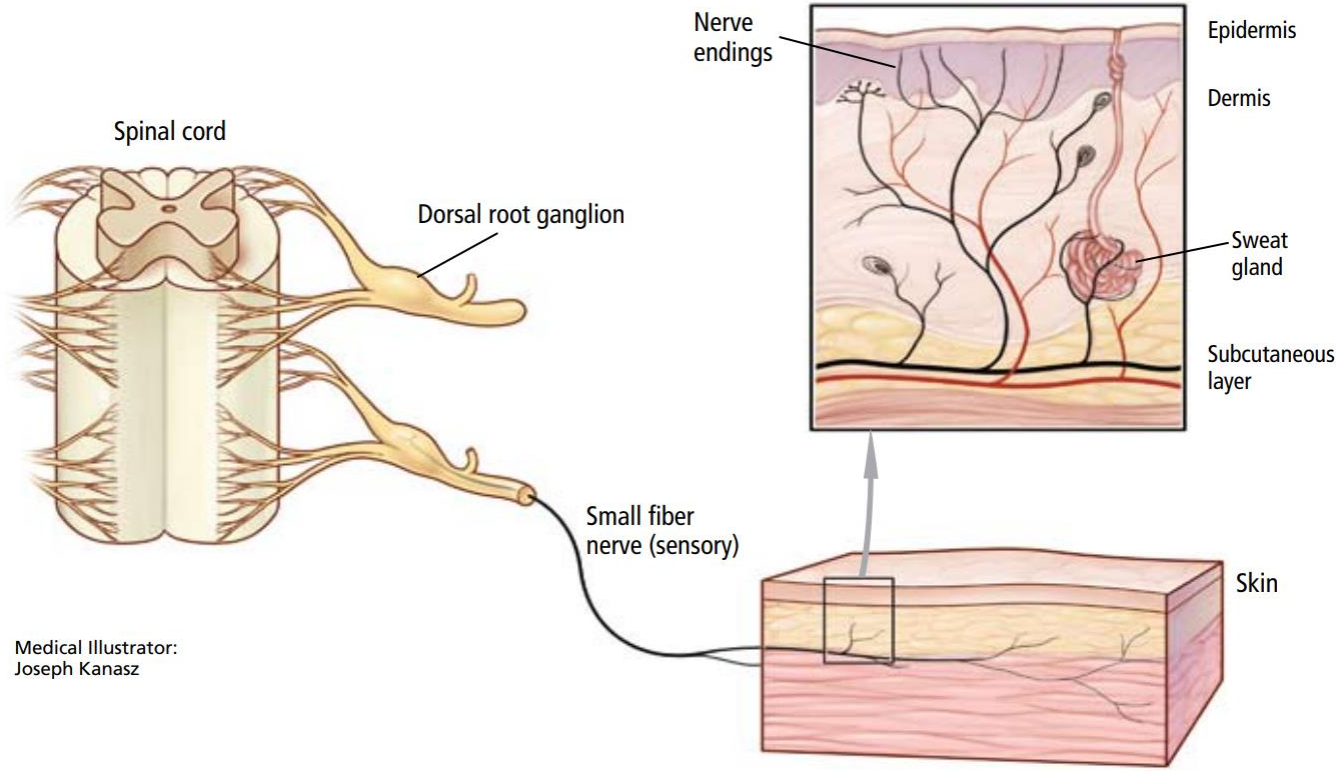
PAIN

Hayden Anderson

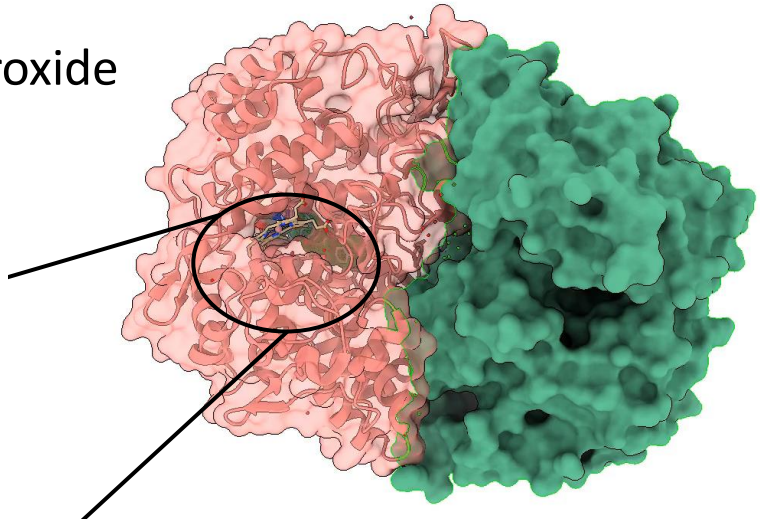
Nociception

Hayden Anderson

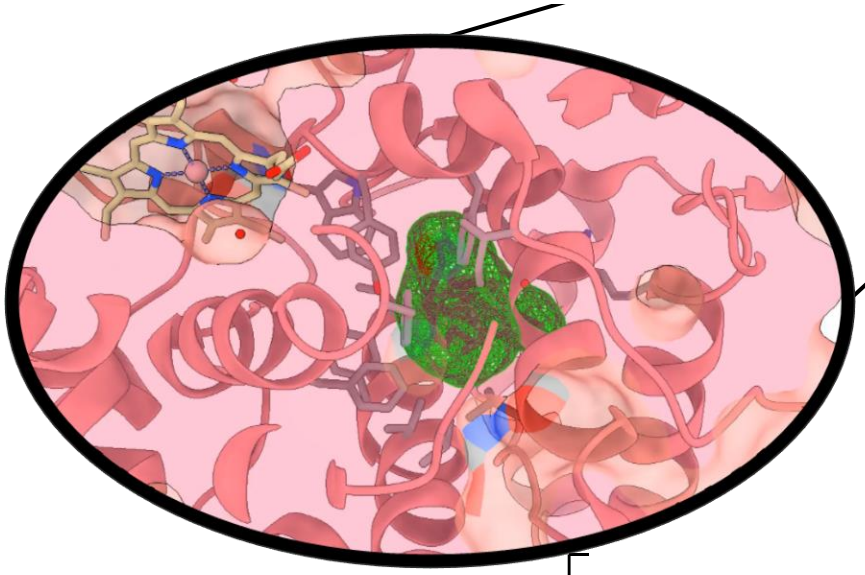

Nerve endings
Epidermis
Dermis
Sweat gland
Subcutaneous layer
Spinal cord
Dorsal root ganglion
Small fiber nerve (sensory)
Skin
Medical Illustrator:
Joseph Kanasz

Cyclooxygenase or Prostaglandin-Endoperoxide Synthase (PTGS):

- Exists as a C_2 symmetric homodimer
- Contains a porphyrin chelated iron atom
- Isoforms are differentially expressed in different tissues



Ovine Cyclooxygenase-1 complexed with endogenous substrate arachadonic acid



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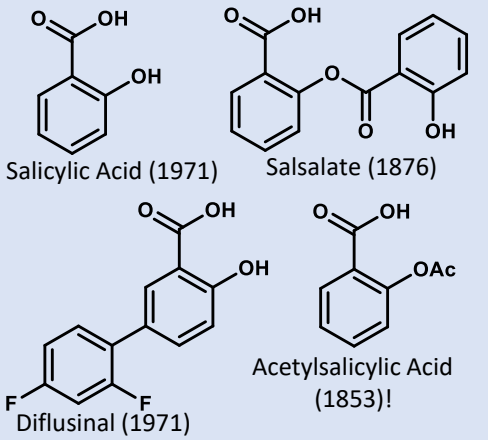
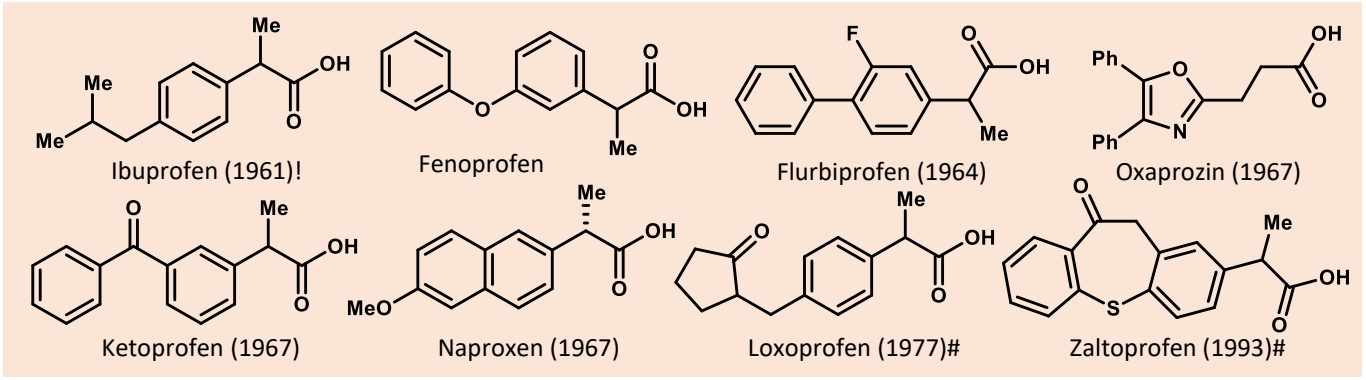
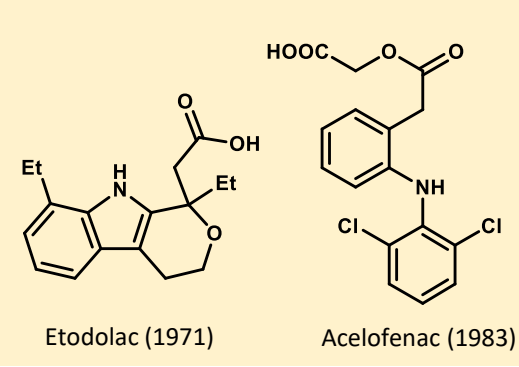
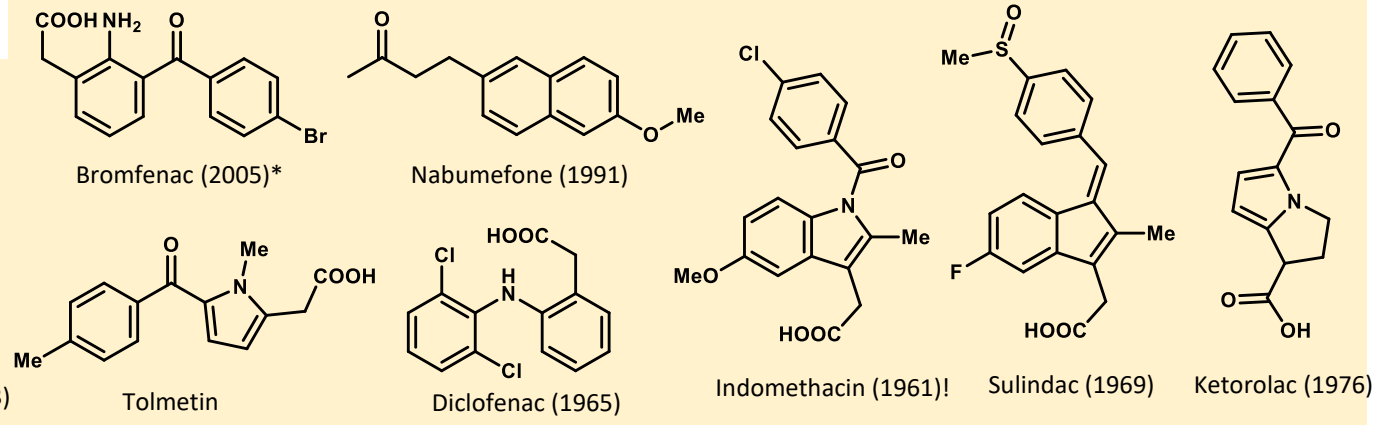
NSAIDs

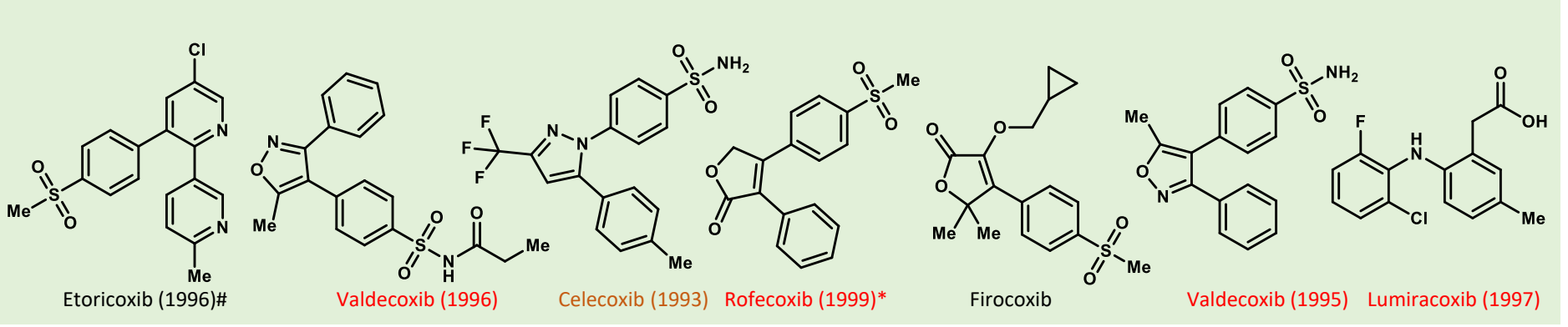
2 August 2022

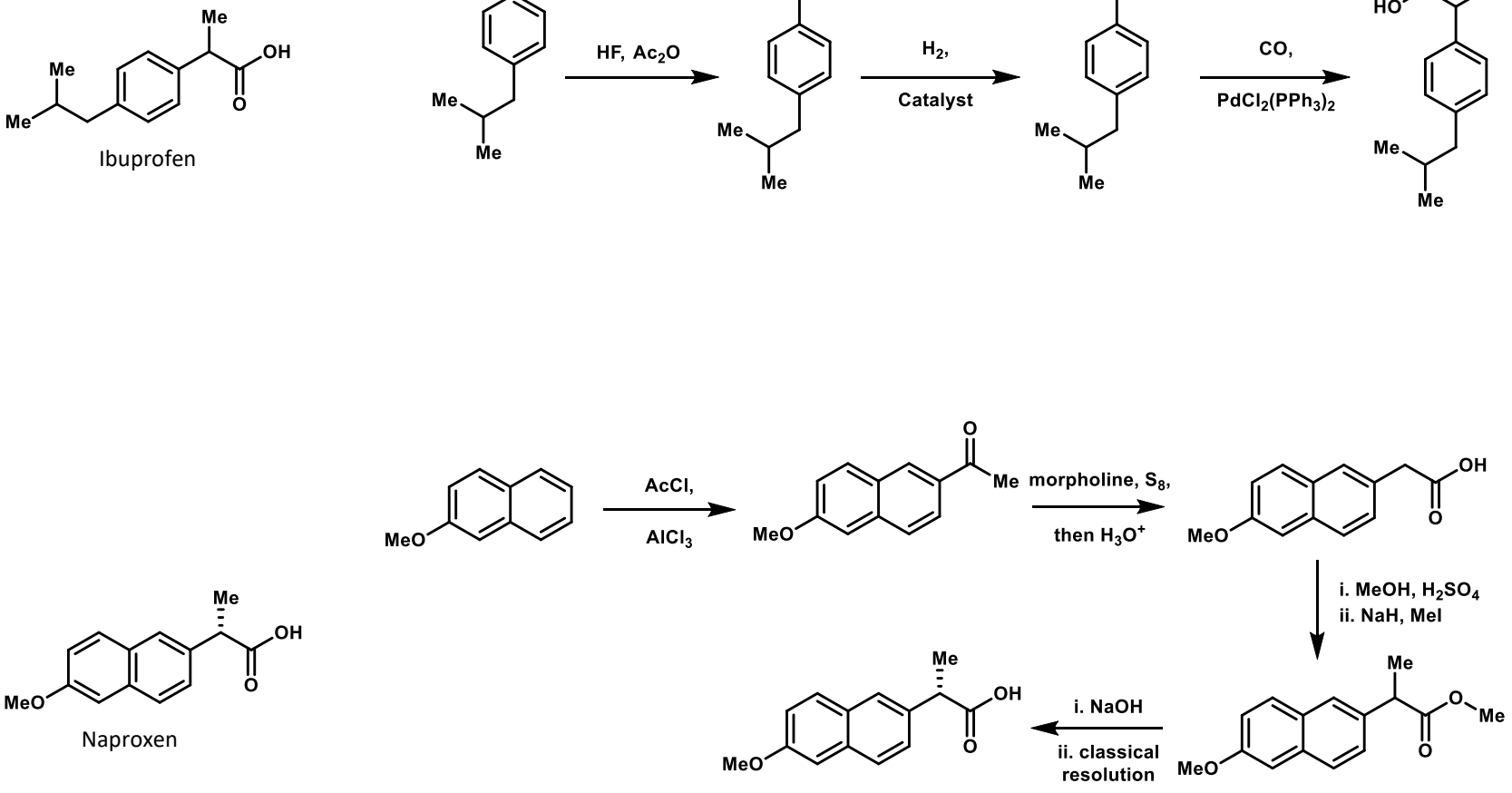










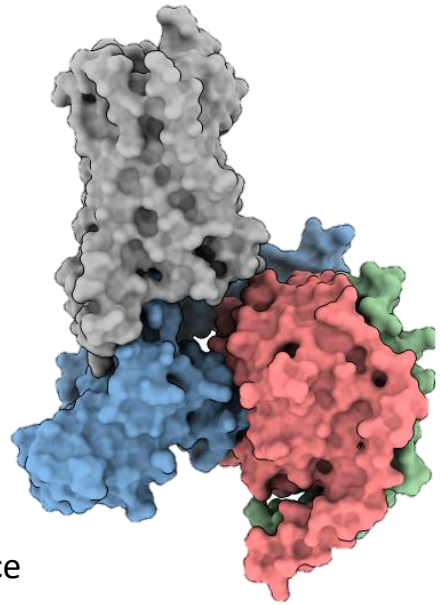

Ibuprofen
HF, Ac2O
H2,
Catalyst
CO,
PdCl2(PPh3)2
Naproxen
AcCl,
AlCl3
morpholine, S8,
then H3O+
i. MeOH, H2SO4
ii. NaH, MeI
i. NaOH
ii. classical
resolution

H. Anderson

Opioid Receptors

2 August 2022

- G-protein coupled receptors (GPCRs) that bind to endogenous ligands known as endorphins (portmanteau of *endogenous morphine*)
- Upon release of the GDP, the $G_{\beta\gamma}$ subunit is released and inhibits calcium channels preventing the release of neurotransmitters into the synaptic cleft
- Further, G_{α} or $G_{\beta\gamma}$ also activate the potassium rectifying ion channel leading to hyperpolarization of the membrane

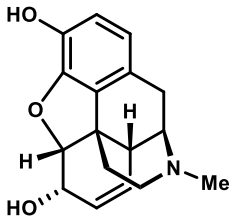


μ-opioid receptors:

- Primary target of morphine
- Agonism has side effects of respiratory depression, reduced GI motility, physical dependence and often itching

δ-opioid receptors:

- Discovered in the vas deferens of mice
- Agonism may actually increase respiratory rate in low doses
- Agonists modulate effectiveness of μ-opioid agonists
- Agonism can result in seizures at high doses

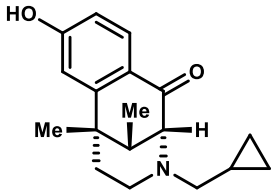


κ-opioid receptors:

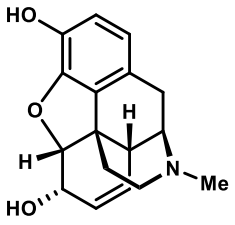
- Identified through interactions with ketocyclazocine
- Believed to have some role in consciousness as agonists result in feelings of paranoia, hallucinations and dissociation

N-opioid receptors:

- Discovered 30 years after the others
- Has received less attention because of its late discovery
- In primates, agonists provided potent analgesia without any of the side effects commonly associated with opioids

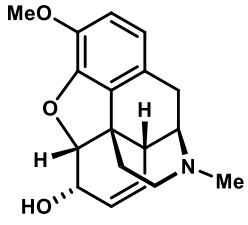


Opiates

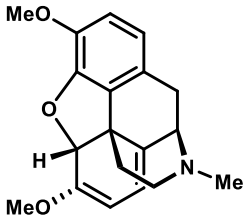


morphine

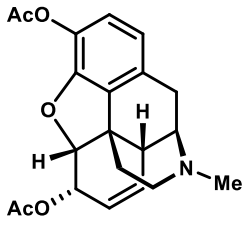
- Originally used as part of opium from *Papaver somniferum*
- The extract contains up to 25% morphine, 6.4% codeine and 6.4% thebaine



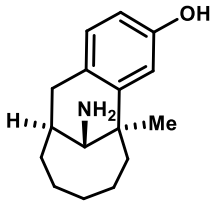
codeine



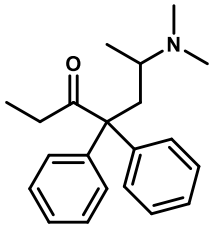
thebaine



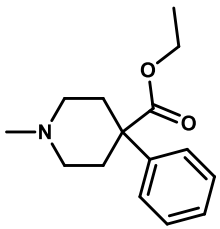
heroin



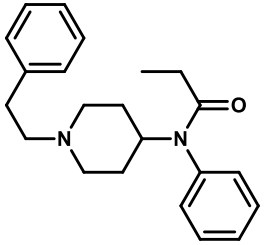
dezocine



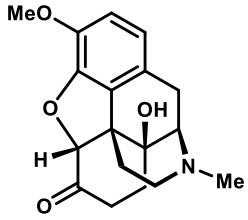
methadone



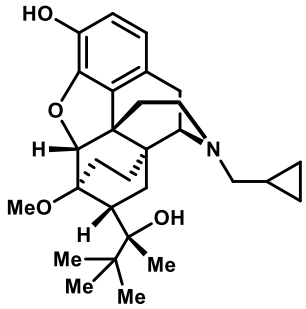
demerol



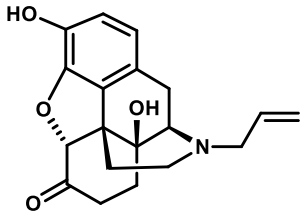
fentanyl



oxycodone

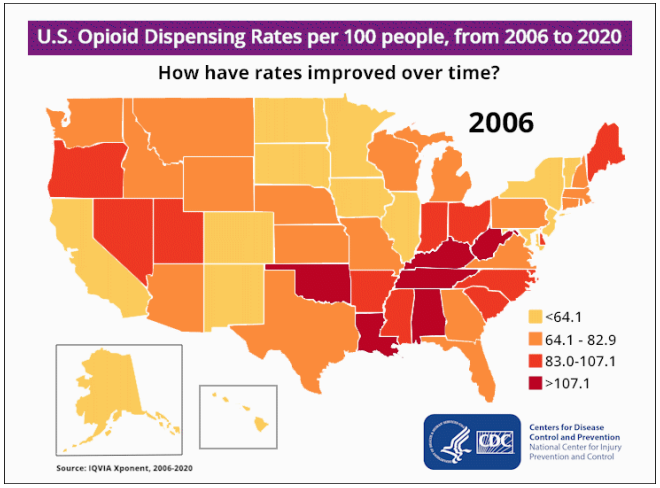


buprenorphine



naloxone

Analgesic	Relative Strength
Ibuprofen	1/222
Codeine	1/10
Morphine	1
Methdone	3
Fentanyl	75
Carfentanil	10,000



- Initially hailed as miracle drugs with minimal chance of abuse until 1920
- Doctors were trained not to prescribe opiates until 1970s
- Opiate prescriptions topped 255 million in 2012 or about 0.81 prescriptions per capita
- In 2022, about 0.43 prescriptions per capita were made

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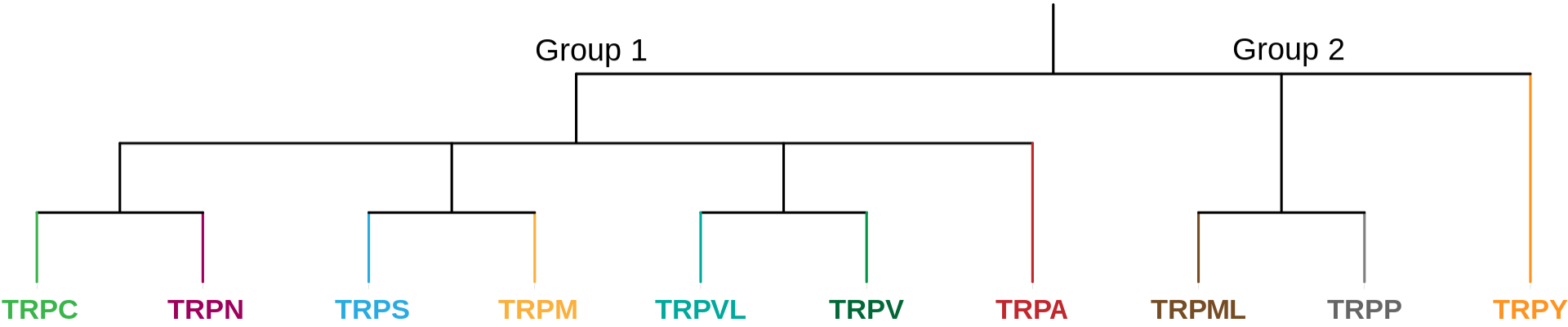
Transient Receptor Potential (TRP) Channels

2 August 2022



- Gated ion channels with varying degrees of selectivity for Ca^{2+}, Na^{+} and K^{+} ions
- Responsible for mediating the transduction of various extracellular stimuli into intracellular responses
- 9 established families which all posses 6 trans-membrane segments
- Little structural or sequence homology between the subfamilies
- Group 1 and group 2 differ in the length of the length of their extracellular segments

TRPC5:
Implicated in mercury poisoning and dental cold sensing

TRPC6:
Implicated in depression and anxiety

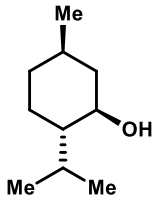
TRPM5:
Plays a key role in the sensation of taste and insulin secretion

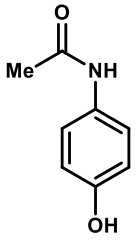
PKD1:
Detects and mediates fluid flow through the kidneys

TRPML1:
Transports iron atoms into the cell from lysosomes

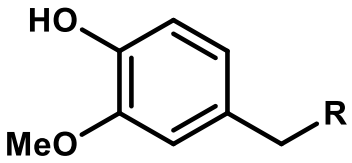
TPRM3 and TRPV2:
Detection of noxious heat > 40 °C and 52 °C, respectively

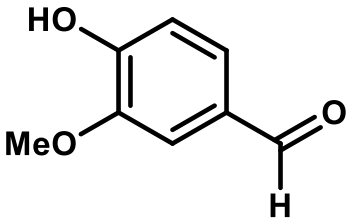
TRPM8:
Responsible for the detection of noxious cold and is activated chemically by menthol

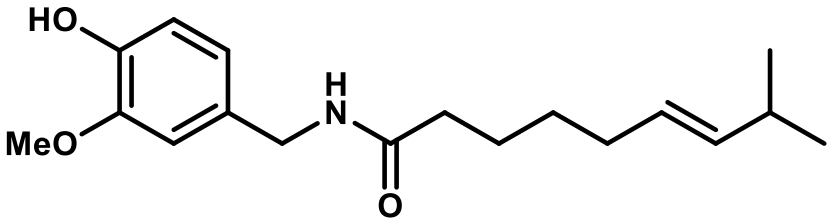




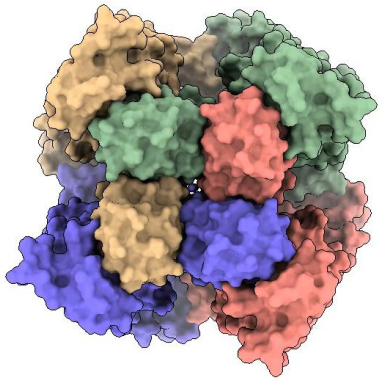
TRPA1:
Responsible for the taste of garlic and transmission of pain signal due to injury or inflamtion

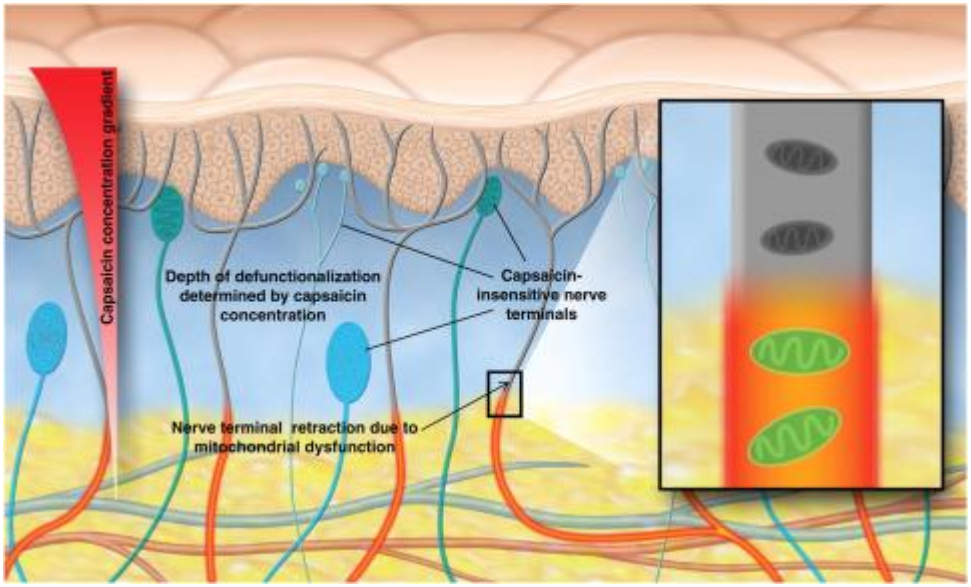


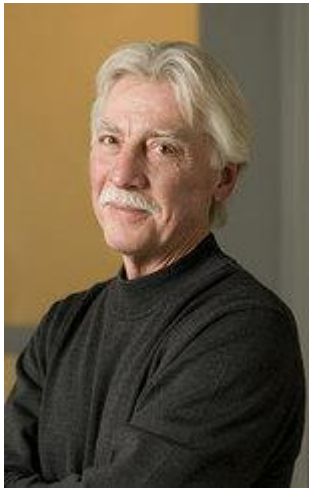




- Non-selective ion channel permeable to Ca^{2+}, K^{+}, and Na^{+}
- Responsible for the detection of noxious heat above
- Also activated by low pH and chemical agents
- Analgesics targeting TRPV1 are typically topical and are aimed at inflammatory and rheumatic pain







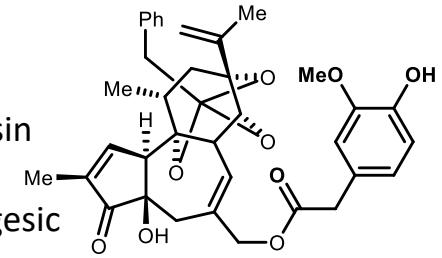
Paul Wender

Cynthia Jesudason

Hiroyuki Nakahira

Norikazu Tamura

Resiniferatoxin:



- Produced by *Euphorbia resinifera* (resin spurge)
- Used in ancient medicine for its analgesic properties
- Has a Scoville rating of 16 billion units
- Has been termed the “molecular scapel”

With Anne Louise Tebbe and Yoshihide Ueno

- Completed the first asymmetric total synthesis of resiniferatoxin
- 44 total steps
- 0.25% yield
- Relied on the Achmatowicz reaction and a zirconocene mediated ring closure

Others:

- 3 other syntheses by Inoue have been published in 2017, 2021, and 2022
- 41, 20 and 27 steps respectively

Masayuki Inoue

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Resiniferatoxin Total Synthesis

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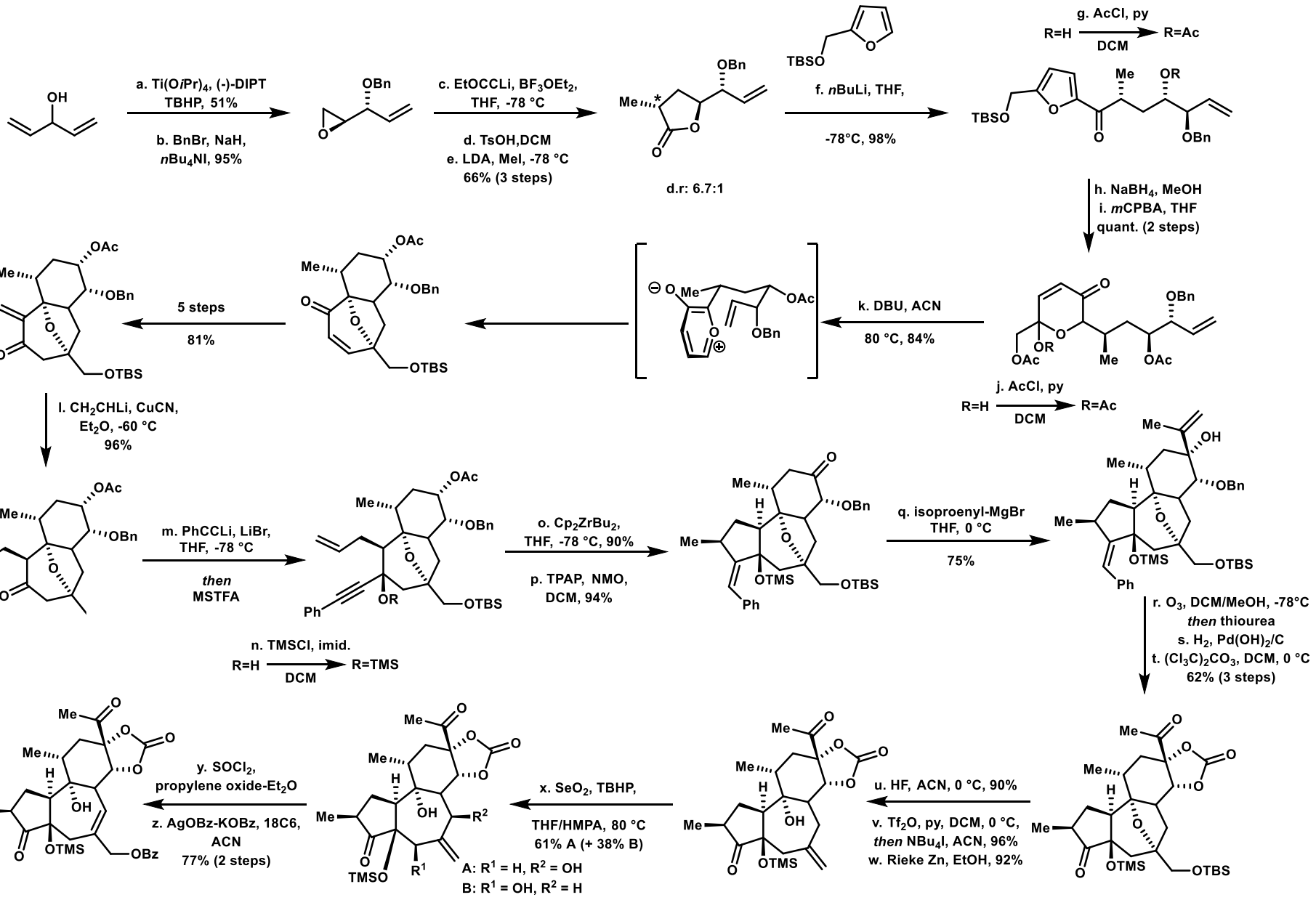
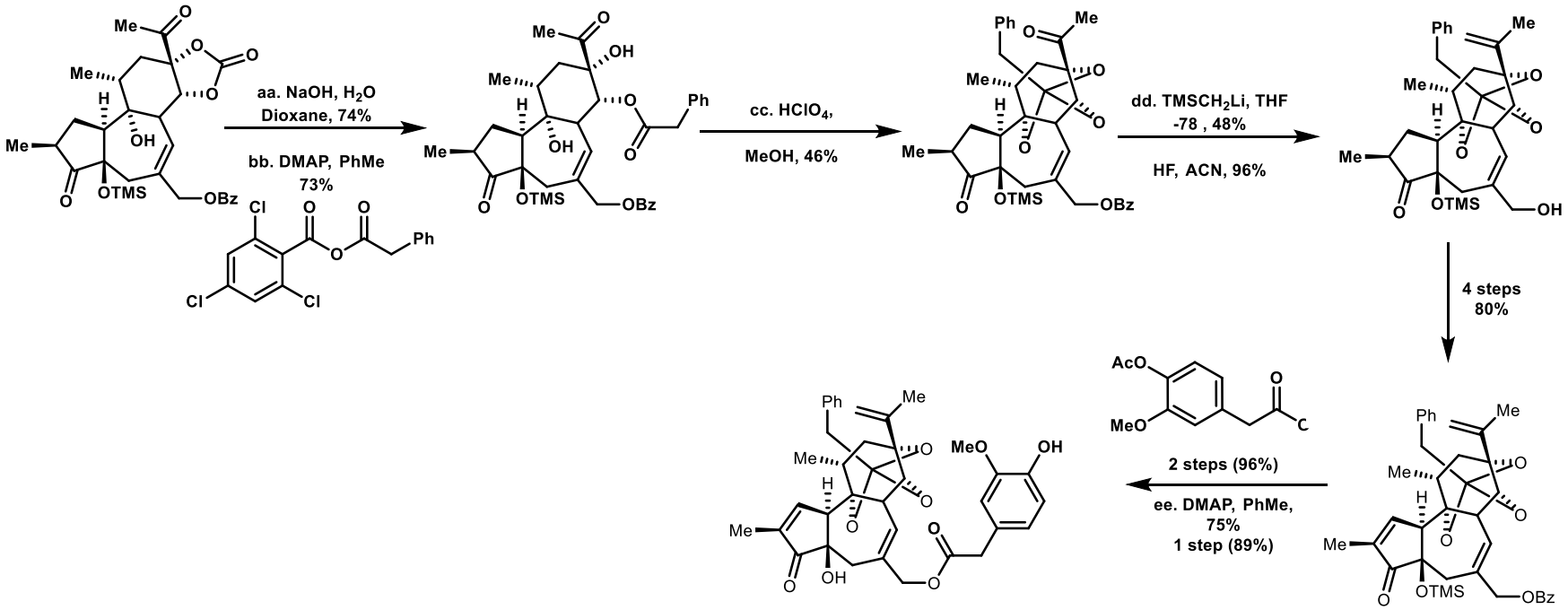
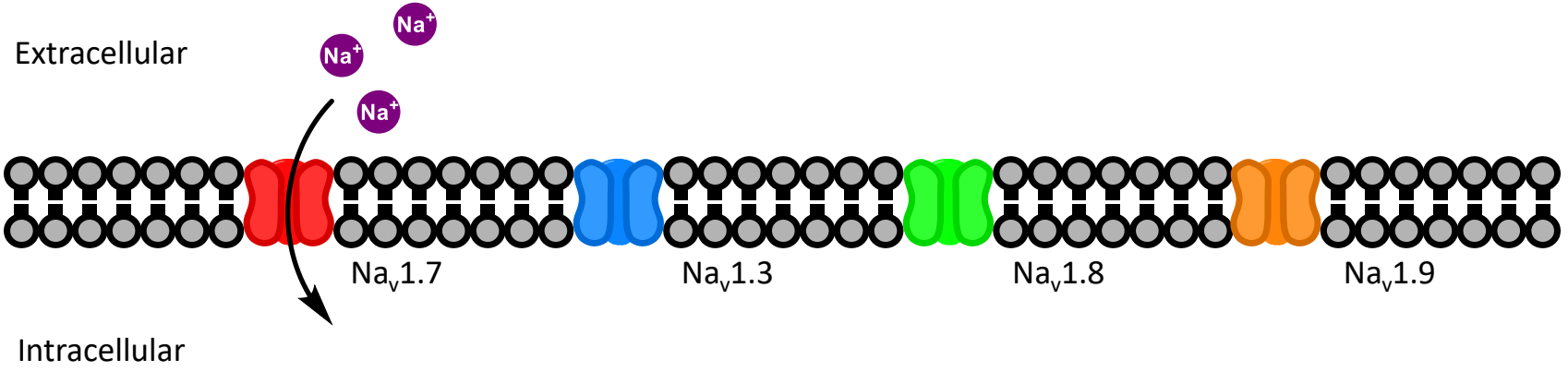


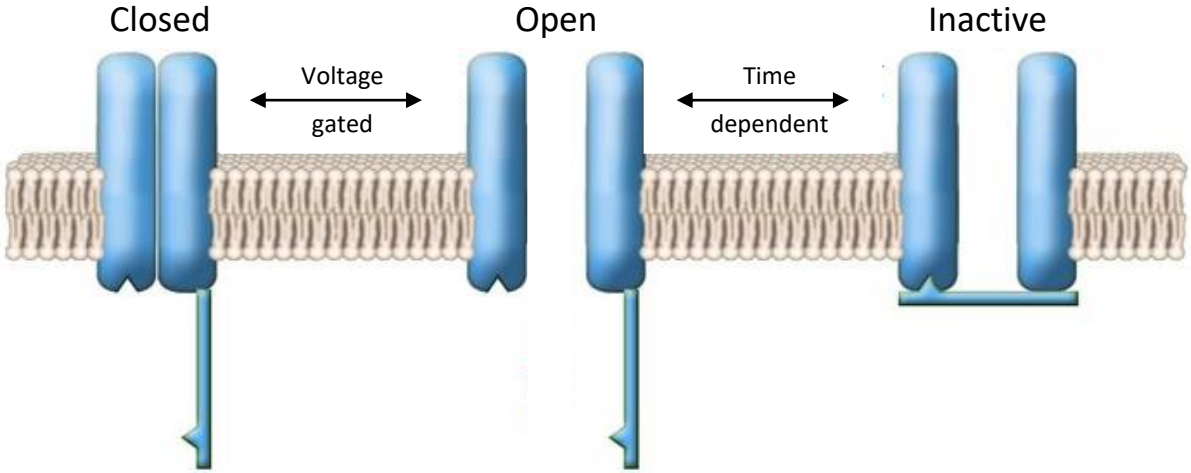

aa. NaOH, H2O
Dioxane, 74%
bb. DMAP, PhMe
73%
cc. HClO4,
MeOH, 46%
dd. TMSCH2Li, THF
-78 , 48%
HF, ACN, 96%
4 steps
80%
ee. DMAP, PhMe,
75%
1 step (89%)
2 steps (96%)

Voltage Gated Sodium Channels

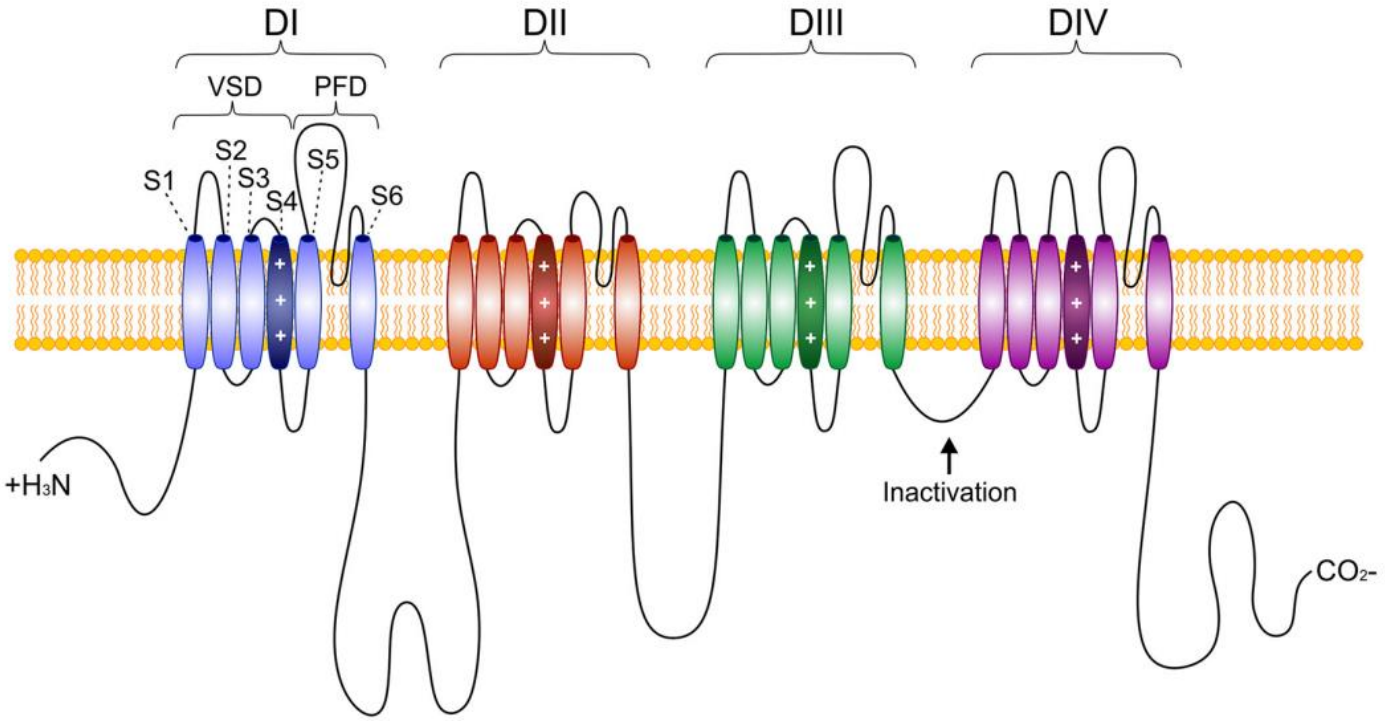


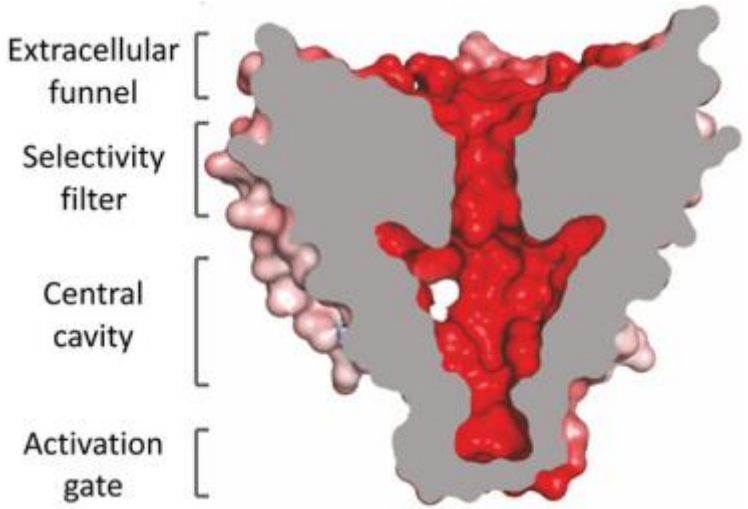
- Amplify the depolarizations of stimulus transducers
- Begin the process of action potential firing
- Have three different states
- Are grouped according to TTX sensitivity

$$\begin{array}{ccccccccc} \alpha C_0 & \leftrightarrow & \alpha C_1 & \leftrightarrow & \alpha C_2 & \leftrightarrow & \alpha C_3 & \leftrightarrow & \alpha C_4 & \leftrightarrow & \alpha O \\ & & & & & & & & \updownarrow & & \updownarrow \\ & & & & & & & & \alpha C_4 & \leftrightarrow & \alpha OI \end{array}$$



Voltage Gated Sodium Channels





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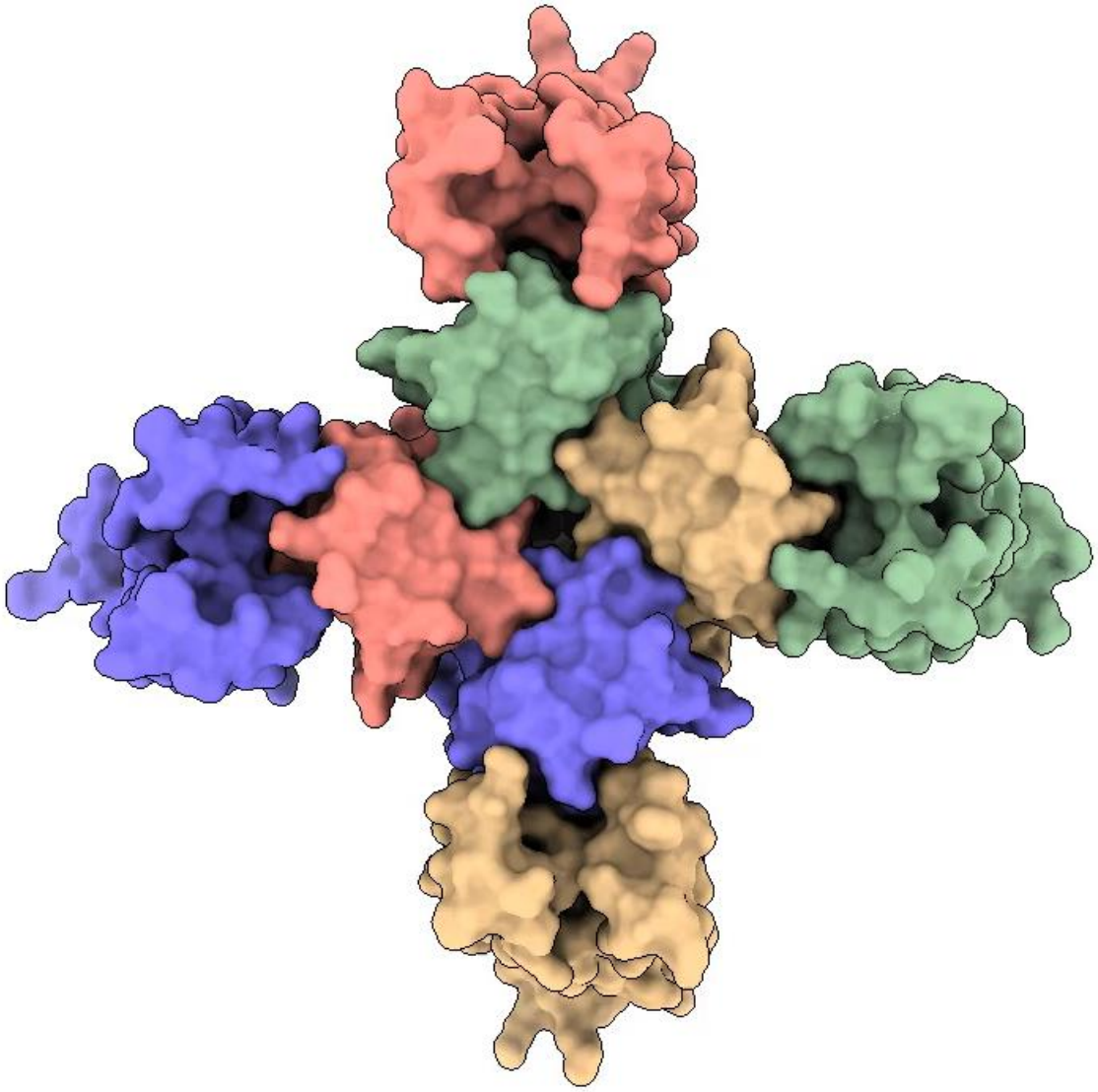
Voltage Gated Sodium Channels

2 August 2022



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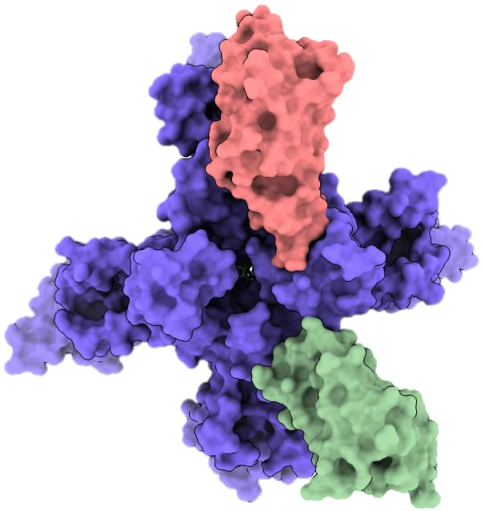
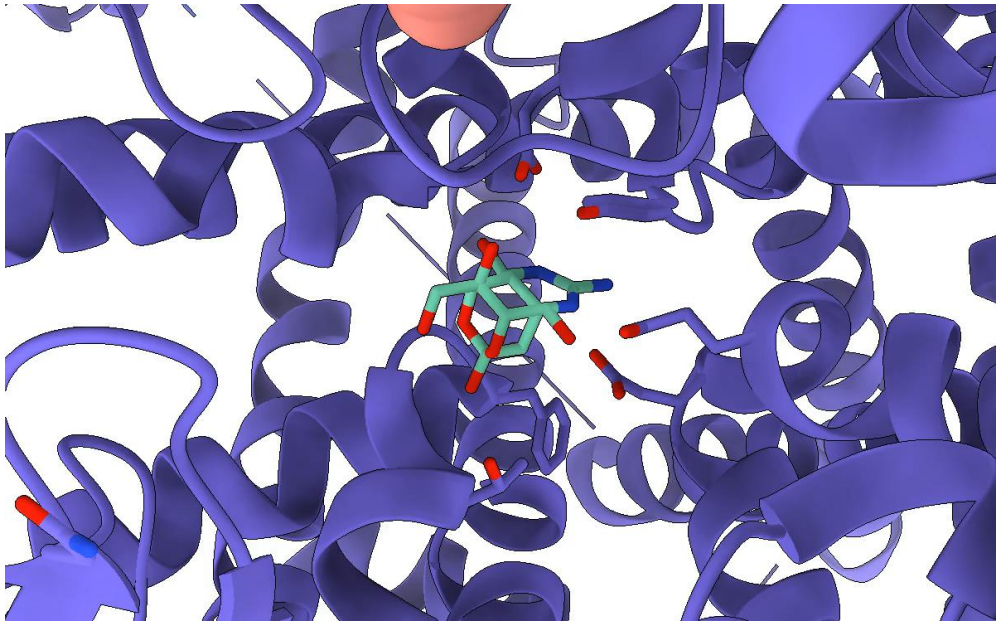
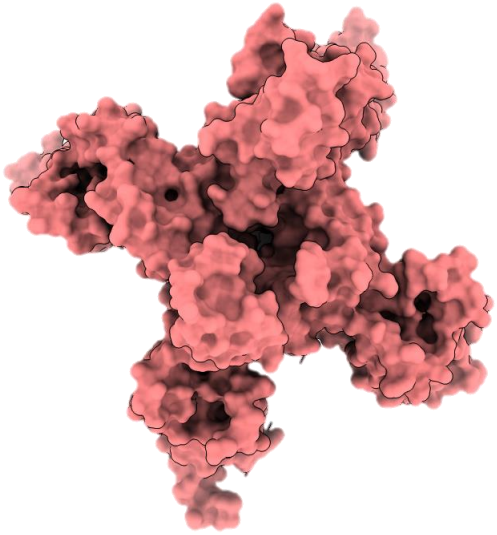
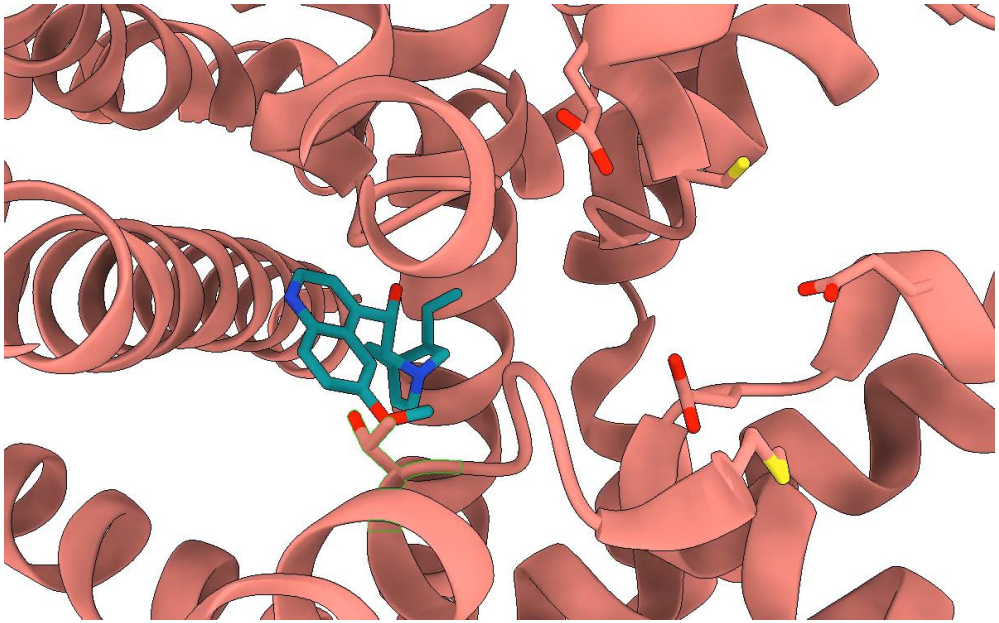
Tettrodotoxin Sensitivity

2 August 2022









$Na_V1.8$

- Exclusively present in the peripheral nervous system
- Levels drop as organism age
- Slow fast-inactivation leads to persistent current
- Expression increases by about 4-fold after injury

$$\alpha C_0 \leftrightarrow \alpha C_1 \leftrightarrow \alpha C_2 \leftrightarrow \alpha C_3 \leftrightarrow \alpha C_4 \leftrightarrow \alpha O$$
$$\updownarrow \qquad \updownarrow$$
$$\alpha C_4 \leftrightarrow \alpha OI$$

Small-Fiber Neuropathy

- Mutation of the SCN10A gene results in faster recovery of $Na_V1.8$ and slower fast-inactivation

Diabetic Neuropathy

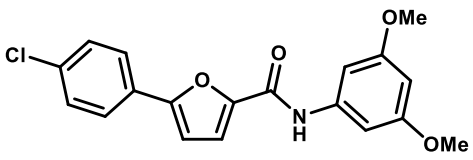
- Methyl glyoxal build up interacts with $Na_V1.8$
- Raises the membrane threshold for fast-inactivation of $Na_V1.8$

Multiple Sclerosis

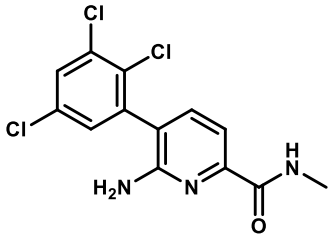
- Expression of $Na_V1.8$ withing the CNS causes misfiring of neurons and contributes to the symptoms of MS

Possum Mice

- A specific mutation of the SCN10A gene causes possum like behavior in mice
- When “scruffed” the mice become temporarily catatonic and “play dead”



A-803467



PF-01247324

$Na_V1.9$

- Difficult to study in isolation
- Least conserved among species
- Activation is very hyperpolarized, and inactivation is ultra slow
- Mutations have been implicated in a number of neuropathies

$Na_V1.3$

- In rats, levels peak early in embryonic development and disappears by adulthood
- Re-expressed after nerve injuries
- Glial cell line derived neurotrophic factor (GDNF) and neural growth factor (NGF) prevent neurotrophic pain
- 85% homologous with $Na_V1.2$

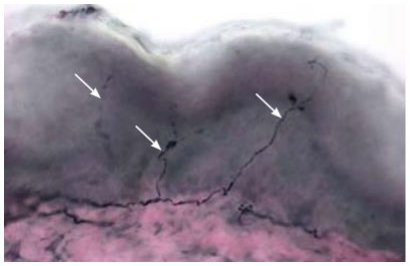
$Na_V1.7$

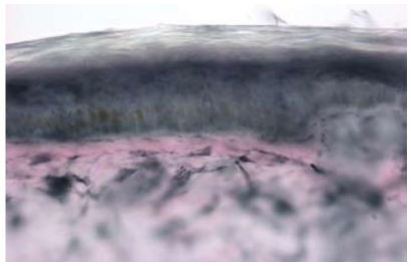
$$\alpha C_0 \leftrightarrow \alpha C_1 \leftrightarrow \alpha C_2 \leftrightarrow \alpha C_3 \leftrightarrow \alpha C_4 \leftrightarrow \alpha O$$
$$\updownarrow \qquad \updownarrow$$
$$\alpha C_4 \leftrightarrow \alpha OI$$

- Minimal depolarization required for activation
- Quickly deactivates once open
- Slow closed-state inactivation
- $Na_v1.7$ works to amplify small membrane depolarizations that are the result of stimulus transduction

Small-Fiber Neuropathy

- Mutation of the SCN9A gene results in $Na_V1.7$ remaining partially open
- Sodium Calcium exchange is then reversed
- Increased cytosolic calcium degrades neurons





Inherited Erythromyalgia

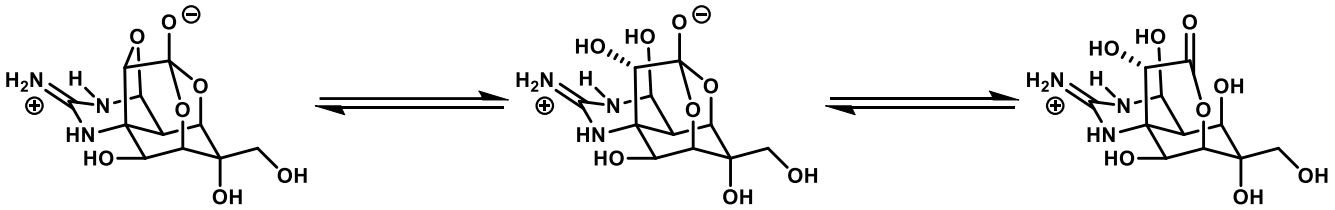
- Gain of function resulting in lowering the activation threshold of $Na_V1.7$

Paroxysmal Extreme Pain Disorder

- Characterized by episodes of extreme pain
- Slower open-state inactivation and faster reset of $Na_V1.7$
- Reopening of inactivated channels

Congenital Insensitivity to Pain (CIP)

- Rare, autosomal recessive disorder
- Characterized by an inability to feel pain
- Results from mutation of the *SCN9A* gene
  - *W987X, I767X or S459X*



Minoru Isobe

With Norio Ohyabu and Toshio Nishikawa

- Completed the first asymmetric total synthesis of (-)–tetrodotoxin
- 69 total steps
- 0.5% yield
- Relied on the careful selection of protecting groups

Tetrodotoxin:

- First isolated in 1909 by Yoshizumi Tahara
- Structural elucidation was published in 1964 by Woodward
- Found in puffer fish (fugu) and in the venom of some octopi (blue ringed octopus)
- Is produced by symbiotic bacteria

Justin Du Bois

Andrew Hinman

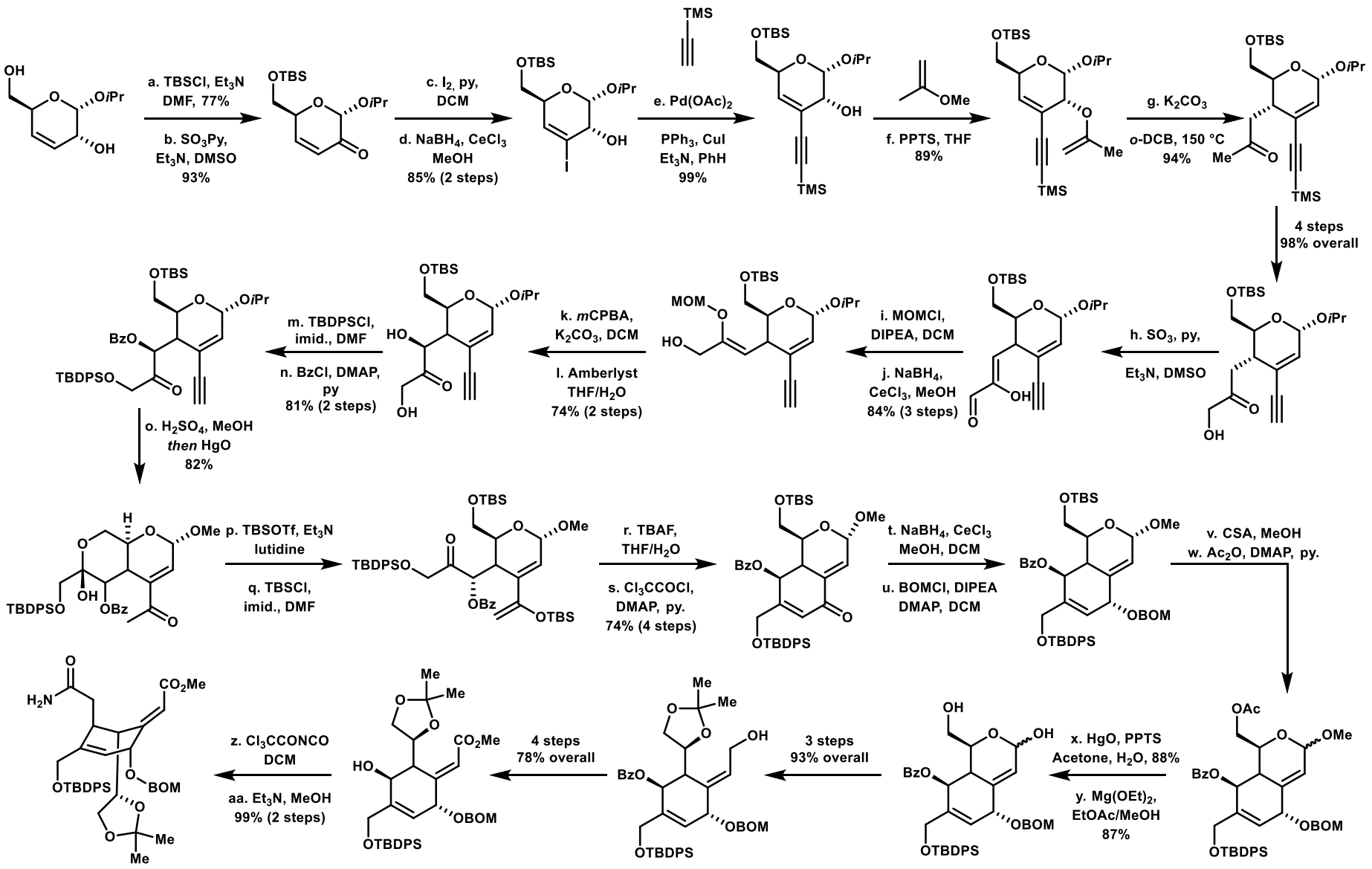
Published 5 months later

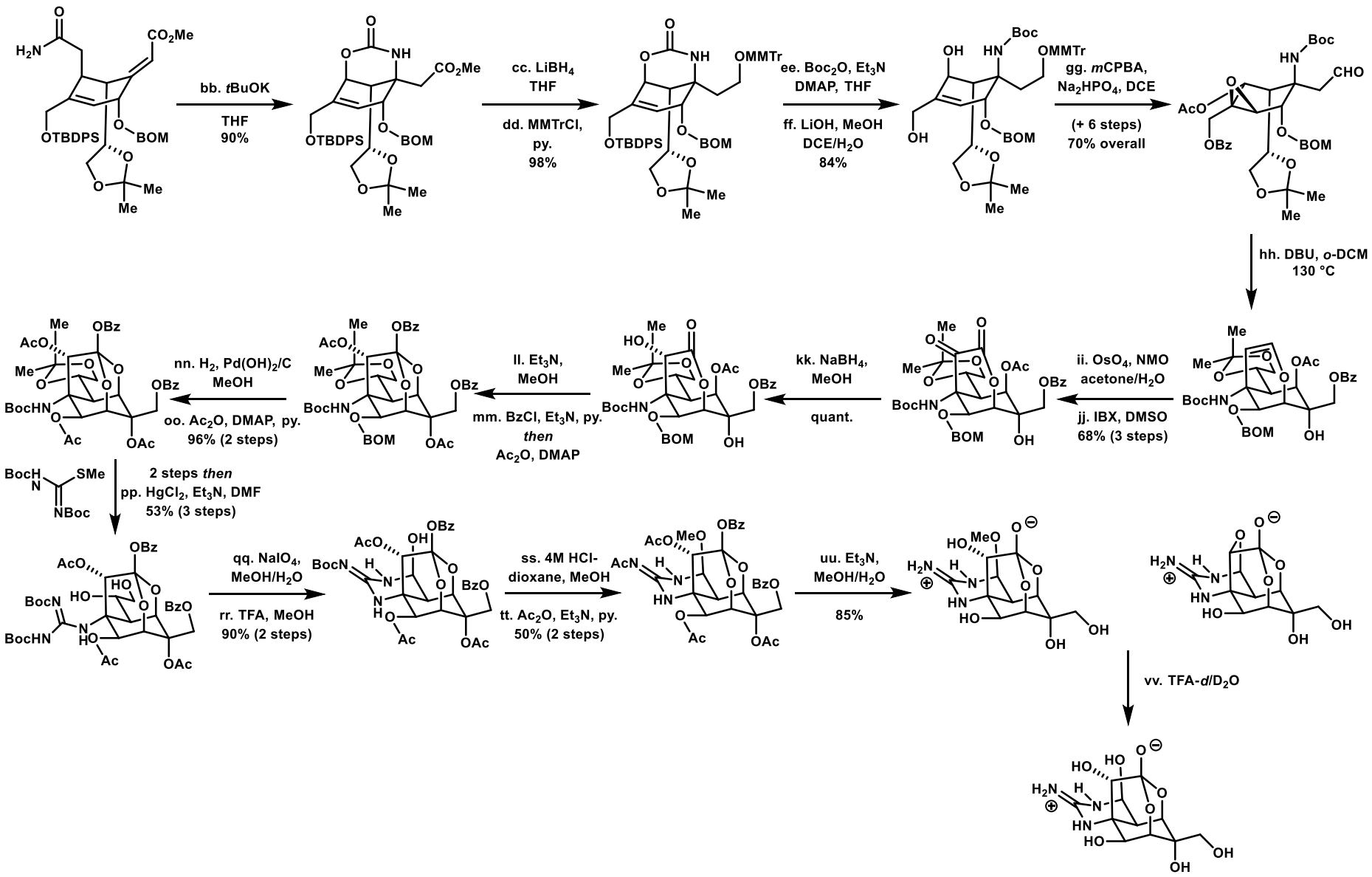
- Completed the total synthesis of (-)–tetrodotoxin
- 28 total steps
- 2.4% yield
- Showcased the power of selective C-H fuctionalization

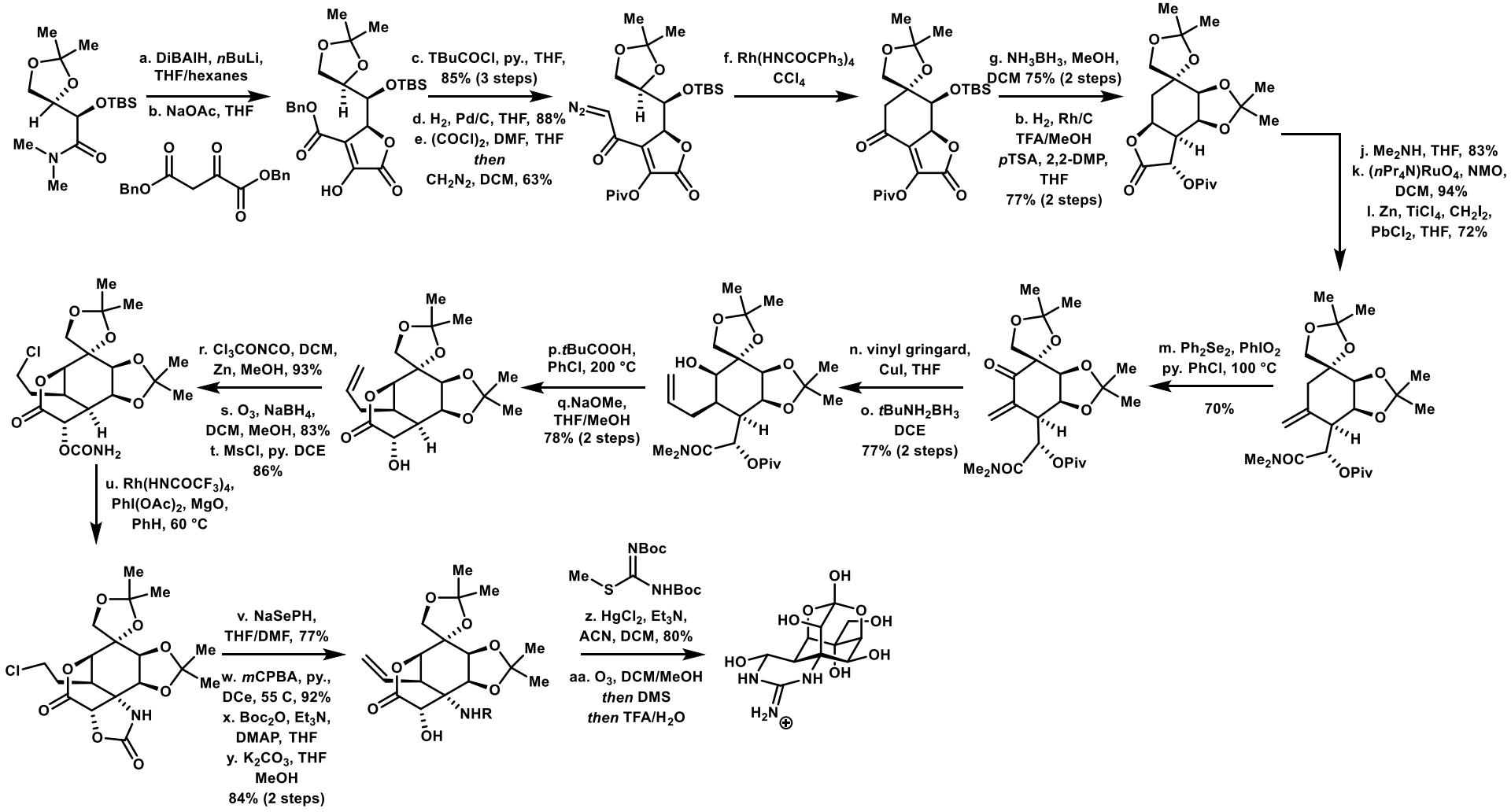
Others:

- Trauner 2022 (22 steps), Isobe 2004 (39 steps), Sato 2008 (34 steps) Sato 2010 (32 steps), Fukuyama 2017 (31 steps), Fukuyama 2020 (29 steps)

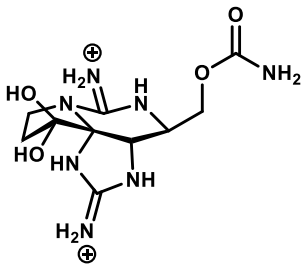
Tetrodotoxin (TTX) Synthesis




bb. tBuOK
THF
90%
cc. LiBH4
THF
dd. MMTrCl,
py.
98%
ee. Boc2O, Et3N
DMAP, THF
ff. LiOH, MeOH
DCE/H2O
84%
gg. mCPBA,
Na2HPO4, DCE
(+ 6 steps)
70% overall
hh. DBU, o-DCM
130 °C
ii. OsO4, NMO
acetone/H2O
jj. IBX, DMSO
68% (3 steps)
kk. NaBH4,
MeOH
quant.
ll. Et3N,
MeOH
mm. BzCl, Et3N, py.
then
Ac2O, DMAP
nn. H2, Pd(OH)2/C
MeOH
oo. Ac2O, DMAP, py.
96% (2 steps)
2 steps then
pp. HgCl2, Et3N, DMF
53% (3 steps)
qq. NaIO4,
MeOH/H2O
rr. TFA, MeOH
90% (2 steps)
ss. 4M HCl-
dioxane, MeOH
tt. Ac2O, Et3N, py.
50% (2 steps)
uu. Et3N,
MeOH/H2O
85%
vv. TFA-d/D2O


a. DiBAlH, nBuLi, THF/hexanes
b. NaOAc, THF
c. TBuCOCl, py., THF, 85% (3 steps)
d. H2, Pd/C, THF, 88%
e. (COCl)2, DMF, THF
then
CH2N2, DCM, 63%
f. Rh(HNCOCPh3)4
CCl4
g. NH3BH3, MeOH, DCM 75% (2 steps)
b. H2, Rh/C
TFA/MeOH
pTSA, 2,2-DMP, THF
77% (2 steps)
j. Me2NH, THF, 83%
k. (nPr4N)RuO4, NMO, DCM, 94%
l. Zn, TiCl4, CH2I2, PbCl2, THF, 72%
m. Ph2Se2, PhIO2
py. PhCl, 100 °C
70%
n. vinyl gringard, CuI, THF
o. tBuNH2BH3
DCE
77% (2 steps)
p. tBuCOOH, PhCl, 200 °C
q. NaOMe, THF/MeOH
78% (2 steps)
r. Cl3CONCO, DCM, Zn, MeOH, 93%
s. O3, NaBH4, DCM, MeOH, 83%
t. MsCl, py. DCE
86%
u. Rh(HNCOCF3)4, PhI(OAc)2, MgO, PhH, 60 °C
v. NaSePH, THF/DMF, 77%
w. mCPBA, py., DCe, 55 C, 92%
x. Boc2O, Et3N, DMAP, THF
y. K2CO3, THF MeOH
84% (2 steps)
z. HgCl2, Et3N, ACN, DCM, 80%
aa. O3, DCM/MeOH
then DMS
then TFA/H2O

Tetrodotoxin



Saxitoxin:

- Isolated in 1957 by E. J. Schantz
- Structure published in 1975 by E. J. Schantz
- Produced by algae and bioaccumulates in shellfish

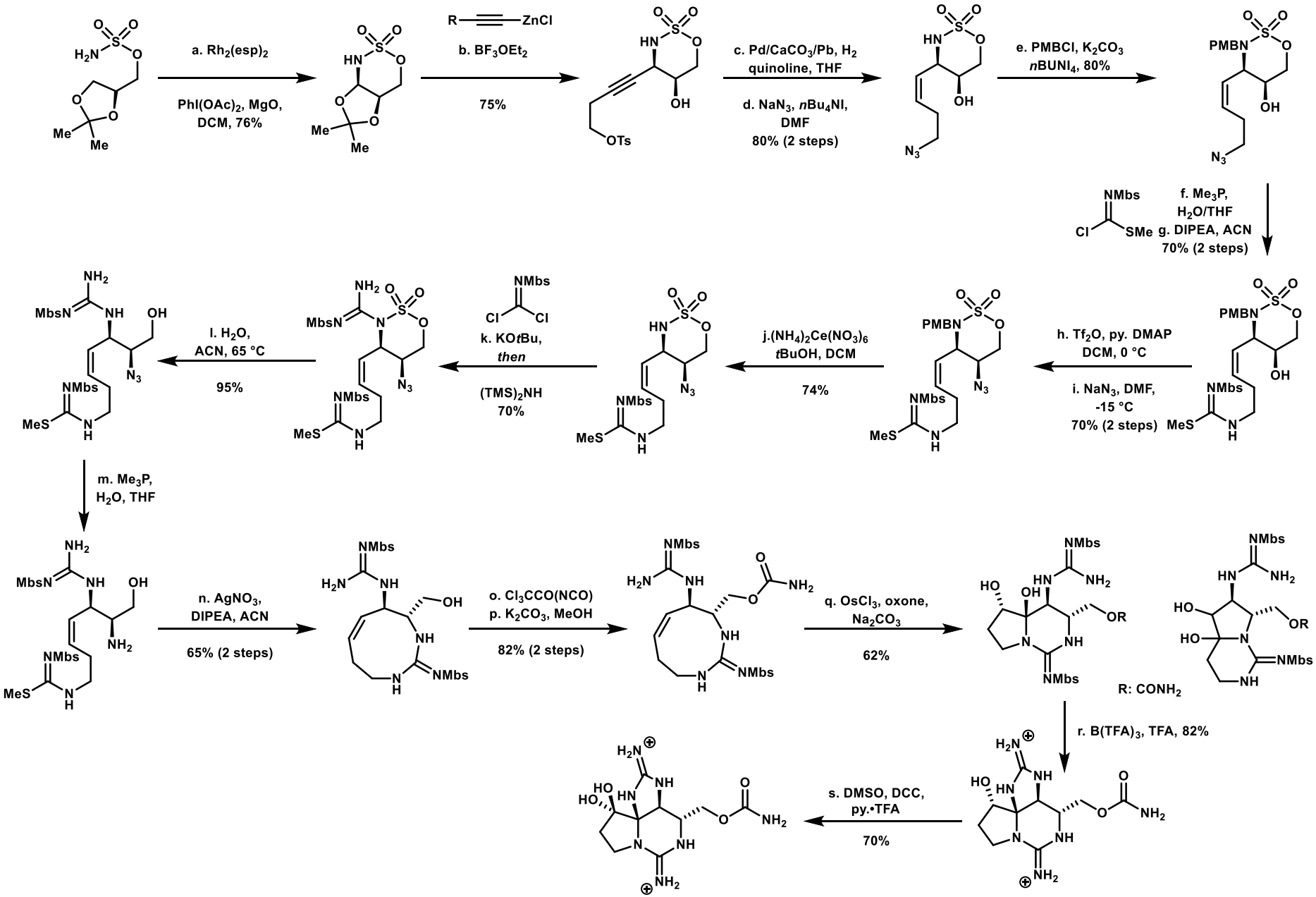
With J. J. Fleming:

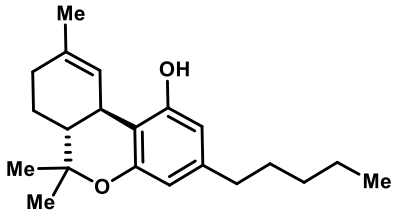
- Completed the first and second asymmetric total syntheses of saxitoxin
- 19 and 14 steps, respectively

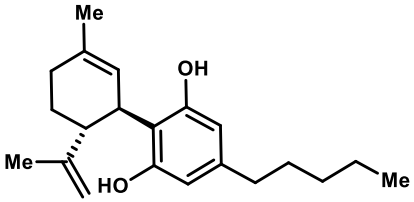
H. Anderson

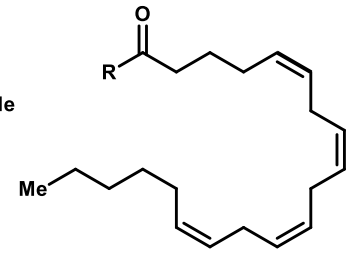
Saxitoxin (TTX) Synthesis

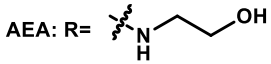
2 August 2022

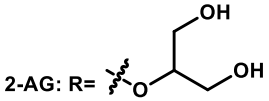


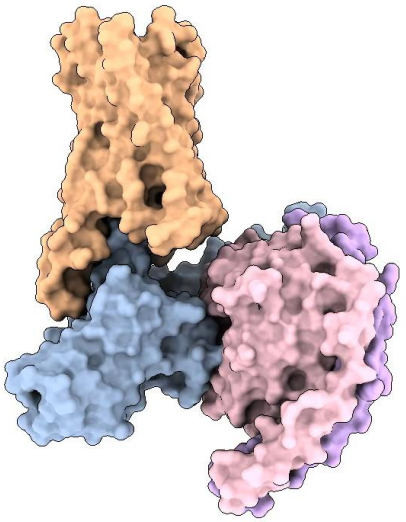












CB_1-GPCR

Cannabinoid Receptors

- Discovery of THC in the 1960s lead to the search for the corresponding receptors
- CB_1 was discovered first followed by CB_2 in 1993
- GPCRs that have 2 endogenous ligands (endocannabinoids 2-AG and AEA)
- Like OPRs, agonism of the CB_1 and CB_2 receptors inhibits calcium channels and prevents the release of neurotransmitters into the synaptic cleft

Potential Therapeutic Targets

CB_1:

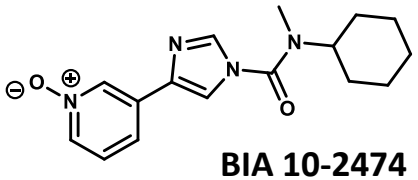
- Side effects: anxiety, panic, euphoria, altered state of consciousness and hallucinations

FAAH (Fatty acid amide hydrolase):

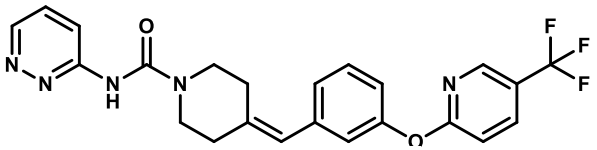
- Inhibition increases levels of endocannabinoids by preventing catabolism

CB_2:

- Theorized to provide the therapeutic effect of cannabinoids without the psychoactive effects



BIA 10-2474



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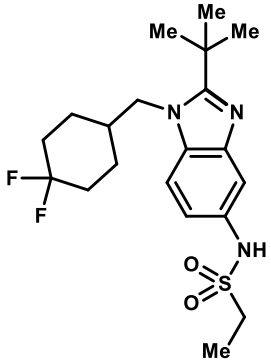

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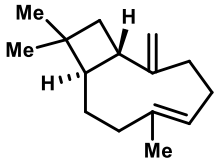
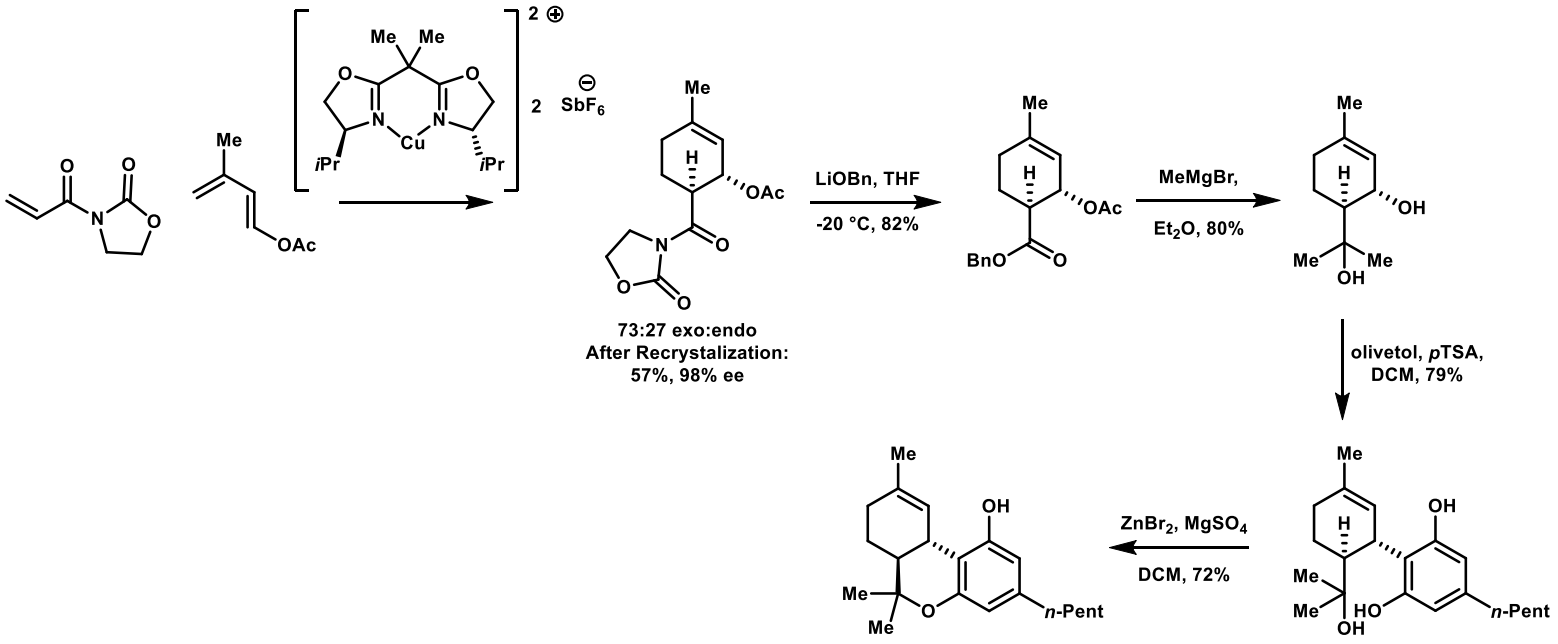
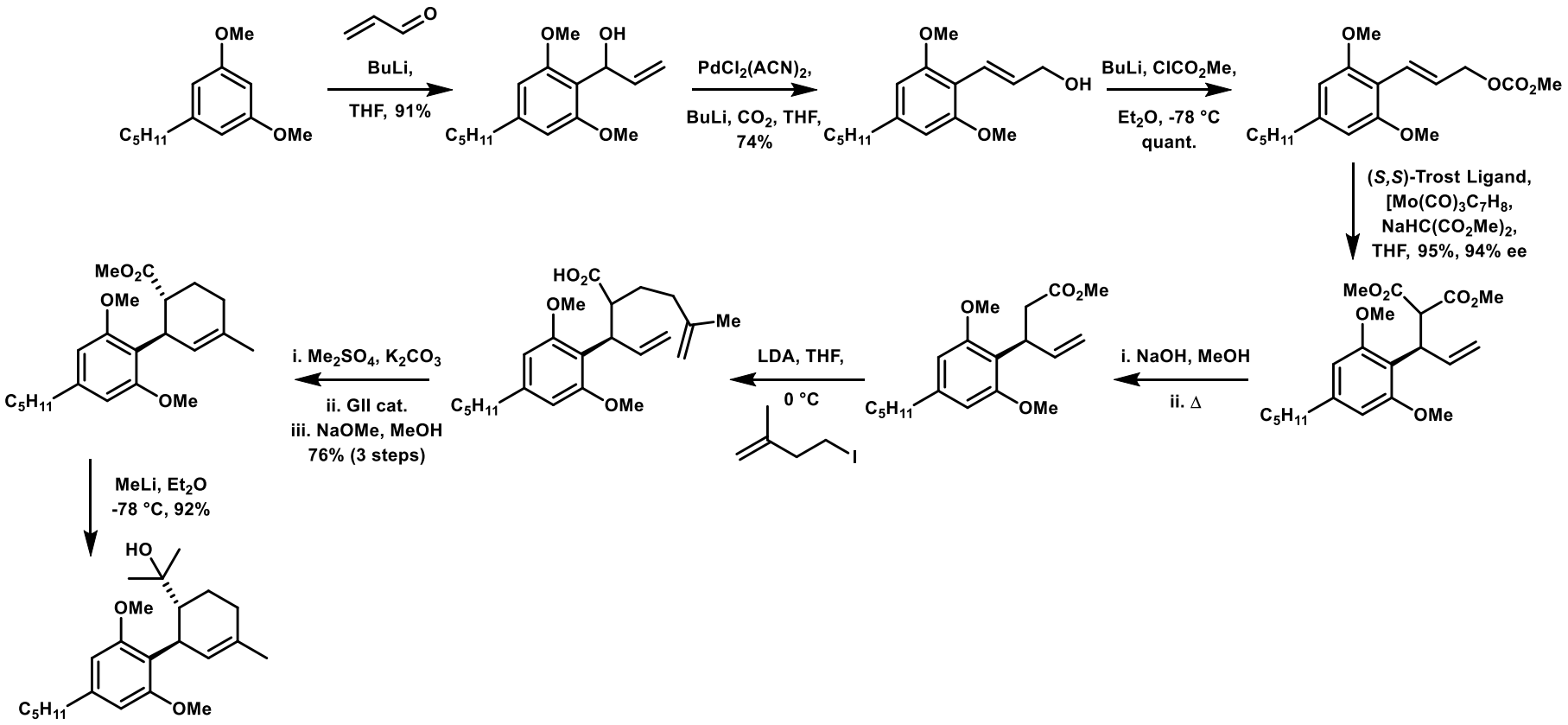


Caryophyllene

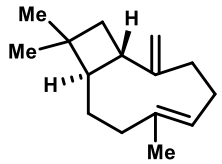
THC Total Synthesis

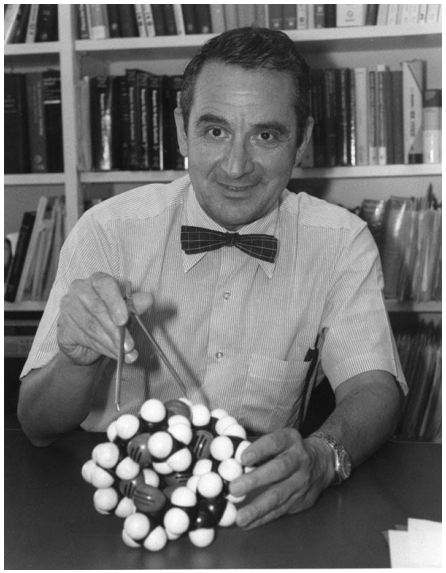



OMe
C5H11
OMe
BuLi,
THF, 91%
OMe OH
C5H11
OMe
PdCl2(ACN)2,
BuLi, CO2, THF,
74%
OMe
OH
C5H11
OMe
BuLi, ClCO2Me,
Et2O, -78 °C
quant.
OMe
OCO2Me
C5H11
OMe
(S,S)-Trost Ligand,
[Mo(CO)3C7H8,
NaHC(CO2Me)2,
THF, 95%, 94% ee
MeO2C
CO2Me
OMe
C5H11
OMe
i. NaOH, MeOH
ii. Δ
OMe
CO2Me
C5H11
OMe
LDA, THF,
0 °C
I
HO2C
OMe
Me
C5H11
OMe
i. Me2SO4, K2CO3
ii. GII cat.
iii. NaOMe, MeOH
76% (3 steps)
MeO2C
OMe
C5H11
OMe
MeLi, Et2O
-78 °C, 92%
HO
OMe
C5H11
OMe

Caryophyllene

Caryophyllene:





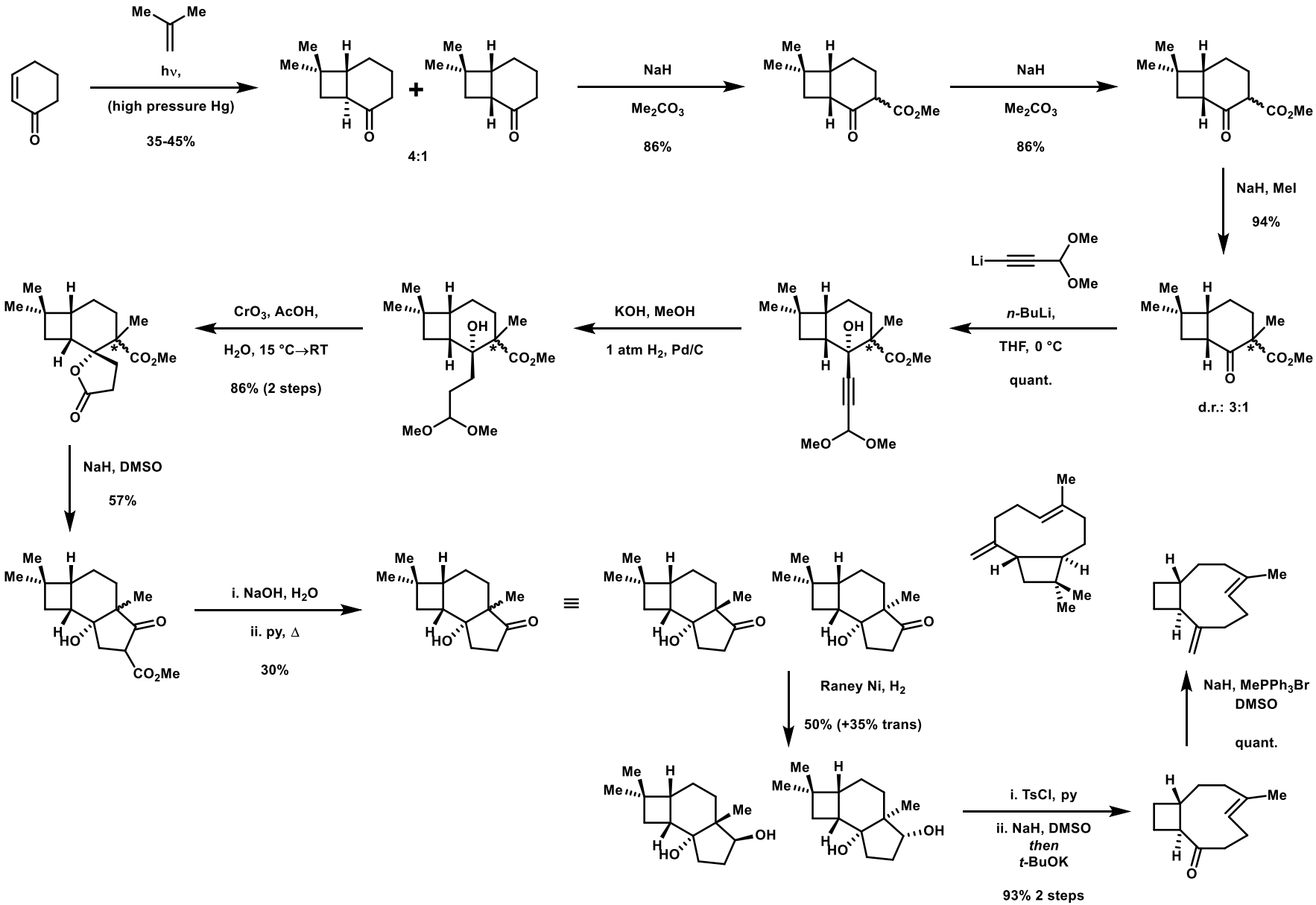
With Rajat B Mitra and Hisashi Uda:

- Completed the first synthesis of caryophyllene and iso-caryophyllene
- Isolated from cannabis, cloves and Indian bay leaves

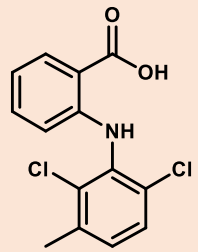
H. Anderson

Caryophyllene Total Synthesis

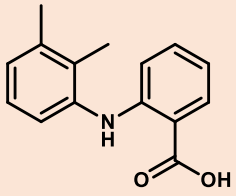
2 August 2022



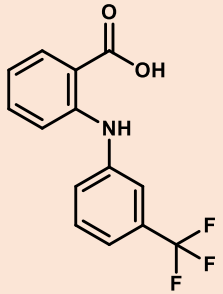
Fin



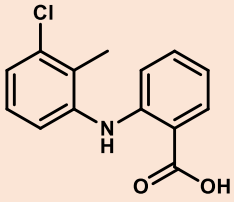
meclofenamic acid



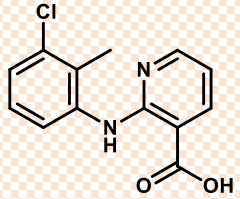
mefenamic acid



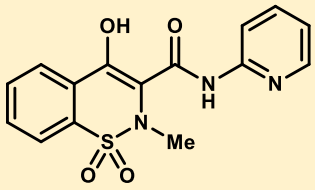
flufenamic acid



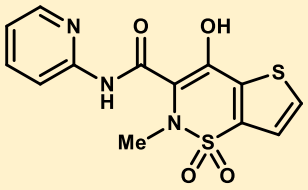
tolfenamic acid



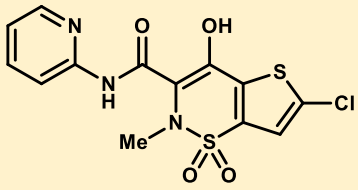
clonixin



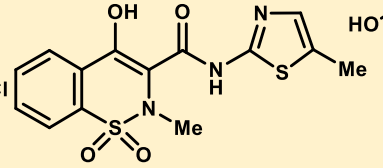
piroxicam



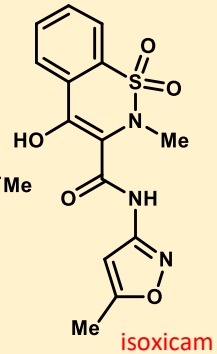
tenoxicam



lornoxicam

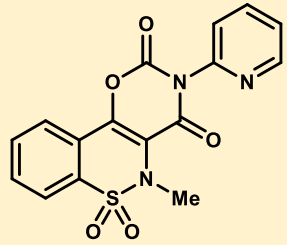


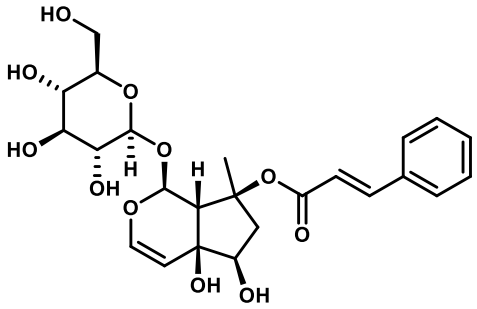
meloxicam



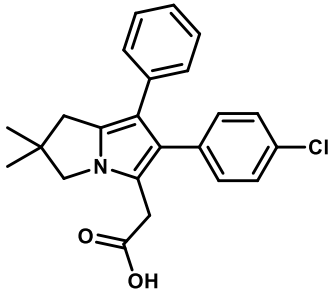
isoxicam

droxicam

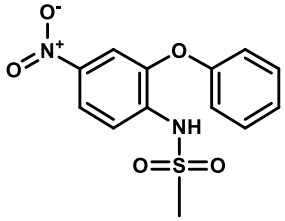




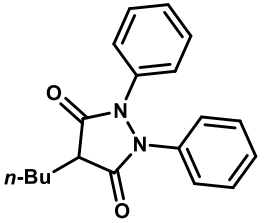
harpagoside



licofelone



nimesulide



phenylbutazone