Thalidomide and Its Chirality: Towards Paradox

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Pharmacy –
a journey from Edo times to modern pharmaceuticals and health economics

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Thalidomide's Return

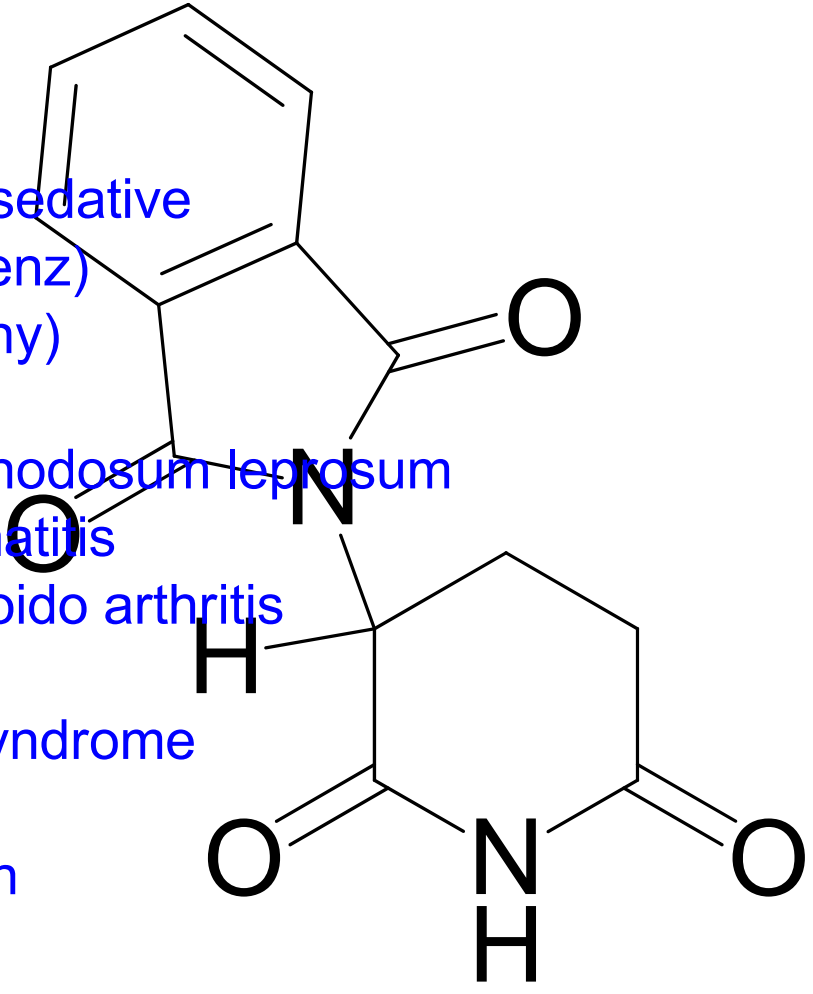
Towards Paradox

Three decades after maiming thousands of children, the drug is increasingly useful- but perilous as ever.

Time 37, June 13, **1994**

Thalidomide (Contergan, Distaval) : From Tragedy to New Drug Discovery

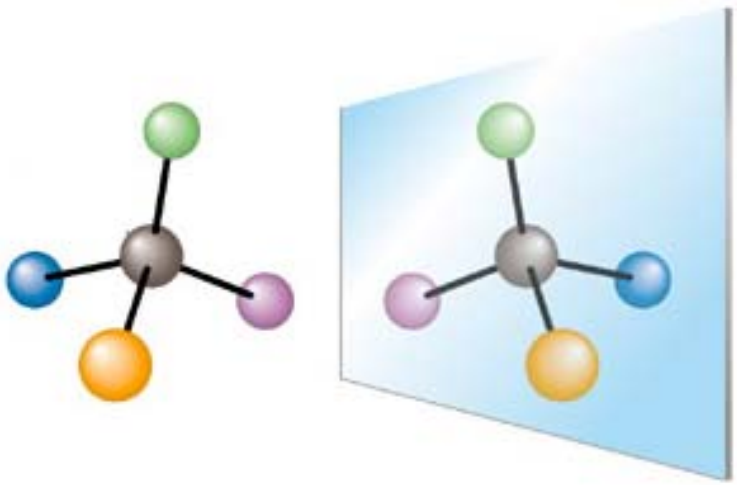
1953: Synthesis at Ciba
1954: Synthesis at Gruenethal
1956: Launch as anticonvulsant, hypnotic, sedative
1961: Report on tragic teratogenicity (W. Lenz)
Withdraw from market (West Germany)
1962: Withdraw from market (Japan)
1965: Therapeutic utility against erythema nodosum leprosum
1977: Therapeutic utility against photodermatitis
1984: Therapeutic utility against phenumatoido arthritis
1985: Therapeutic utility against GVHD
1986: Therapeutic utility against Behcets syndrome
1990: Nominate as an orphan drug in USA
1991: Inhibitory activity of TNF-α production
1993: Therapeutic utility against AIDS
1998: Launch by Celgene as Thalomid



Why the disaster was happened ?

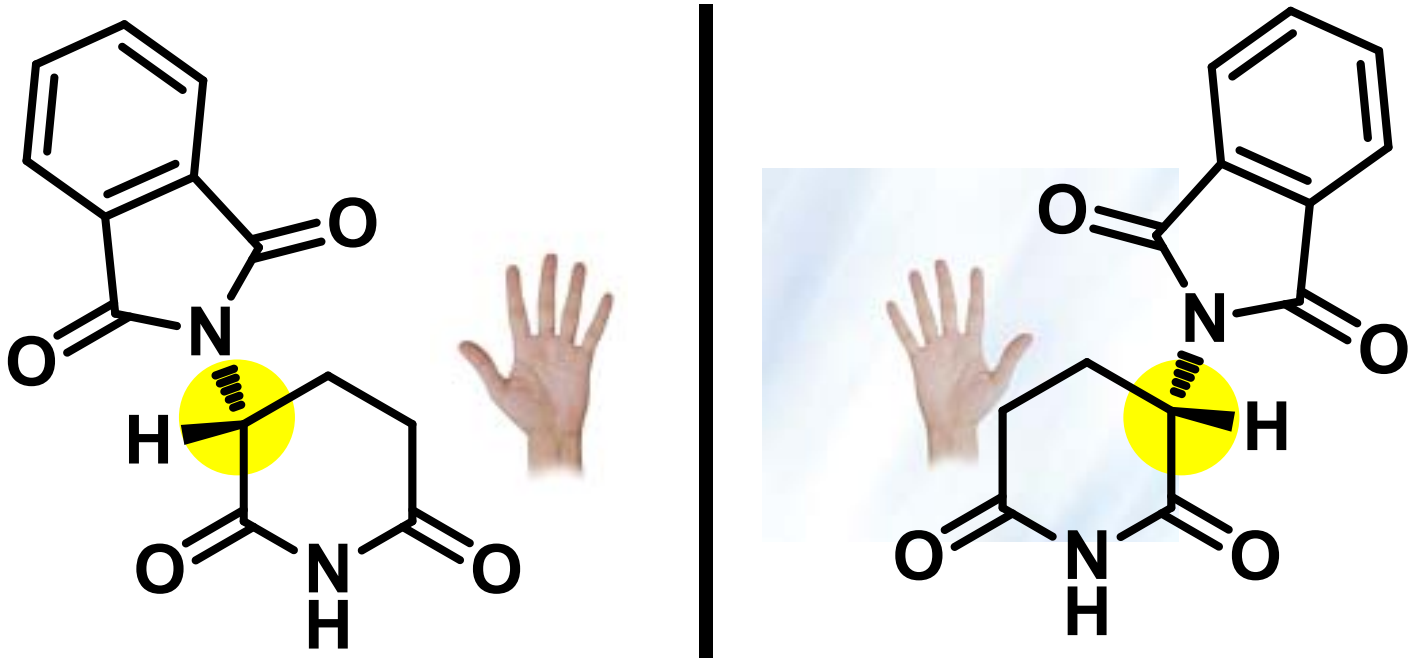
No! Different

Same?



Chiral Molecule

Thalidomide is a Chiral Molecule

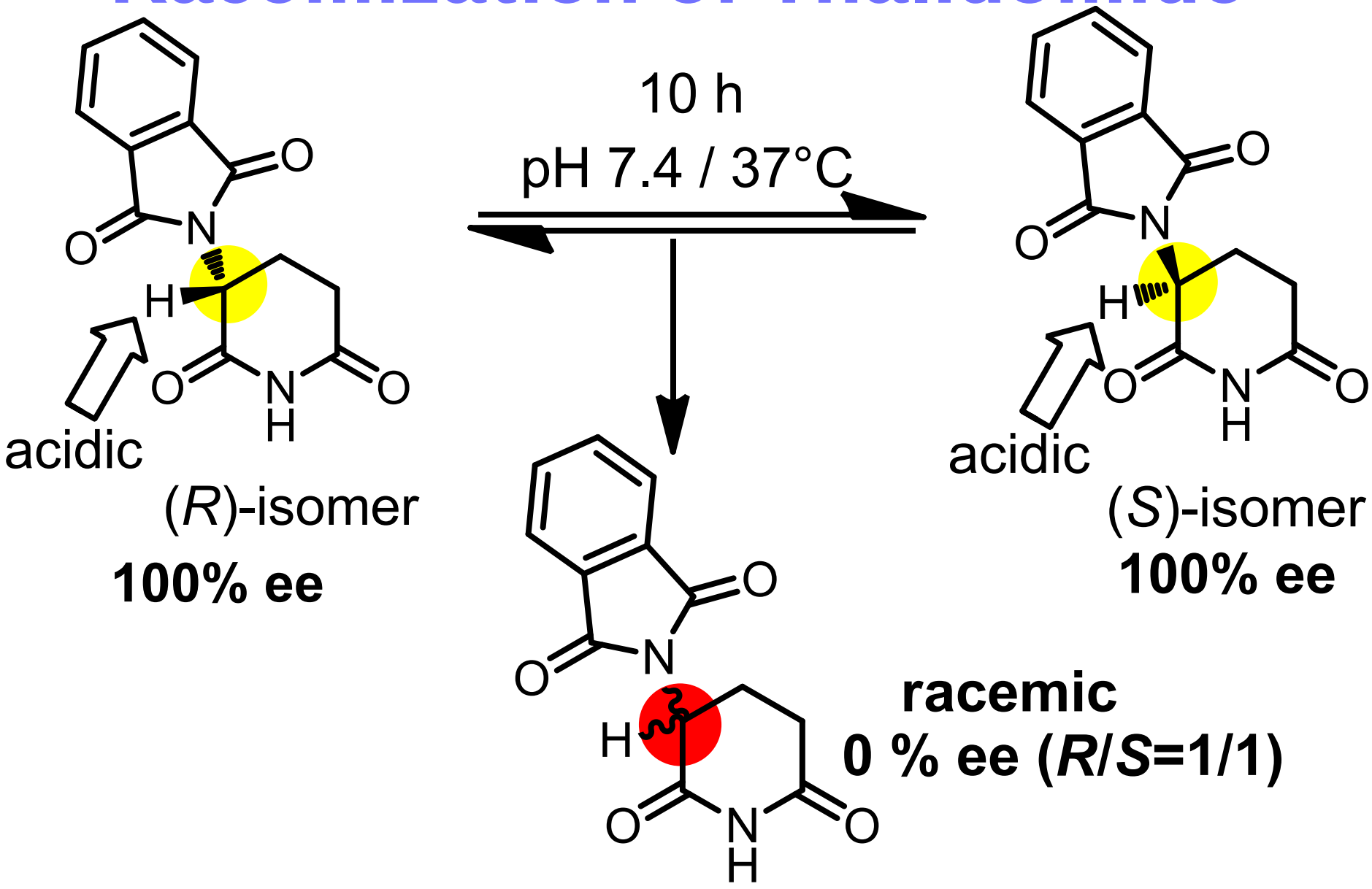


(*R*)-isomer desired effects (sedation)

(*S*)-isomer side effects (teratogenicity)

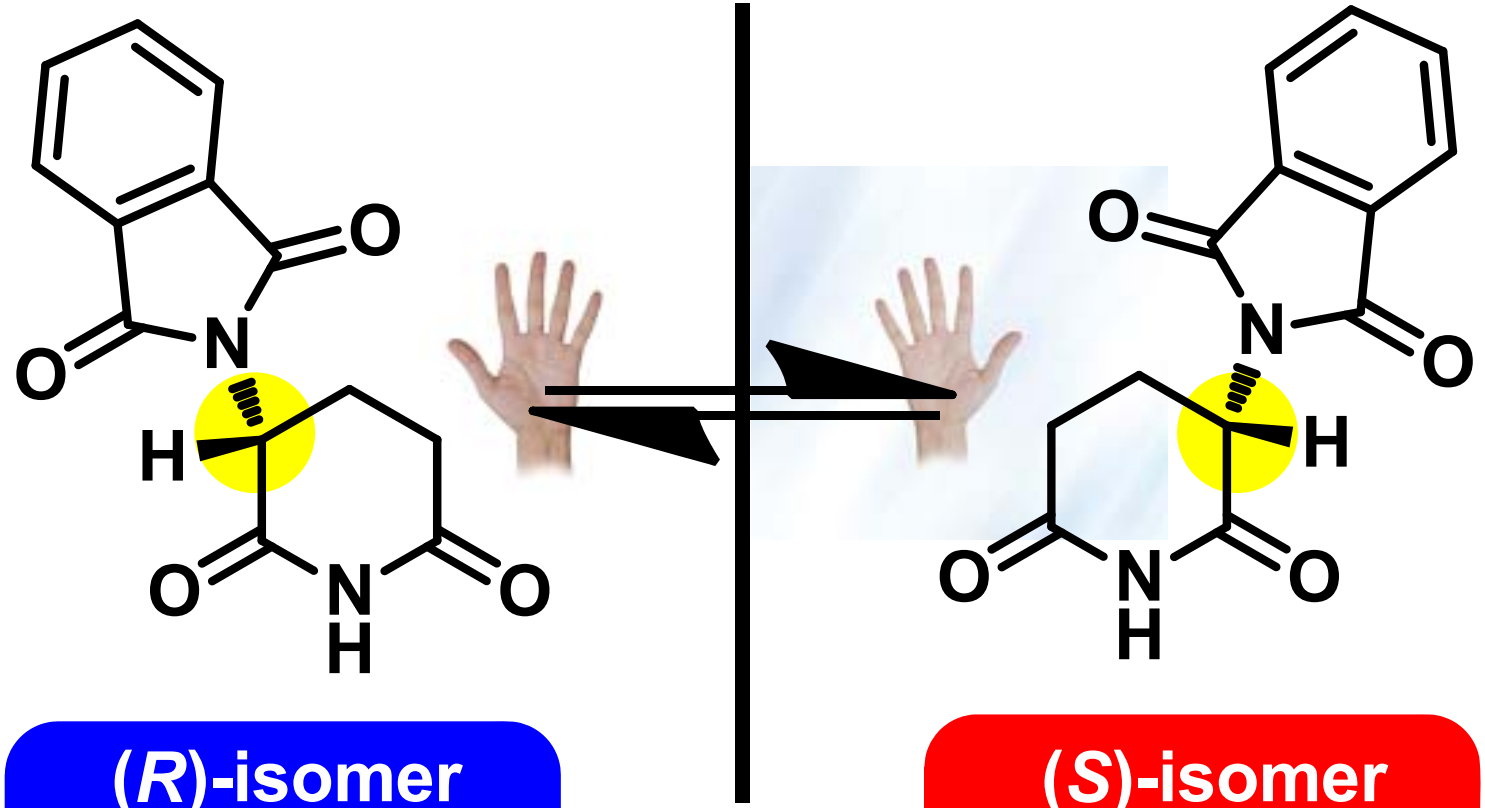
Blaschke, V. G. *et al. Arzneim.-Forsch*. 1979, *29*, 1640.

Racemization of Thalidomide



Nishimura, K. *et al. Chem. Pharm. Bull*. **1994**, *42*, 1157.

Thalidomide is a Chiral Molecule, But...



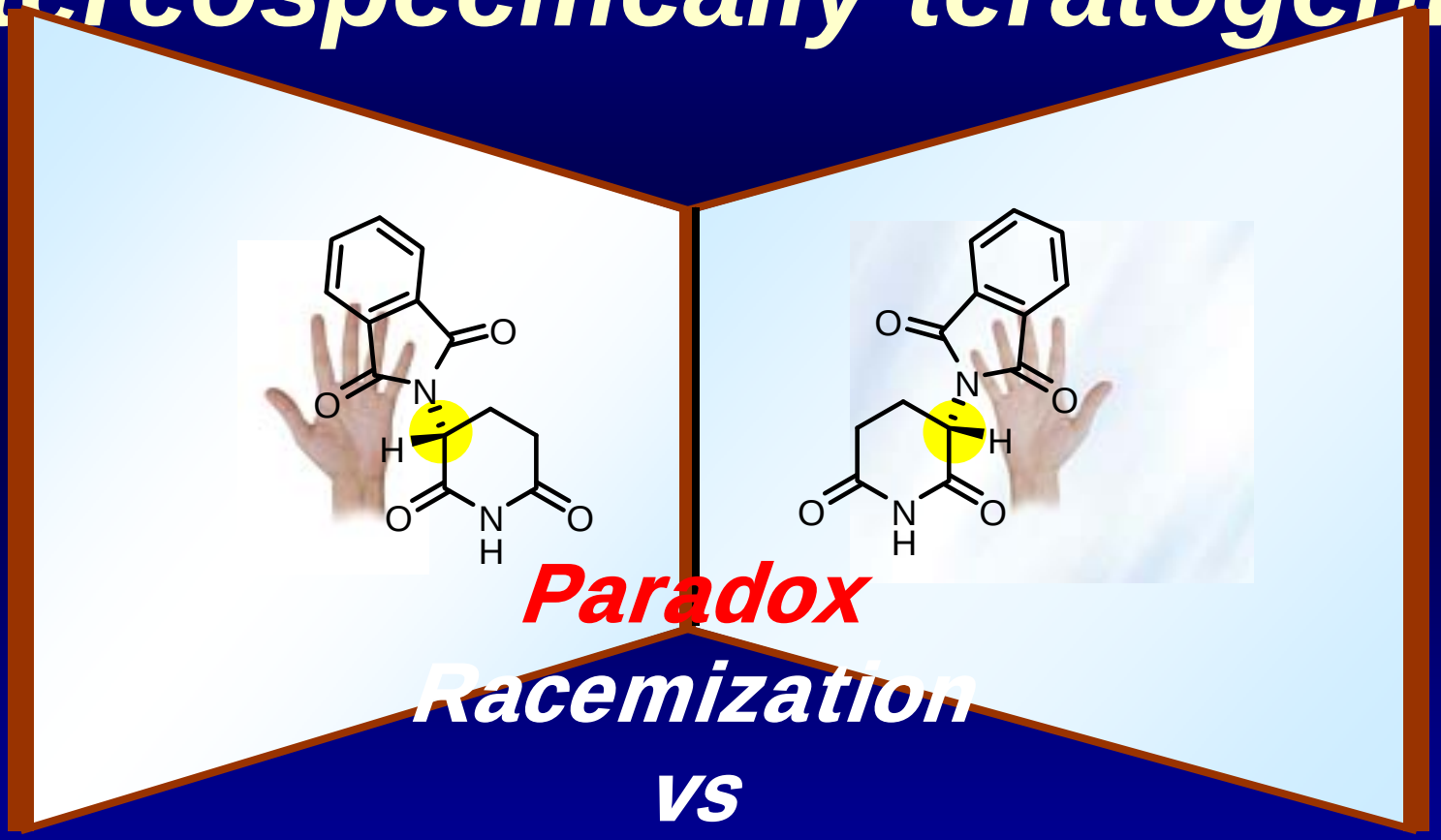
(*R*)-isomer desired effects (sedation)

?

(*S*)-isomer side effects (teratogenicity)

Blaschke, V. G. *et al. Arzneim.-Forsch*. 1979, *29*, 1640.

A fundamental question is whether thalidomide is stereospecifically teratogenic.



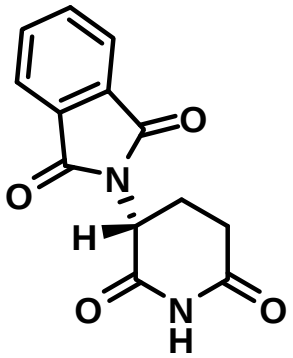
Paradox

Racemization

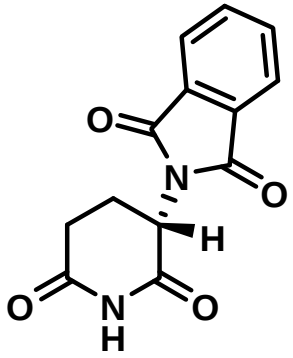
vs

Stereospecific Teratogenicity

Teratogenic Potency of Thalidomide Enantiomers



(*R*)-1

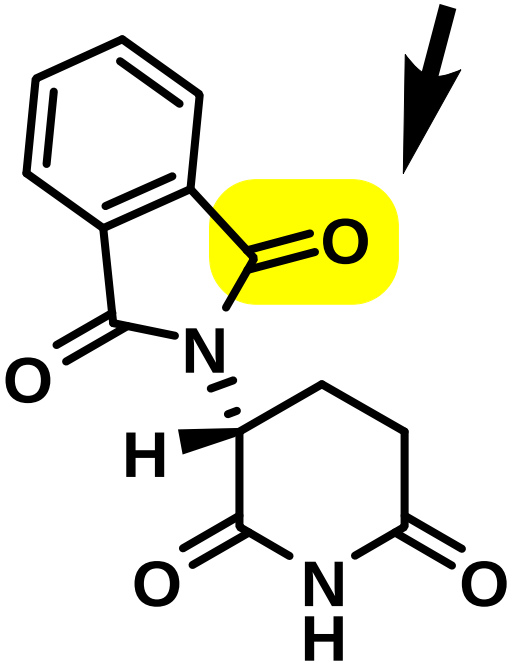


(*S*)-1

V. G. Blaschke. *et al. Arzneim.-Forsch*. 1979, *29*, 1640-1642.

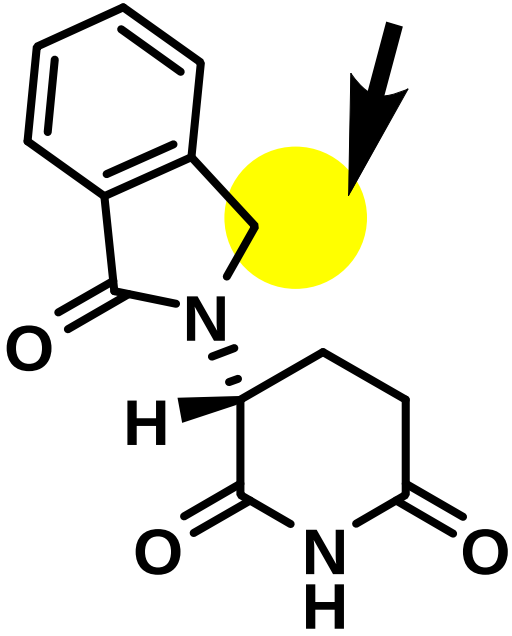
Substance	Dose (mg/kg)	Pregnant mouses	Viable fetuses A	Aborted B	Aborted rate(B/A) (%)	A-B	Abnormal fetuses C	Abnormal rate (%) C/A-B
(*S*)-1	3.12	15	211	15	7.1	196	2	1.0
	12.5	10	135	31	22.9	104	0	0
	50	17	226	59	26.1	167	34	20.3
	100	14	178	21	11.7	157	15	10.4
	200	12	154	29	18.8	125	36	28.8
(*R*)-1	3.12	20	269	28	10.4	241	0	0
	12.5	11	157	17	10.8	140	0	0
	50	15	191	10	5.2	173	0	0
	200	12	160	21	13.1	139	0	0
	400	9	105	12	11.4	93	0	0

EM12 (2-(2,6-dioxopiperidine-3-yl)-phthalimidine: Thalidomide Analogue



(*R*)-Thalidomide

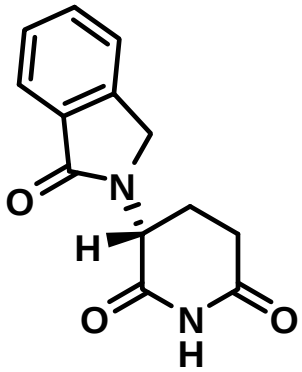
1979
Prof. Gottfried Blaschke
Pharmaceutical Institute.
University of Münster



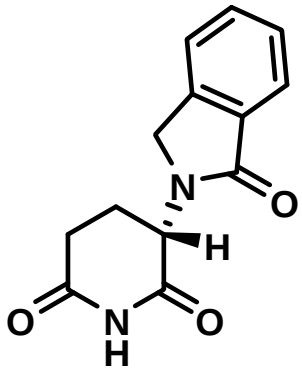
(*R*)-EM-12

1988
Prof. Heinz Nau,
University of Veterinary
Medicine Hannover

Teratogenic Potency of EM12 Enantiomers



(*R*)-EM-12

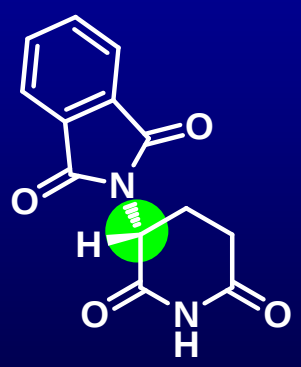


(*S*)-EM-12

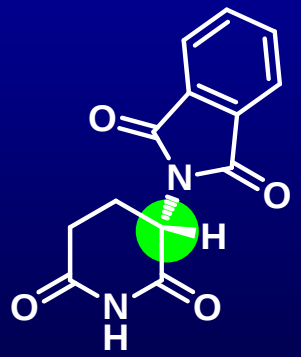
H. Nau. *et al*. *Arch. Toxicol*. 1988, *62*, 205-208.

Substance	Dose mg/kg	Number of		Normal		Litters		Fetuses abnormal
		Pregnant animals	Viable fetuses	Litters	Fetuses	Abnormal	Aborted	
S–(–)-	0.5	3	10	0	0	3	0	10
	1	6	8	0	0	3	3	8
R–(+)-	1	5	11	1	6	3	1	5

Paradox:
Racemization vs Stereospecific Teratogenicity



(*R*)-isomer
desired effects
(sedation)

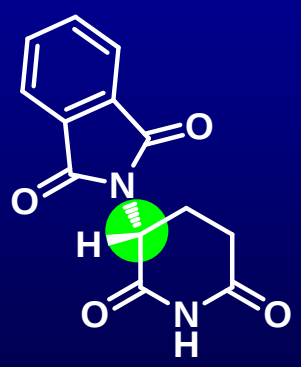


(*S*)-isomer
side effects
(teratogenicity)

Blaschke, V. G. *et al. Arzneim.-Forsch*. 1979, *29*, 1640.

1. Clearness of the biological results using mice by Blaschke.
2. Teratogenicity of thalidomide does not generally appear on rodent such as rat and mouse.
3. Rapid racemization in physiological conditions.
4. Teratogenic study for rabbit did not show the clear difference between those two enantiomers.
5. EM-12, thalidomide analogue, showed the (*S*)-enantiomer of EM-12 had stronger teratogenicity than that of (*R*)-enantiomer.
6. Enantioselective biological studies including metabolism were fragmentally reported by several researchers.

Paradox:
Racemization vs Stereospecific Teratogenicity



(*R*)-isomer
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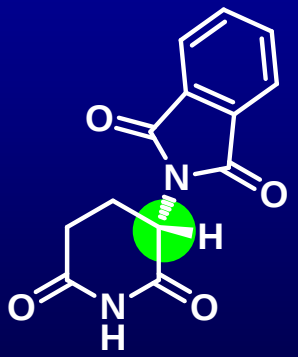
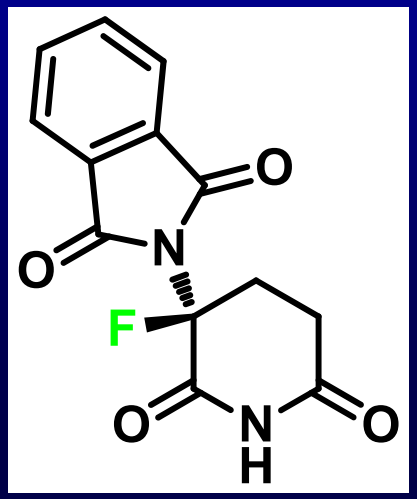


(*S*)-isomer
side effects
(teratogenicity)

Blaschke, V. G. *et al. Arzneim.-Forsch*. 1979, *29*, 1640.



E. Tokunaga

Missing Ring

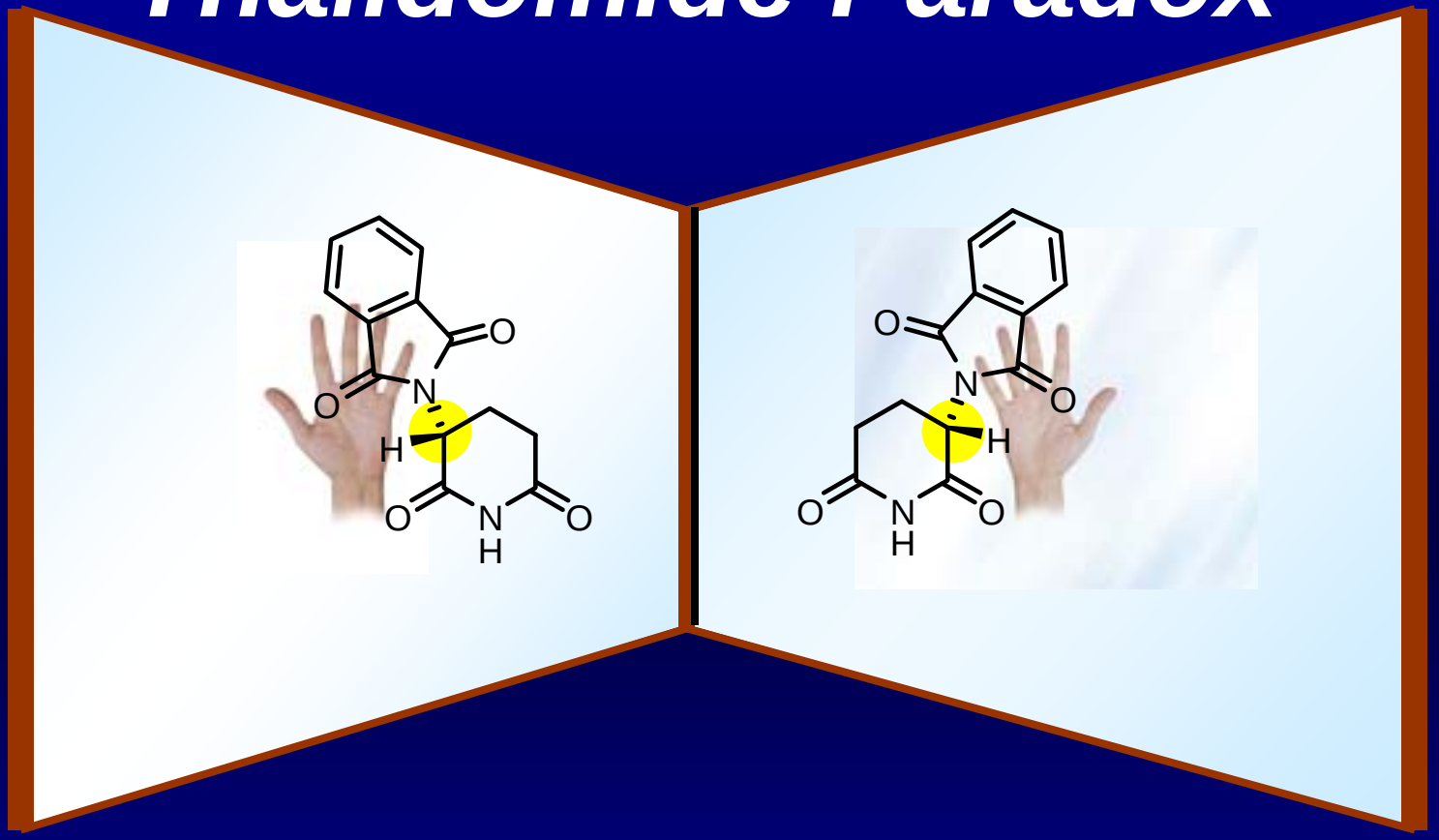
>30 years

Racemization

vs

Stereospecific Teratogenicity

“Thalidomide Paradox”

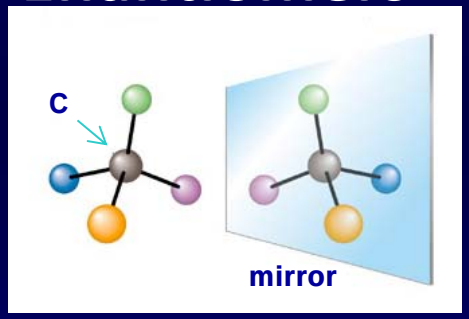


“a self-disproportionation of non-racemic thalidomide in water”

Self-disproportionation of Enantiomers

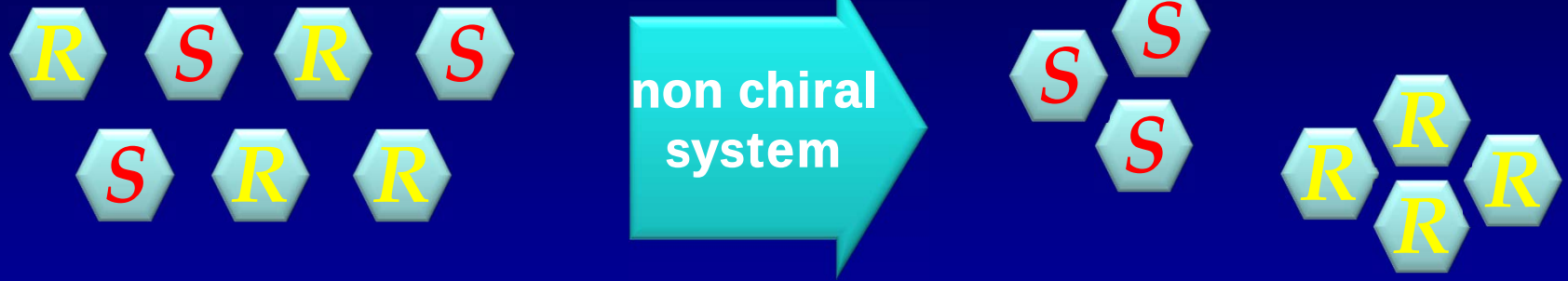
The phenomenon of the optical separation without the application of any external element of chirality.

Enantiomers

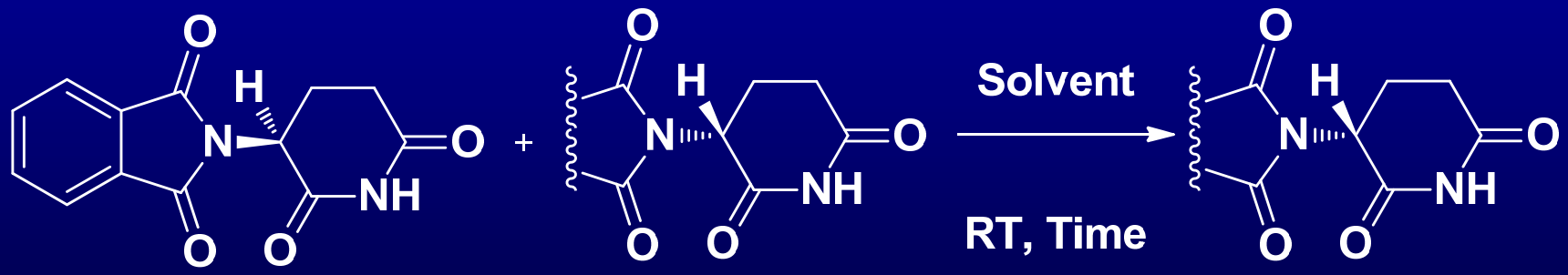


Same Physiological Properties

Chiral materials should be required for separation.



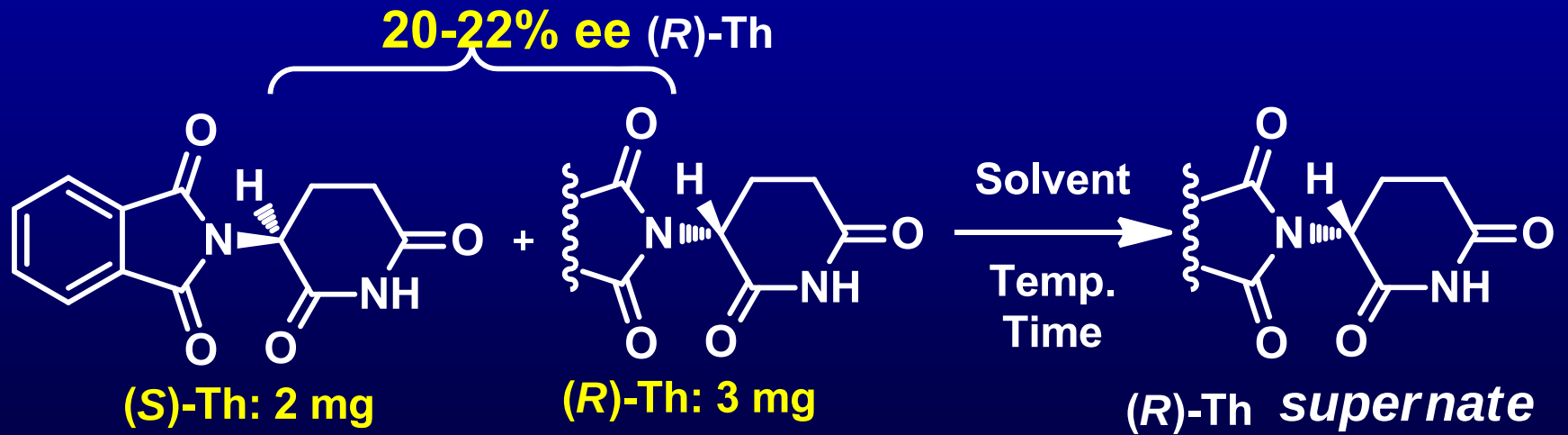
Thalidomide and Chirality: A Fundamental Question is Whether Thalidomide is Stereospecifically Teratogenic.



(*S*)-Th: 2 mg + (*R*)-Th: 3 mg → (*R*)-Th *supernate*

Entry	Solvent 1	Solvent 2	Time (h)	Ee (%)
1	EtOH (8 ml)		-	22
2	EtOH (2 ml)	water (1 ml)	1	
3	$CHCl_3$/EtOH (1:1, 4 ml)	water (1 ml)	1	
4	DMSO (0.2 ml)	water (1 ml)	1	
5	water (8 ml)		1	
6	water (1 ml)		1	
7	water (1 ml)		1	

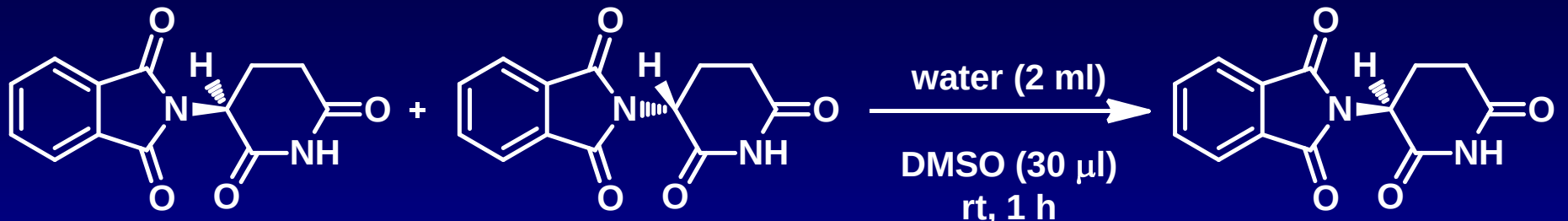
Thalidomide and Chirality: A Fundamental Question is Whether Thalidomide is Stereospecifically Teratogenic.



Entry	Solvent	Temp (°C)	Time (h)	Ee (%)
1	water (1 ml)	37	1	
2	water (1 ml)	37	24	
3	phosphate buffer (pH 7) (1 ml)	rt	1	
4	phosphate buffer (pH 7) (1 ml)	rt	24	
5	phosphate buffer (pH 7) (1 ml)	37	1	
6	phosphate buffer (pH 7) (1 ml)	37	24	

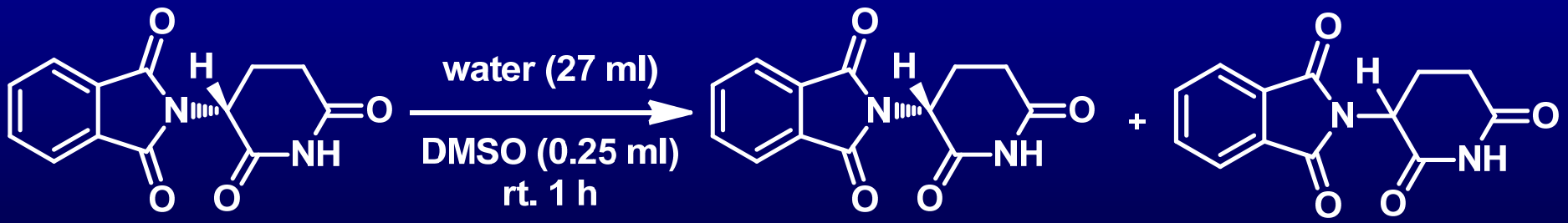
Thalidomide and Chirality: A Fundamental Question is Whether Thalidomide is Stereospecifically Teratogenic.

(*S*)-Th: 2 mg + (*R*)-Th: 3 mg → (R)-Th: 82% ee *supernatant*



(*S*)-Th: 3 mg + (*R*)-Th: 2 mg → (S)-Th: 90% ee *supernatant*

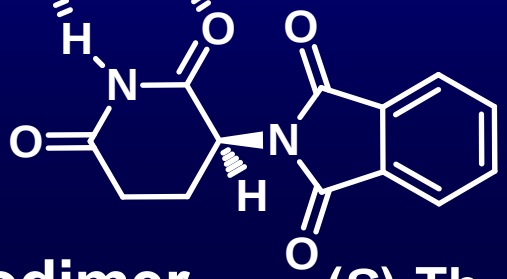
Thalidomide and Chirality: A Fundamental Question is Whether Thalidomide is Stereospecifically Teratogenic.



Run	(*R*)-Th (Ee)	(*R*)-Th (filtrate)			Th (precipitate)	
		Amount (mg)	Yield (%)	Ee (%)	Amount (mg)	Ee (%)
1	20%	3.9	39	82	40.5	13 (R)
2	20%	3.8	38	84	43.0	15 (R)
3	21%	3.7	37	78	38.4	19 (R)

Racemic Quenching of Thalidomide

(*S*)-Th



homodimer

(*S*)-Th

Solubility

369.4 μg/ml in water

monomer

(*S*)-Th

Soluble

5.7 fold

monomer

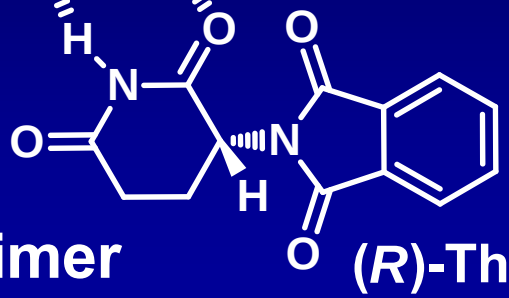
(*S*)-Th (*R*)-Th

Solubility

64.8 μg/ml in water

(*S*)-Th

Poorly Soluble

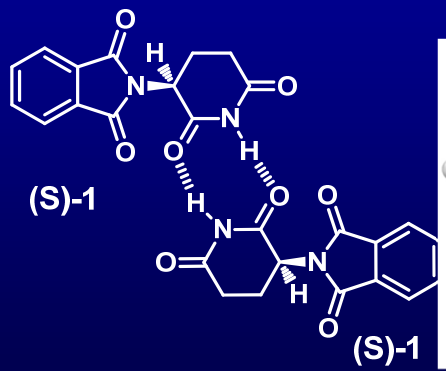


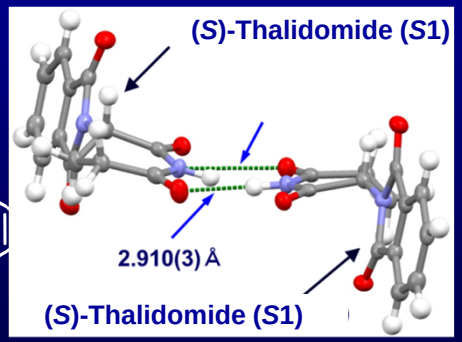
heterodimer

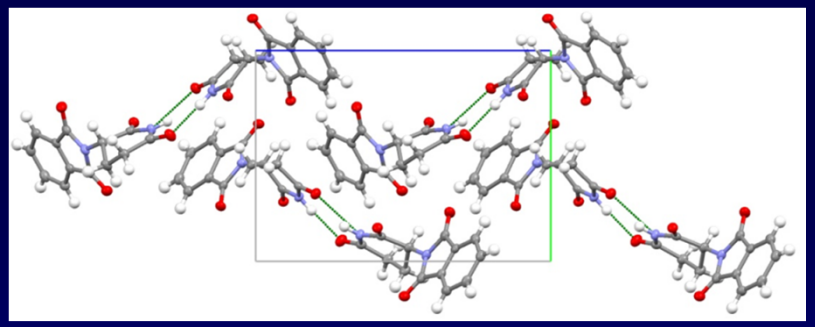
(*R*)-Th

Proposed Mechanism for Thalidomide

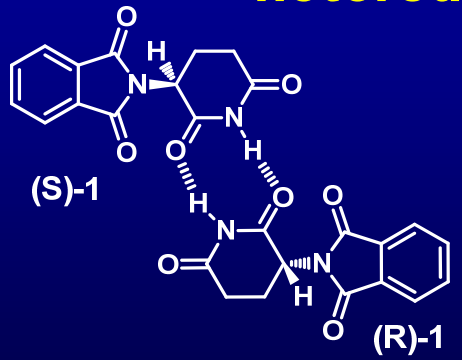
homodimer

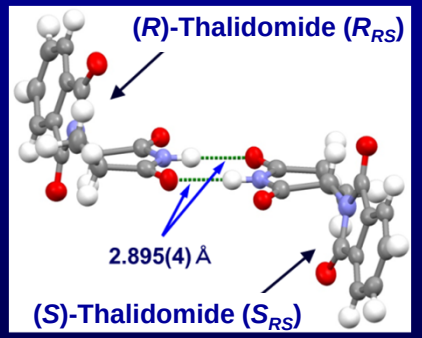


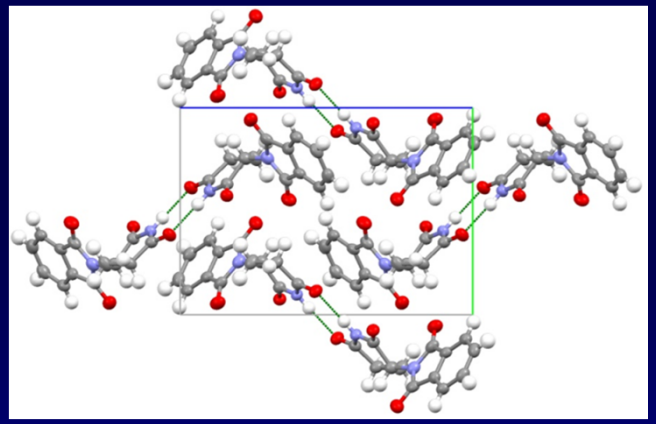




heterodimer

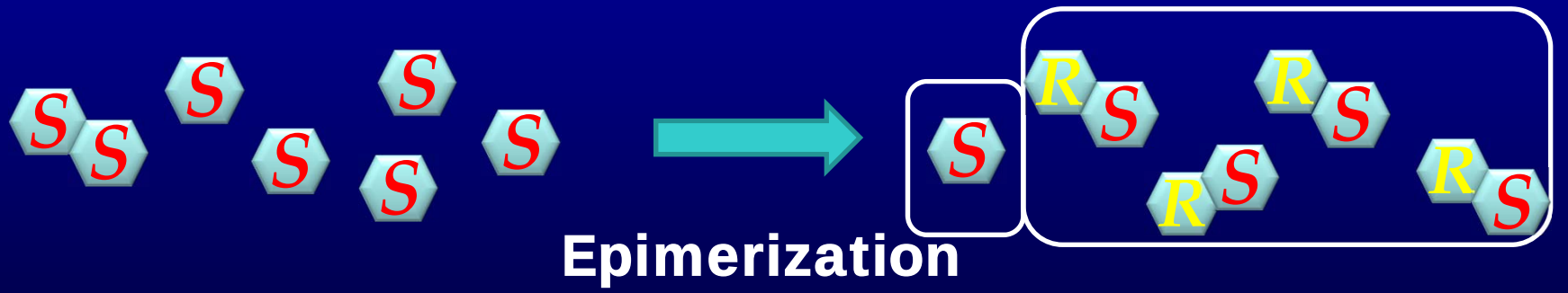






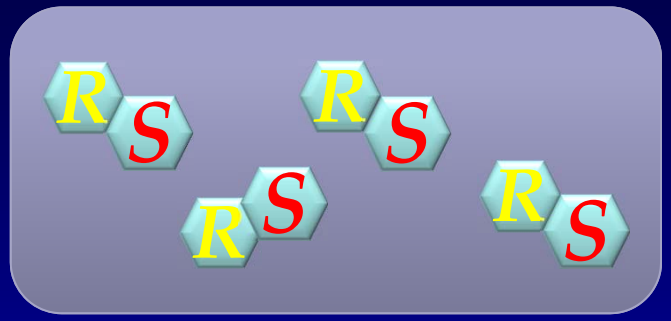
Suzuki, T.; Tanaka, M.; Shiro, M.; Shibata, N.; Osaka, T.; Asahi, T. *Phase Transitions*. **2010**, *83*, 223-234.

Our Novel Hypothesis for Mechanism of Enantiospecific Biological Effects



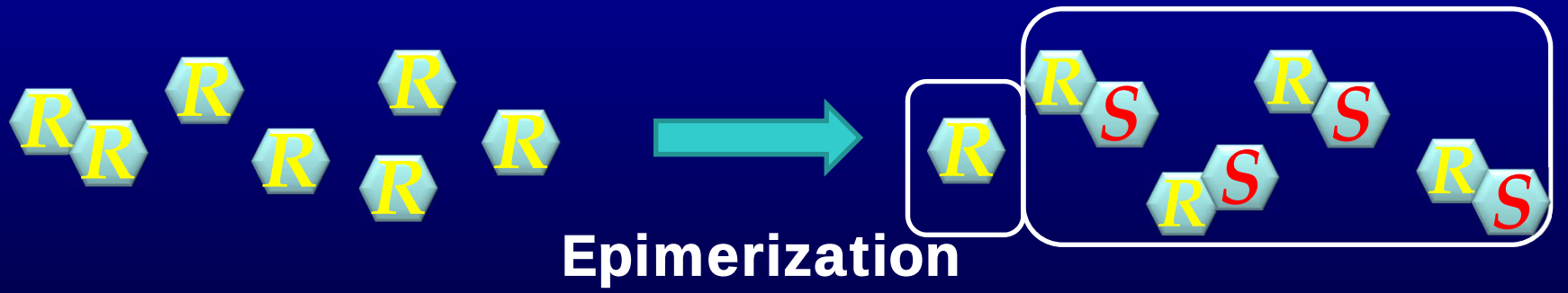
Racemic Quenching

(*S*)-isomer in the Blood



Separated-out from Biological System as precipitate

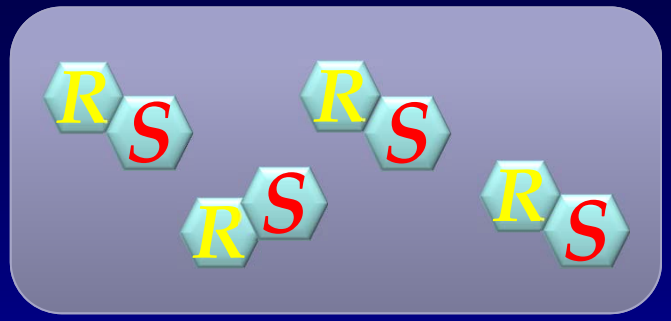
Our Novel Hypothesis for Mechanism of Enantiospecific Biological Effects



Epimerization

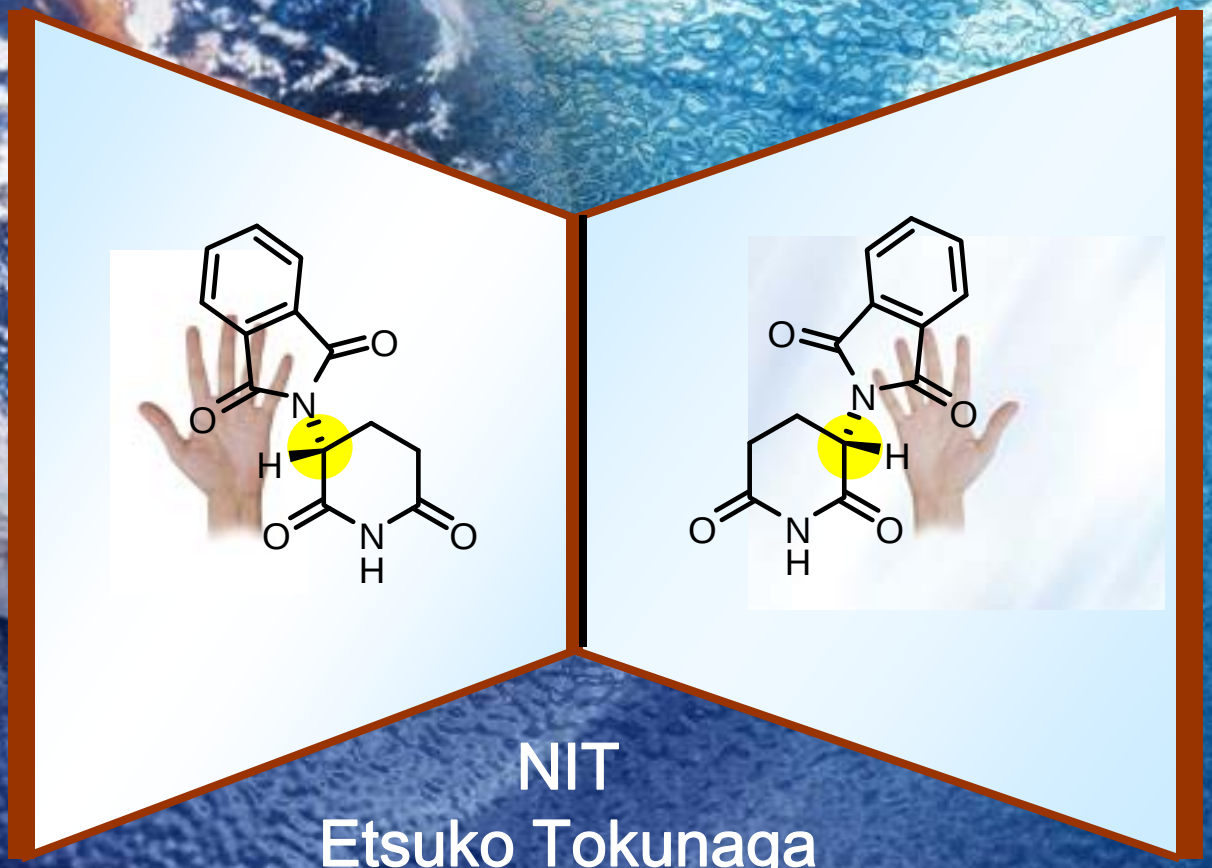
Racemic Quenching

(*R*)-isomer in the Blood



Separated-out from Biological System as precipitate

Acknowledgment



NIT
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