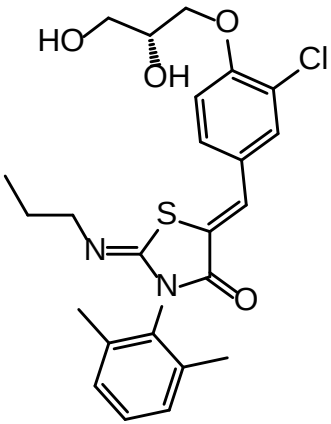


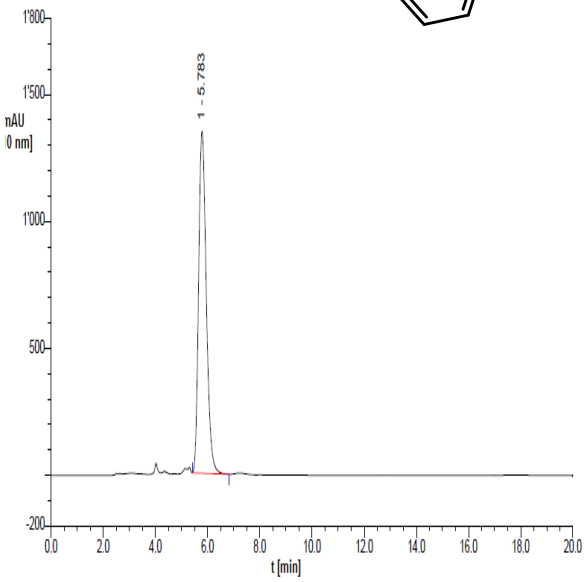




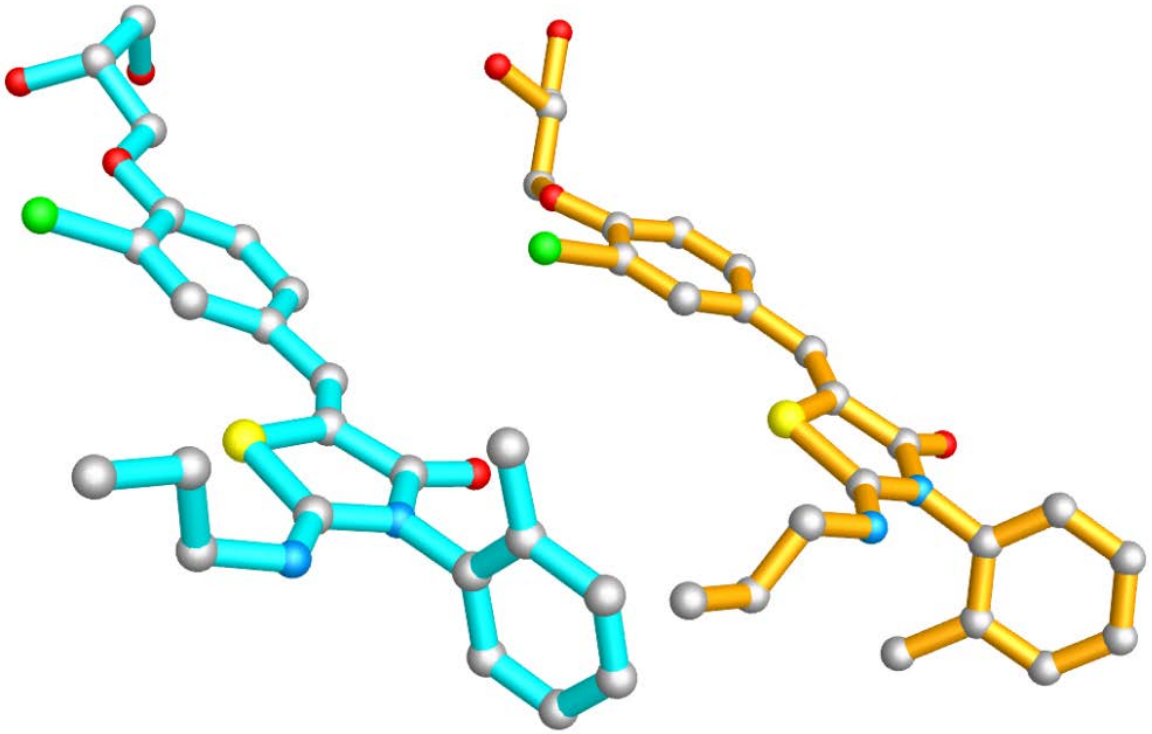






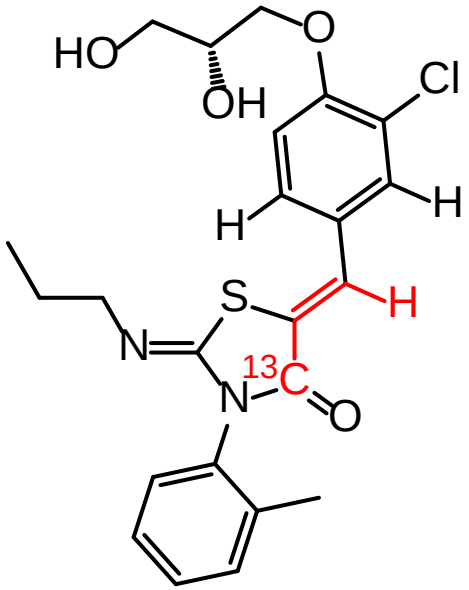


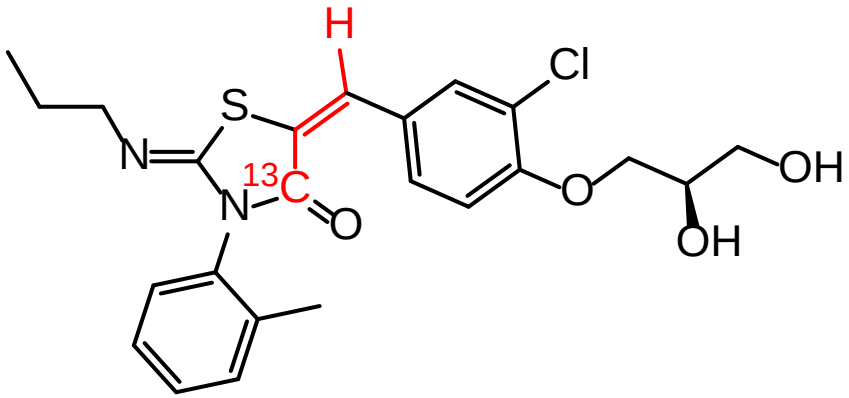
Ponesimod X-ray



Ponesimod in Solution
LC-NMR Studies

- When solutions of ponesimod are exposed to light, formation of an isomer is observed
- This isomer is in equilibrium with ponesimod





Ottana et al. *Bioorg. Med. Chem.* **13**(2005)4243-4252
Hansen *Prog. NMR Spectr.* **14**(1981) 175-296

	ponesimod	6.3 Hz
$^3J_{CH}$	□□□	

A. Preiß, M. Elend

Ponesimod Biology

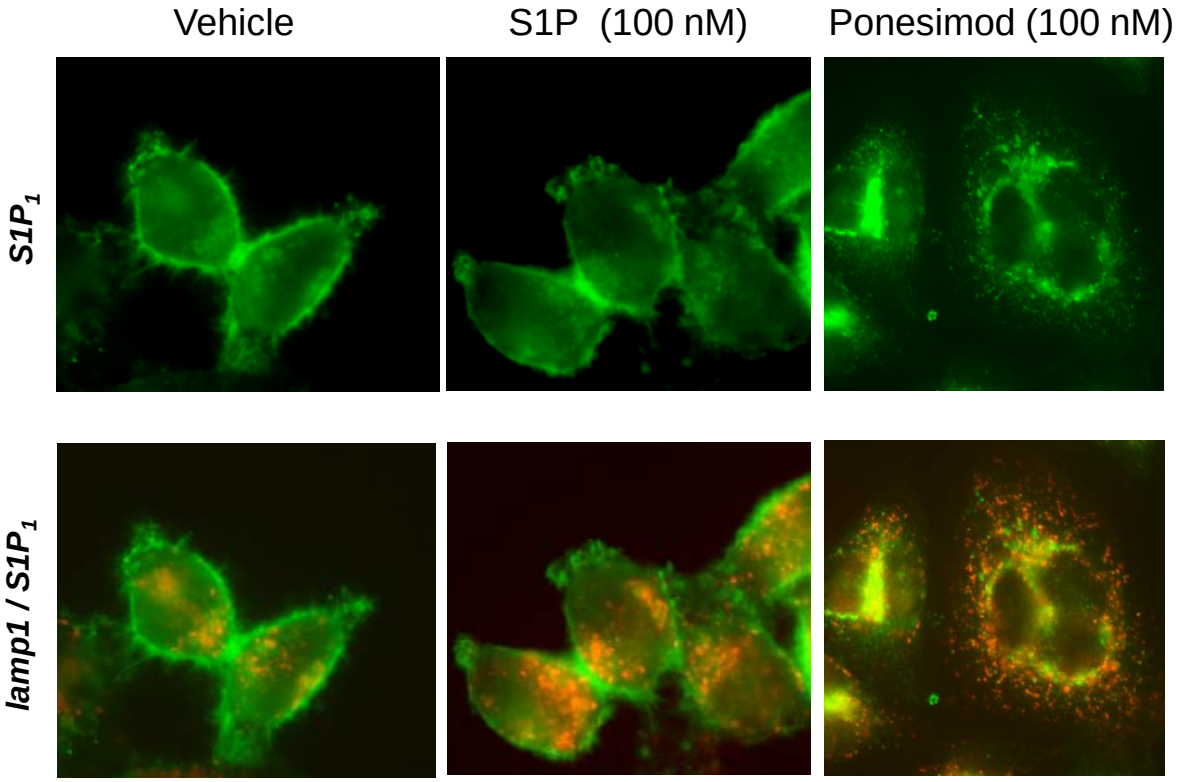
Ponesimod
Potency and Selectivity

GTPγS EC_{50} [nM][a]*				
$S1P_1$	**$S1P_2$**	**$S1P_3$**	**$S1P_4$**	**$S1P_5$**
5.7	>10000	105	1108[c]	59.1[c]

→ potent and selective $S1P_1$ agonist

[a]CHO-S1P membranes; [b]Hela-$S1P_1$ cells; [c]partial agonist; *values are geomeans, n ≥ 3

$S1P_1$ Receptor Internalisation
Hela-$S1P_1$ Cells After 24 h Incubation



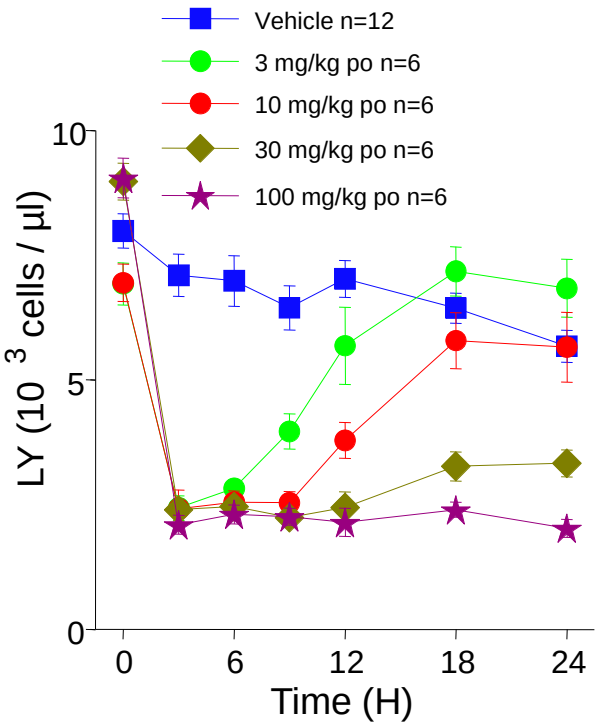
→ ponesimod: efficient $S1P_1$ receptor internalisation and lysosomal targeting

Ponesimod Biology Summary

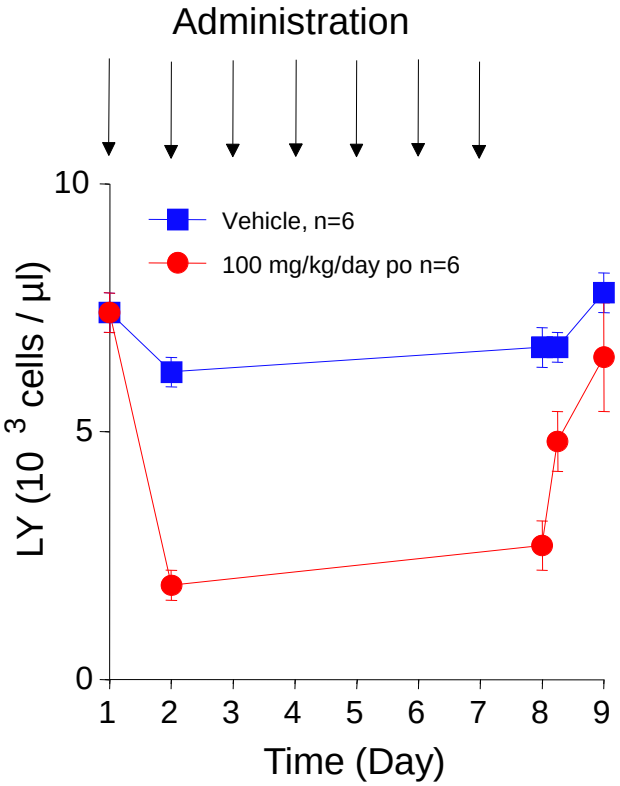
	S1P	Ponesimod
Potent & efficient Gα – mediated $S1P_1$ signaling	+	+
Efficient $S1P_1$ receptor internalization	-	+
$S1P_1$ receptor transfer to lysosomes	-	+
$S1P_1$ receptor degradation	-	+
Desensitization / functional antagonism	-	+

Ponesimod Pharmacology

Ponesimod – In Vivo Pharmacology
Single and Multiple Dosing in Male Wistar Rats



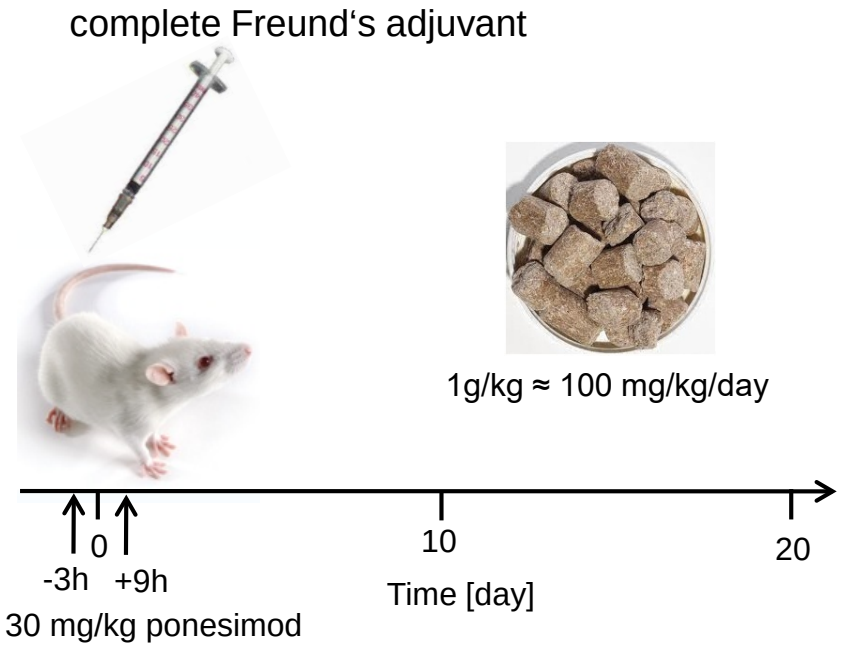
→ rapid and dose dependent LC reduction

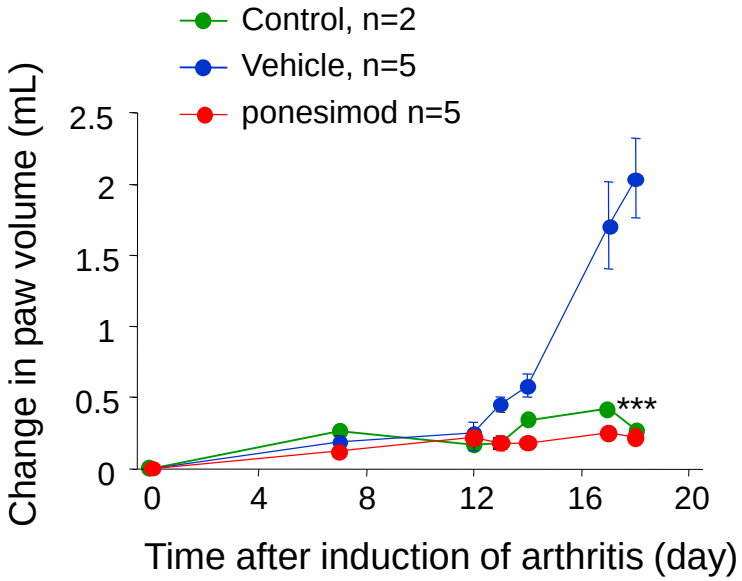


→ rapid reversibility of LC reduction

Ponesimod – In vivo Pharmacology
Rheumatoid Arthritis Model

- Ponesimod was shown to be efficacious in several animal models of autoimmune diseases e.g.
  - Adjuvant-induced arthritis (AIA) model





Summary

Iminothiazolidinones

- SAR leading to the identification of ponesimod
- Regio-isomers, constitutional isomers, atropisomers

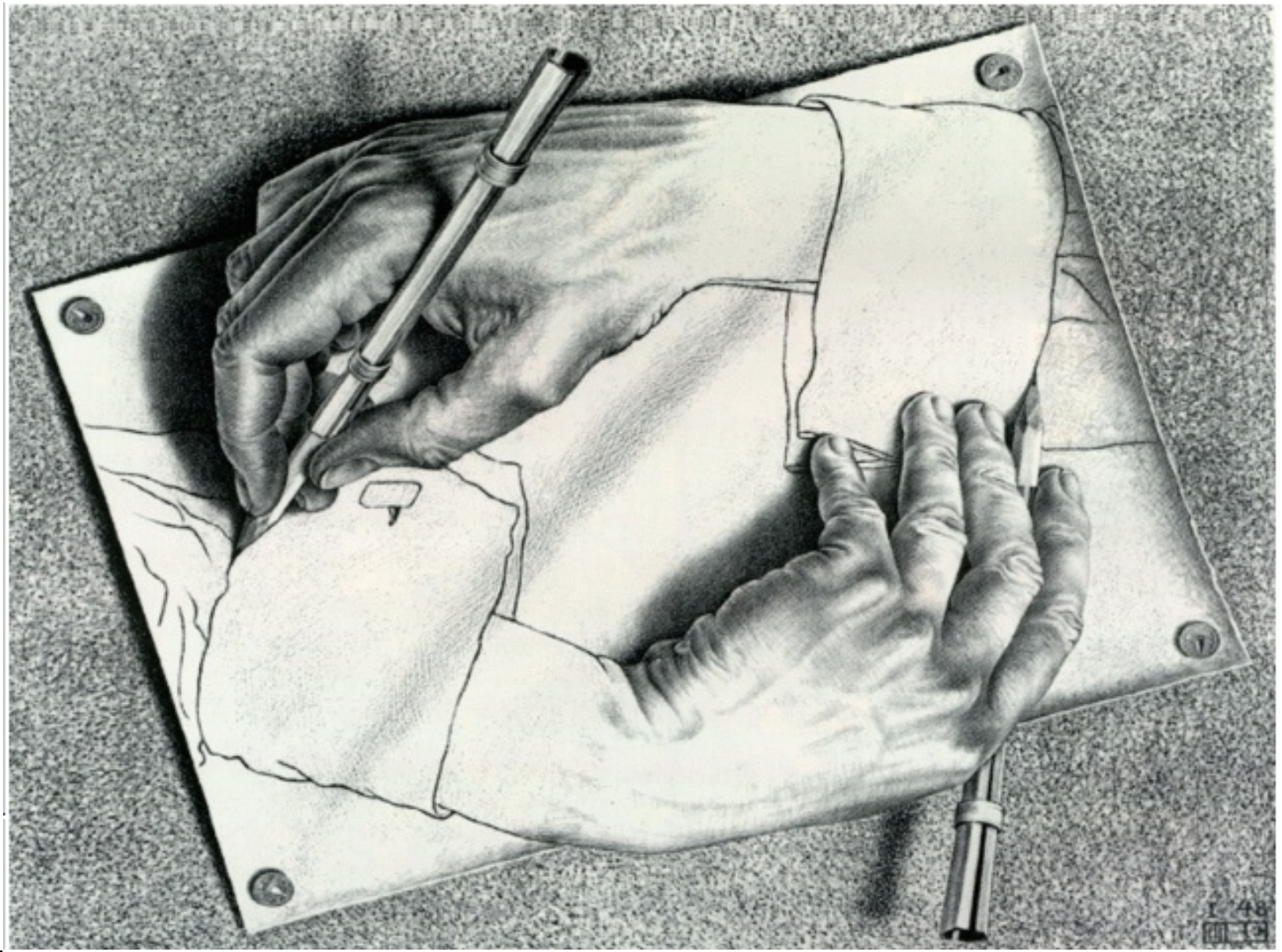
Ponesimod

- is a potent, selective $S1P_1$ receptor agonist
- leads to $S1P_1$ receptor internalisation and degradation
- acts as a functional receptor antagonist
- reduces blood lymphocyte counts with rapid onset and reversibility
- is efficacious in models of autoimmune diseases
- is in Phase II clinical trials for MS and psoriasis

Bolli et al. *J. Med. Chem.* **53**(2010) 4198-4211

Piali et al. *J. Pharmacol. Exp. Ther.* ***337(***2011) 547-556

Bolli et al. *Curr. Top. Med. Chem.* ***11****(2011) 726-757*



M. C. Escher

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