Biosynthesis

Biosynthesis of Alkaloids

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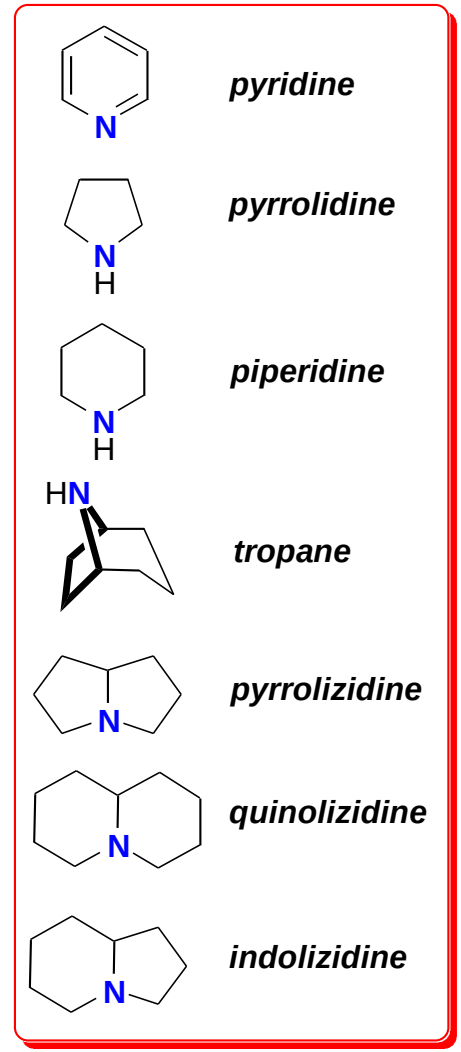
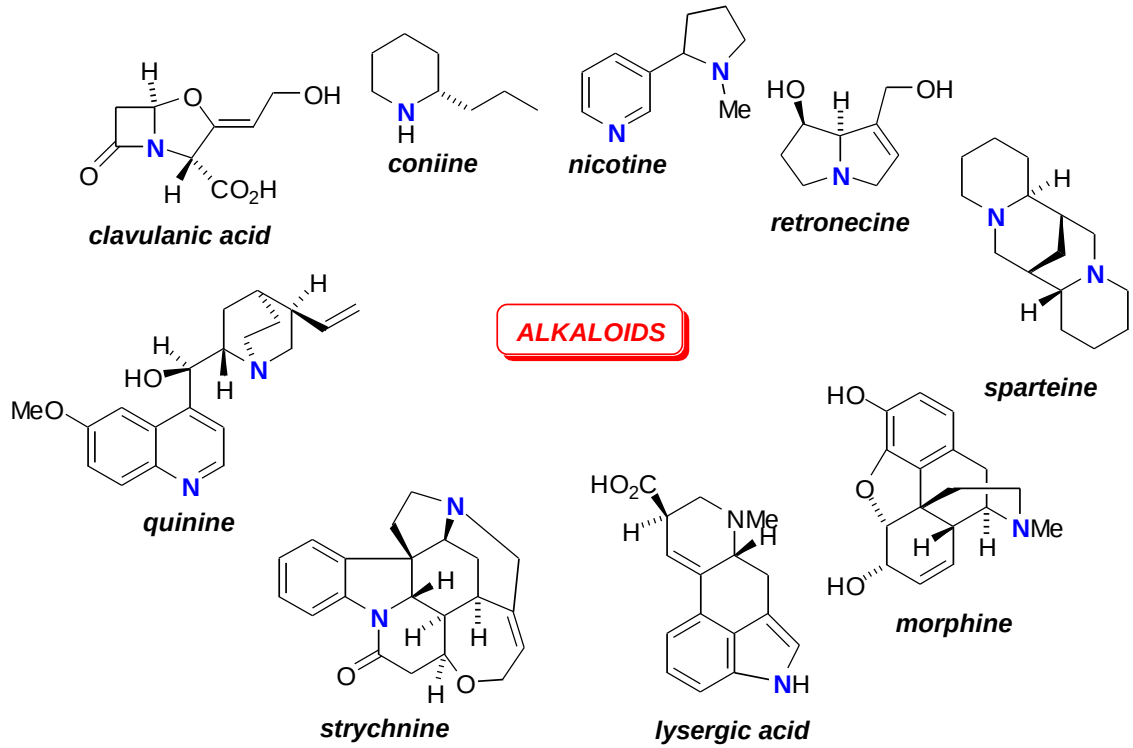
Format & Scope of Lectures

- ***What are alkaloids?***
  - definitions, 1° metabolism → α-amino acids (Lys, Orn)
    - the citric acid cycle – oxaloacetate & α-ketoglutarate
    - pyridoxal – transamination, racemisation & decarboxylation
- ***Lysine and ornithine derived alkaloids***
  - pyridine, pyrrolidine, piperidine, tropane, pyrrolizidine, quinolizidine & indolizidine
- ***Phenylalanine & tyrosine derived alkaloids***
  - *monocyclic alkaloids (mescaline case study – 'elucidating a biosynthetic pathway')*
    - *biological hydroxylation of aromatic rings - the 'NIH shift'*
  - *benzylisoquinolines (opium, aporphine & erythrina alkaloids)*
    - *oxidative phenolic coupling & dienone – phenol rearrangements*
  - *amaryllidaceae alkaloids*
- ***Tryptophan derived alkaloids***
  - simple indole alkaloids: *e.g*. serotonin
  - mixed tryptophan/mevalonate (isoprenoid) alkaloids:
    - DMAPP derived: ergot alkaloids
    - secologanin derived: vinca-, strychnos- & quinine alkaloids *etc.*
- ***Non-ribosomal peptides & derivatives***
  - cyclic di-peptides (diketopiperazines)
  - penicillins & cephalosporins
  - cyclic polypeptides



Alkaloids

- ***Definitions:***
  - ***originally –*** '*a natural product that could be extracted out of alkaline but not acidic water*' (*i.e.* containing a basic amine function that protonated in acid)
  - ***more generally -*** '*any non-peptidic & non-nucleotide nitrogenous secondary metabolite*'



Why do Organisms Make Alkaloids?

...indeed, secondary metabolites in general:

1. At some specific, as yet unidentified, point in the life of the organism these compounds have a 1° metabolic function

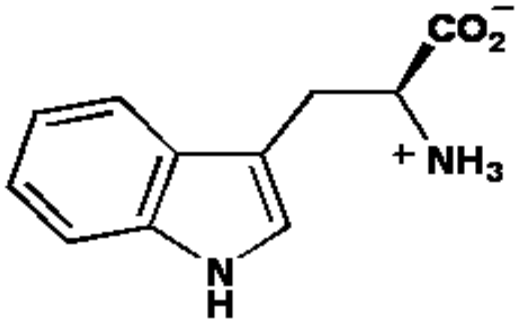
2. These compounds are evolutionary relics, once having had a metabolic function but no longer

3. These compounds are waste/detoxification/overflow(resevoir) products

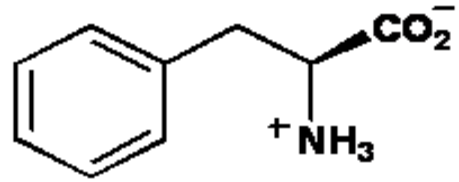
4. ***These compounds are ecological survival agents (repelents/attractants etc.) evolved to enhance an organisms ability to adapt to its environment***
   - *e.g.* frog toxins being highly distasteful to predators

5. The *processes* of secondary metabolism allow a network of highly evolved enzymes to persist which although not currently required for 1° metabolism may be called on if adjustment to changing circumstances is required

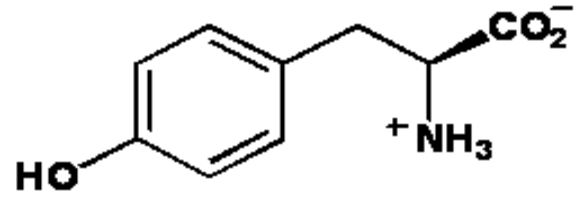
α-Amino Acids used to make Alkaloids



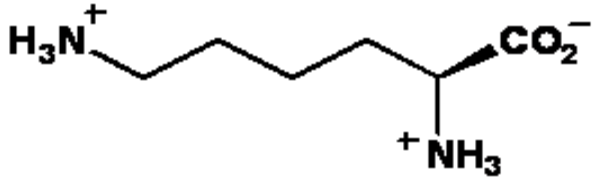
tryptophan



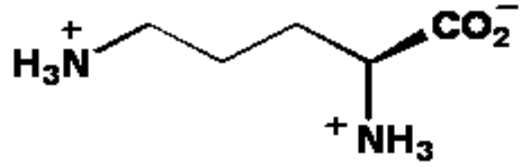
phenylalanine



tyrosine

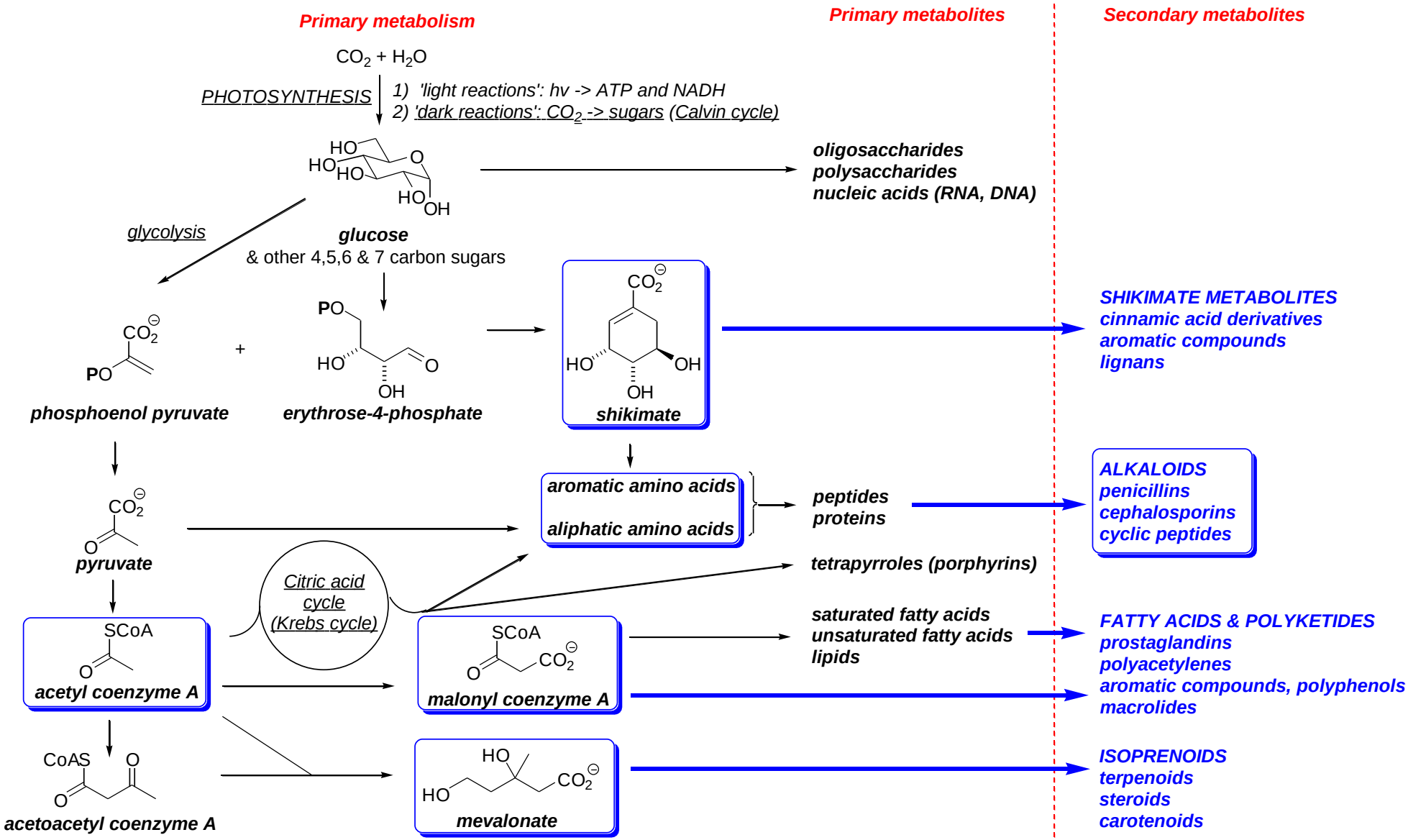


lysine



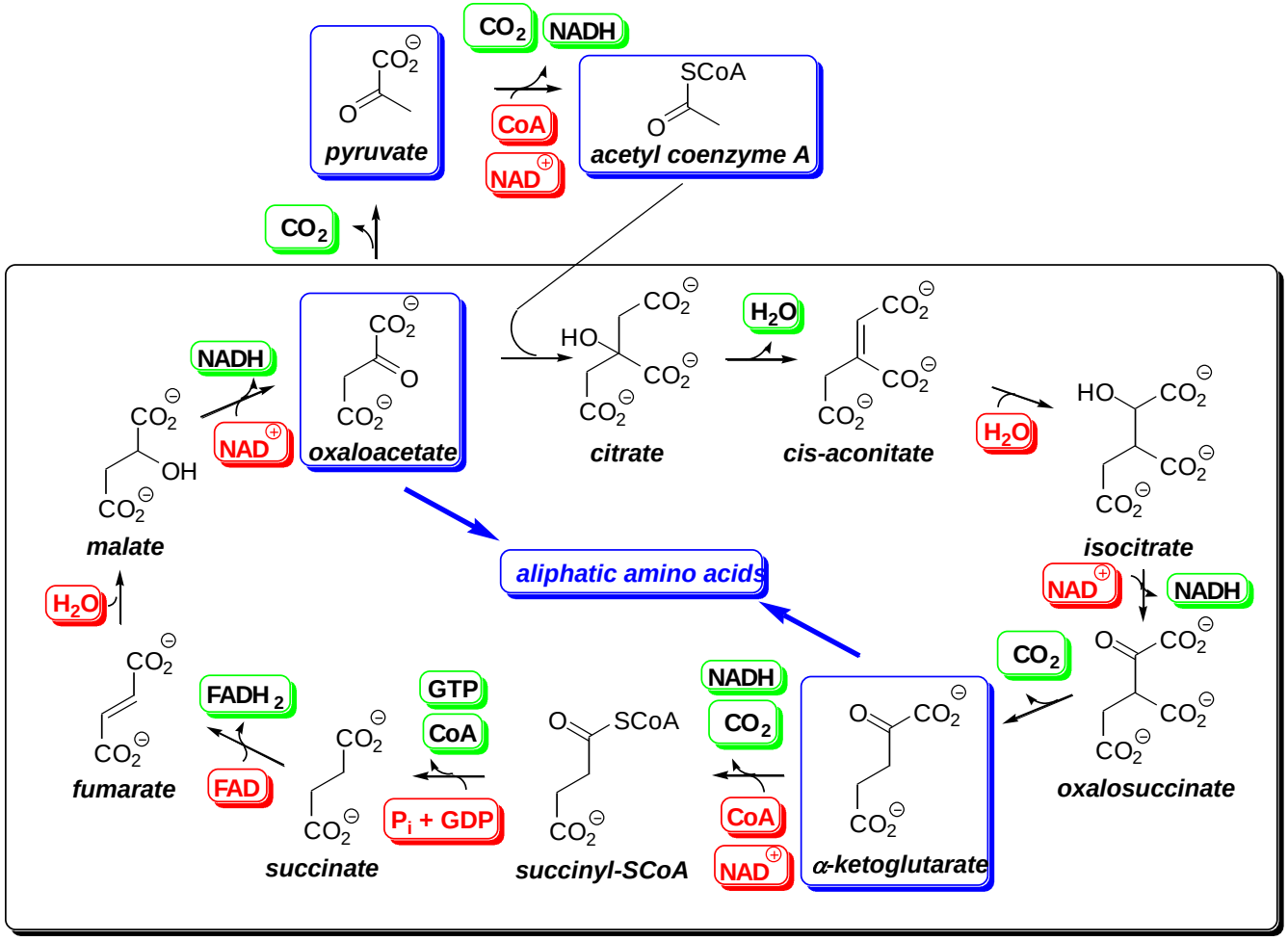
ornithine

Primary Metabolism - *Overview*



The Citric Acid Cycle

- ***The citric acid (Krebs) cycle*** is a major catabolic pathway of 1° metabolism that provides two key building blocks for aliphatic amino acid biosynthesis - ***oxaloacetate*** & ***α-ketoglutarate:***



THE CITRIC ACID CYCLE

OVERAL STOICHIOMETRY

1x *'acetate'*

\+

1x O_2

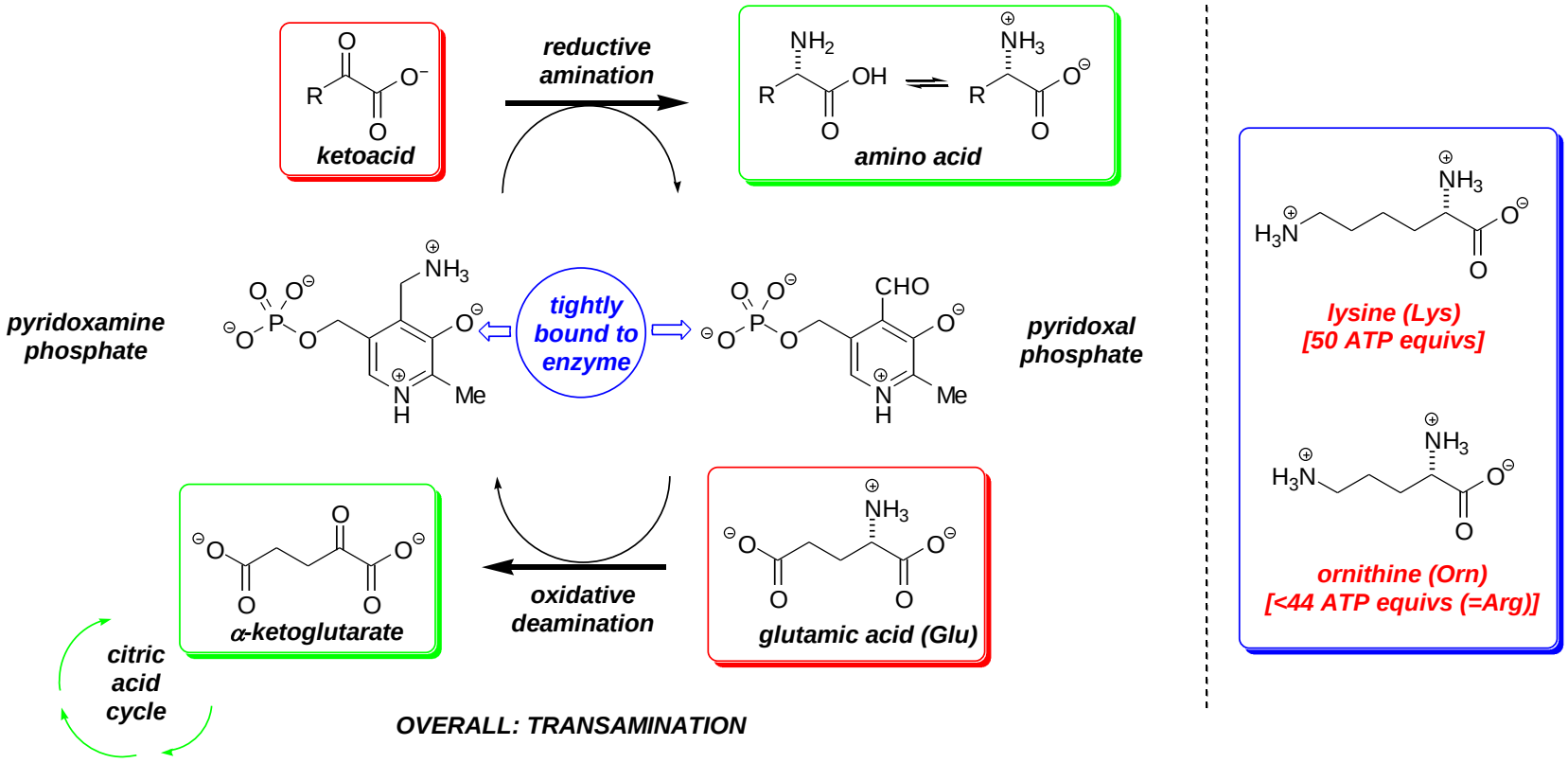
2x CO_2

\+

12x ATP *energy!*

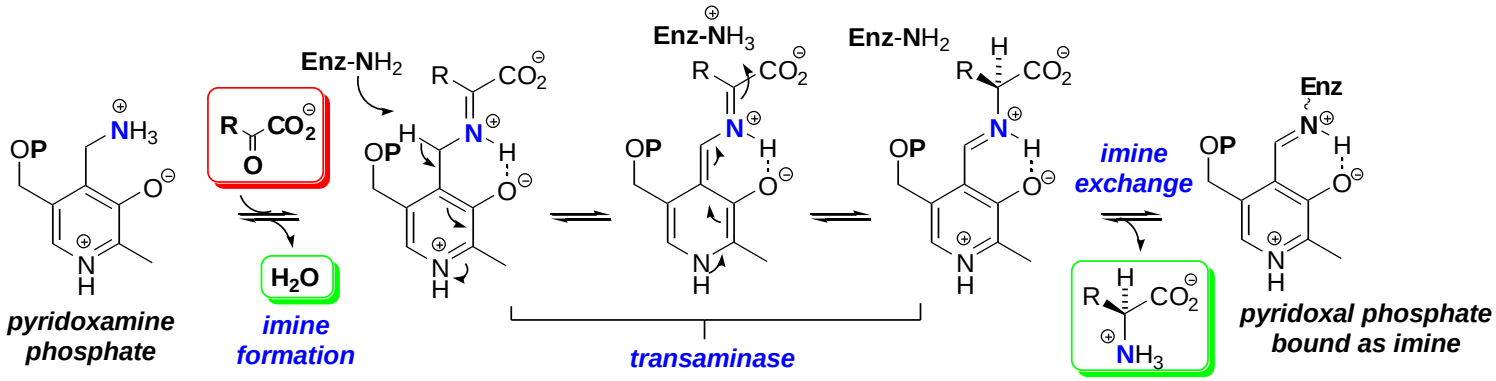
The Biosynthesis of Lysine & Ornithine

- ***Lysine & ornithine*** - the two most significant, *non-aromatic* α-amino acid ***precursors to alkaloids:***
  - *NB.* lysine (Lys) is proteinogenic whereas ornithine (Orn) is not
  - phenylalanine (Phe), tyrosine (Tyr) & tryptophan (Trp) from ***shikimate*** are the other important precursors
  - biosynthesis is *via* reductive amination of the appropriate α-ketoacid mediated by ***pyridoxal-5'-phosphate*** (***PLP***)

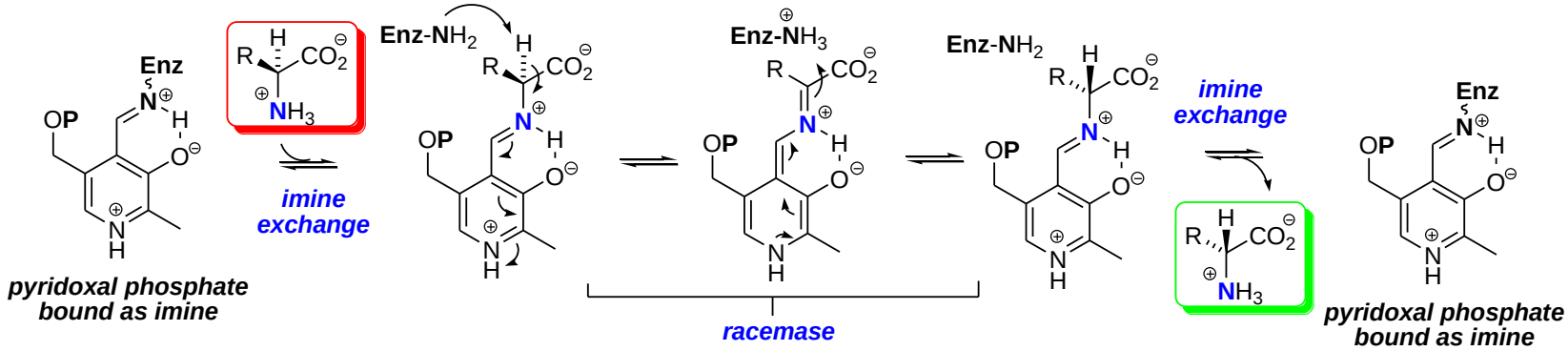


PLP Chemistry – *Transamination & Racemisation*

- ***Transamination*** – LHS → RHS (***reductive amination***); RHS → LHS (***oxidative deamination***):

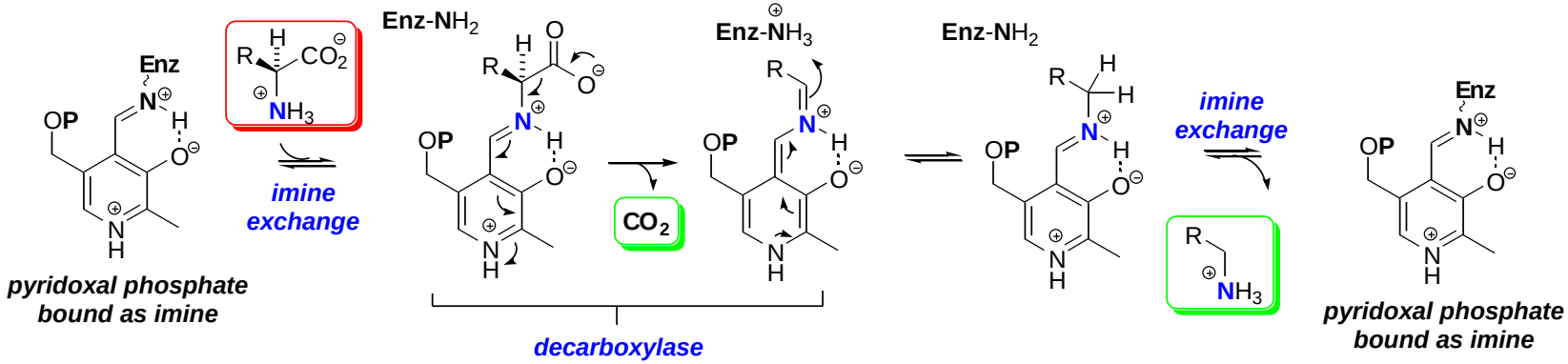


- ***Racemisation***:

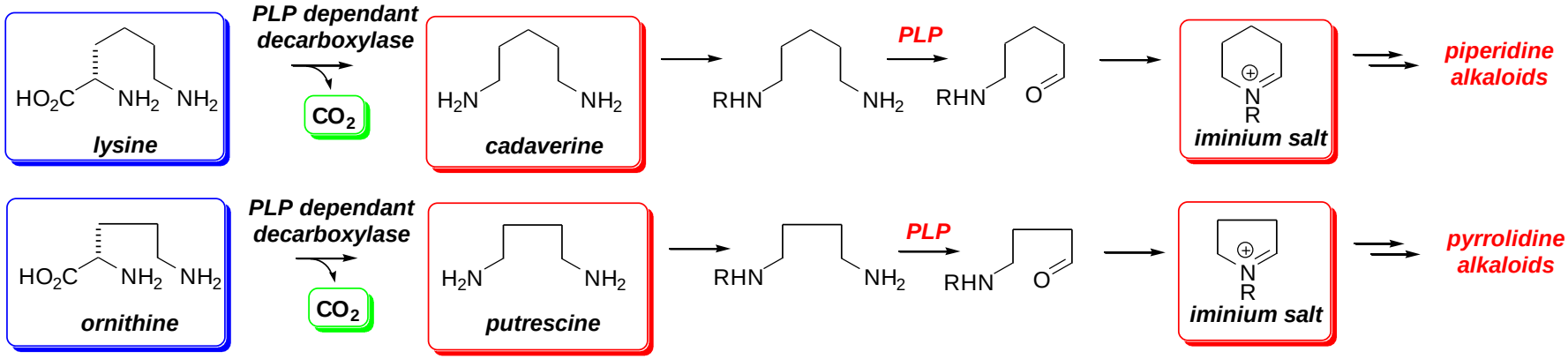


PLP Chemistry – *Decarboxylation*

- ***Decarboxylation***:

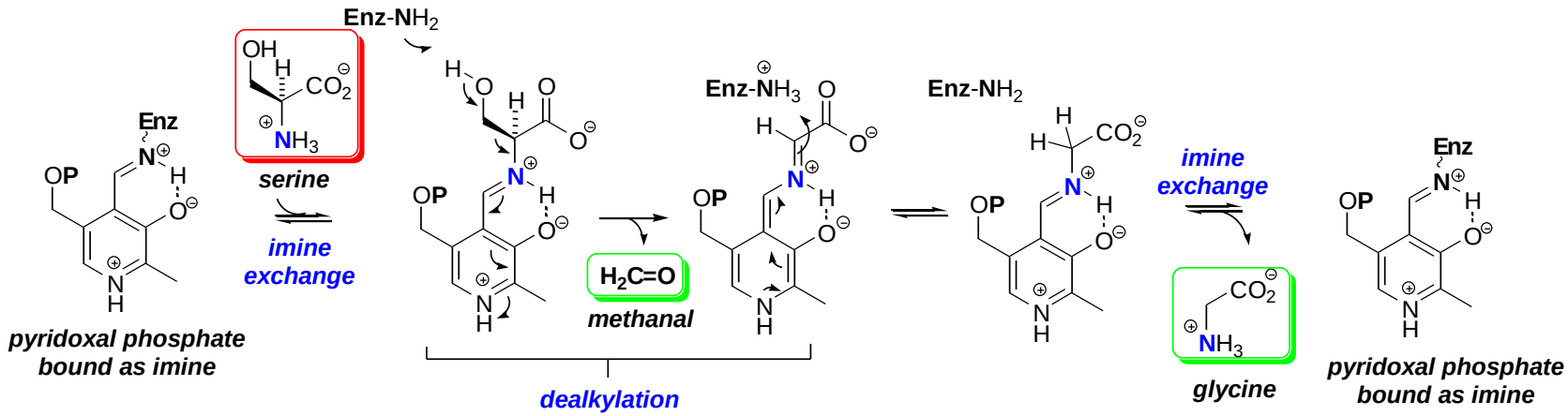


- Decarboxylation of ***lysine*** & ***ornithine***:

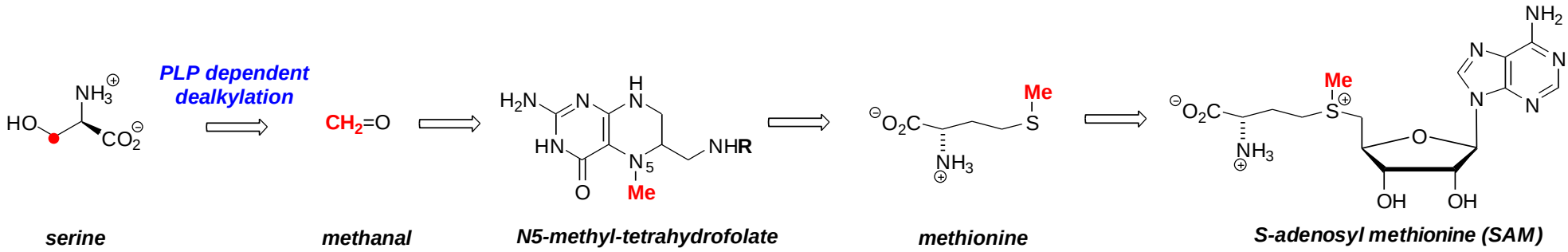


PLP Chemistry – *Dealkylation*

- ***Dealkylation***:
  - The cleavage of one carbon from serine is achieved by a PLP-dependent enzyme *via* dealkylation:

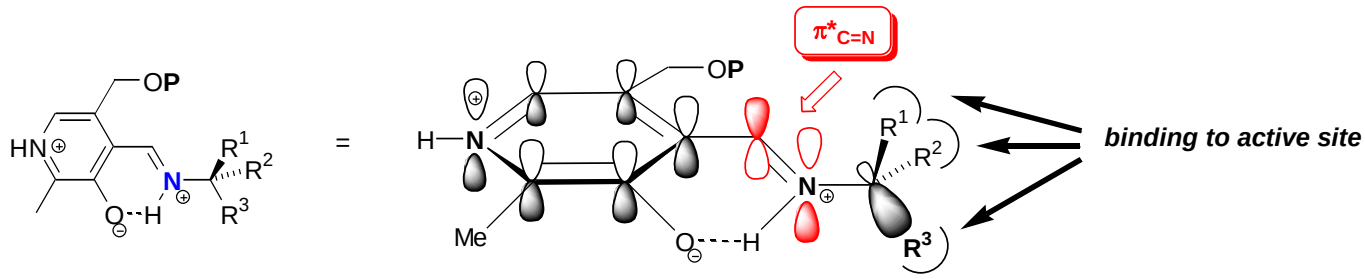


  - The carbon extruded as methanal in this process ends up as the methyl group of ***SAM*** (*via N*5-methyl tetrahydrofolate):

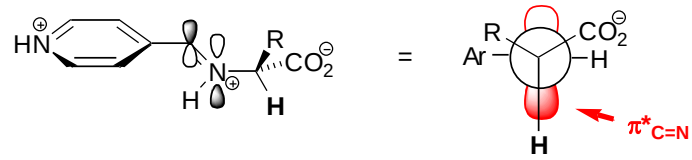


Control of PLP Activity – *Stereoelectronics*

- How does an enzyme control whether the PLP co-factor effects ***racemisation, decarboxylation or dealkylation?***
  - *i.e.* which bond will be cleaved?

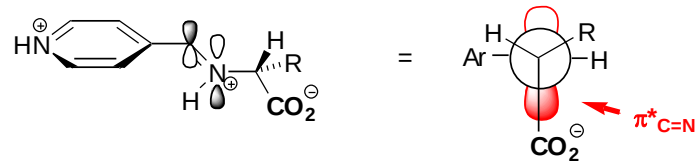


racemisation
(α proton loss)



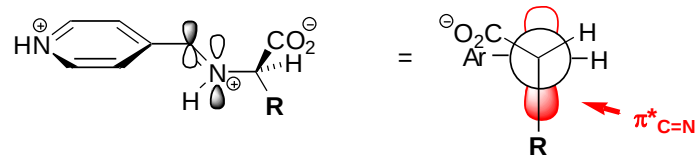
σC-H -> π*C=N *anti peri planar*

decarboxylation



σC-C -> π*C=N *anti peri planar*

dealkylation
(loss or R group)



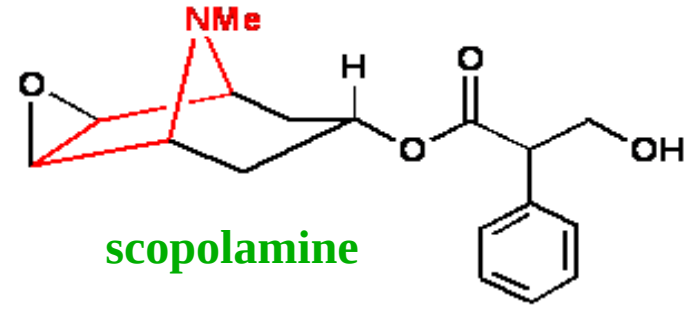
σC-C -> π*C=N *anti peri planar*

Pyridine, Piperidine & Pyrrolidine Alkaloids

Pyridine Alkaloids

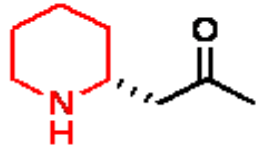
nicotine

Pyrrolidine Alkaloids

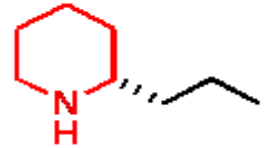


scopolamine

Piperidine Alkaloids



pelletierine

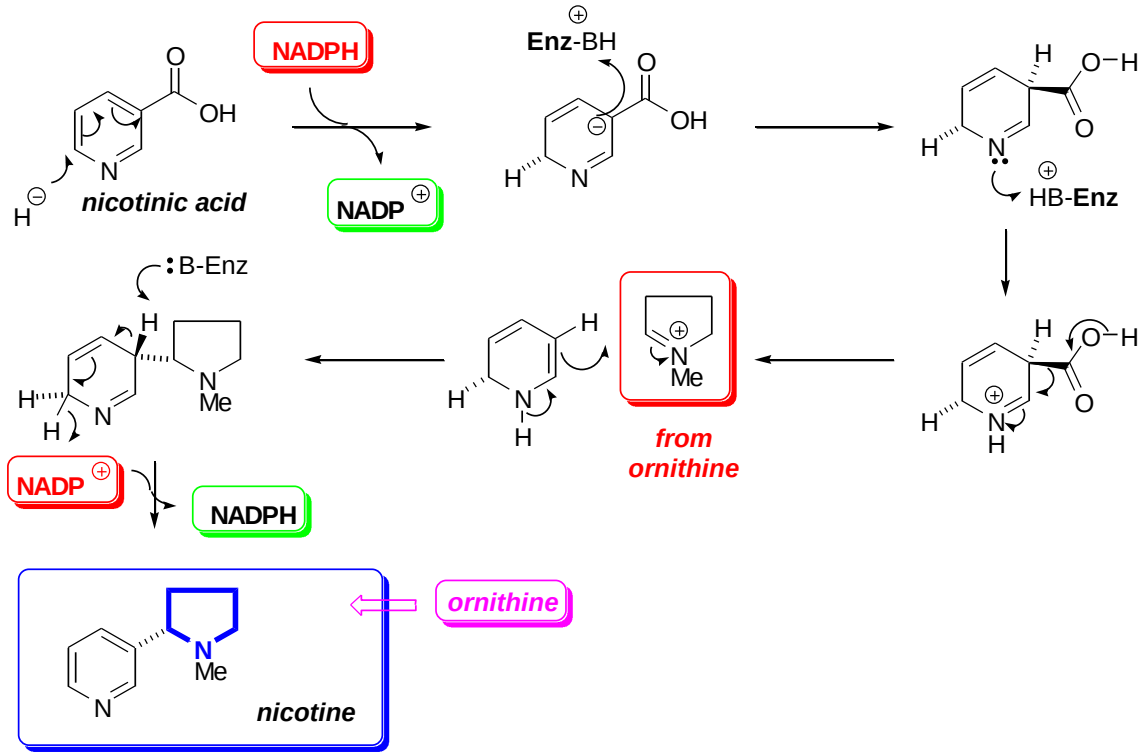


coniine

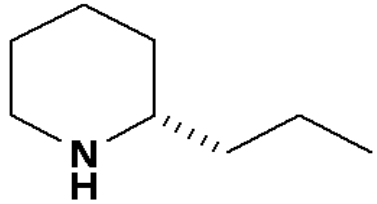
pseudopelletierine

Pyridine/Pyrrolidine Alkaloid – *Nicotine*

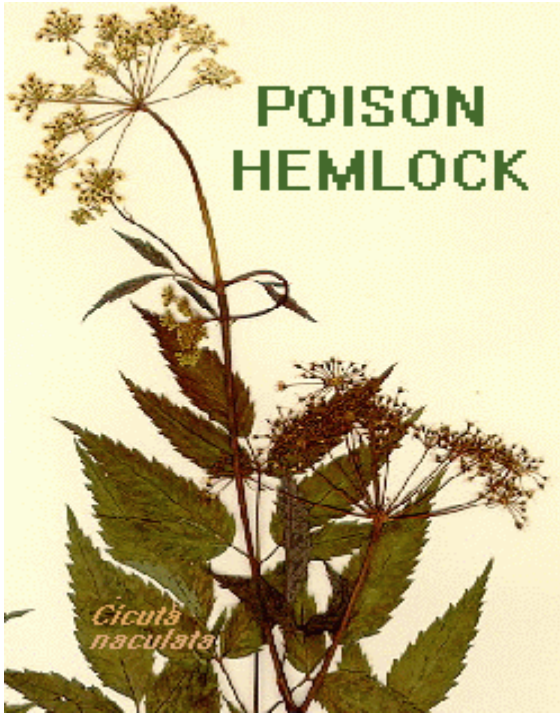
- ***Nicotine***: constituent of dried leaves of the tobacco plant (*Nicotiana tabacum*)
  - salts also sold as insecticides
  - origin of ***nicotinic acid*** component: *plants* – from aspartic acid; *animals* – from tryptophan



Hemlock Alkaloids



Coniine

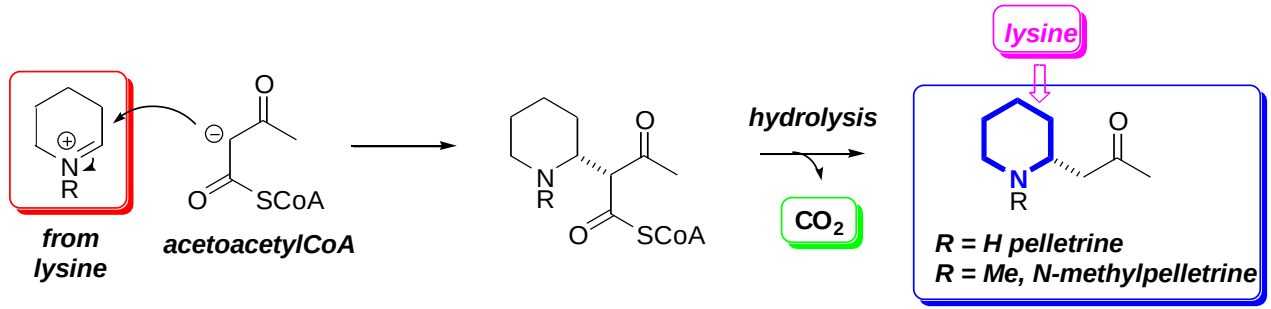


Socrates drinking poison hemlock, 399 B.C.

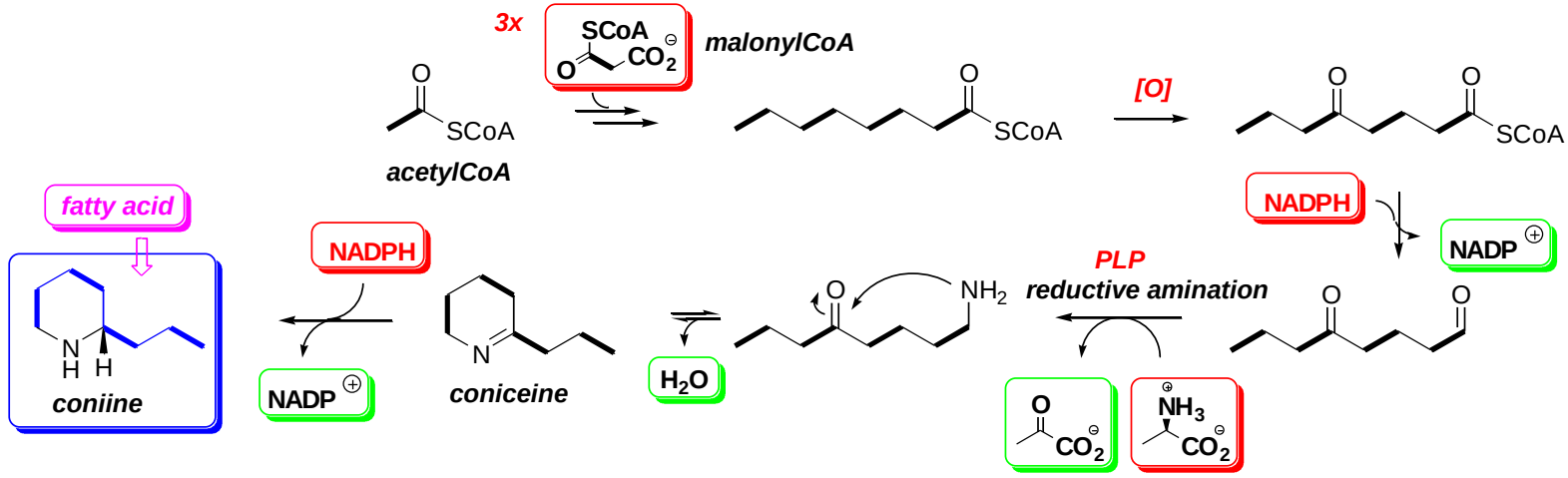
"The Death of Socrates" by Jacques-Louis David (1787)

Piperidine Alkaloids – *Pelletierine & Coniine*

- ***Pelletierine***:

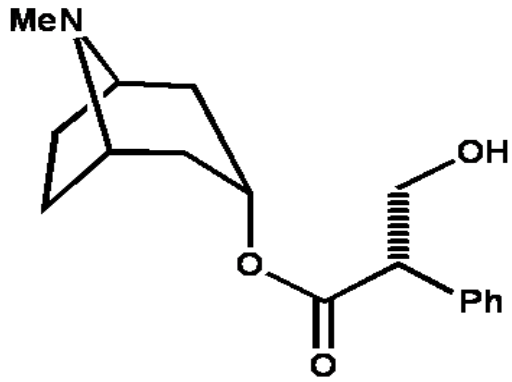


- ***Coniine***:
  - in 399 BC Socrates was sentenced to death for impiety and executed by being forced to drink a potion made from poison hemlock. The toxic component in hemlock is coniine. Although by analogy with the above pathway, biosynthesis from lysine might be suspected, it is in fact of ***fatty acid*** origin



Tropane Alkaloids

Atropa belladonna
Deadly nightshade

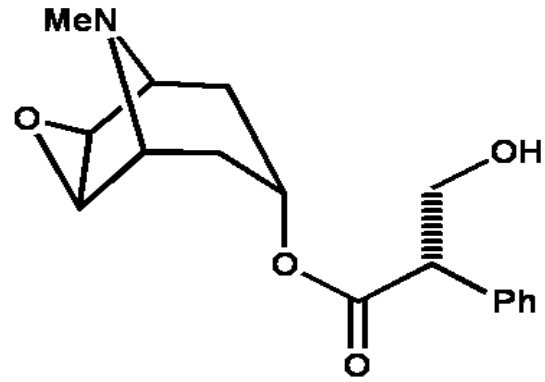


(±)-atropine
[(-)-hyoscyamine]

Hyoscyamus niger
Henbane

Tropane Alkaloids

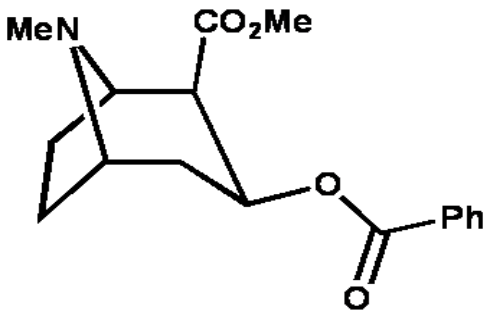
Datura stramonium
Thorn apple



scopolamine (hyoscine)

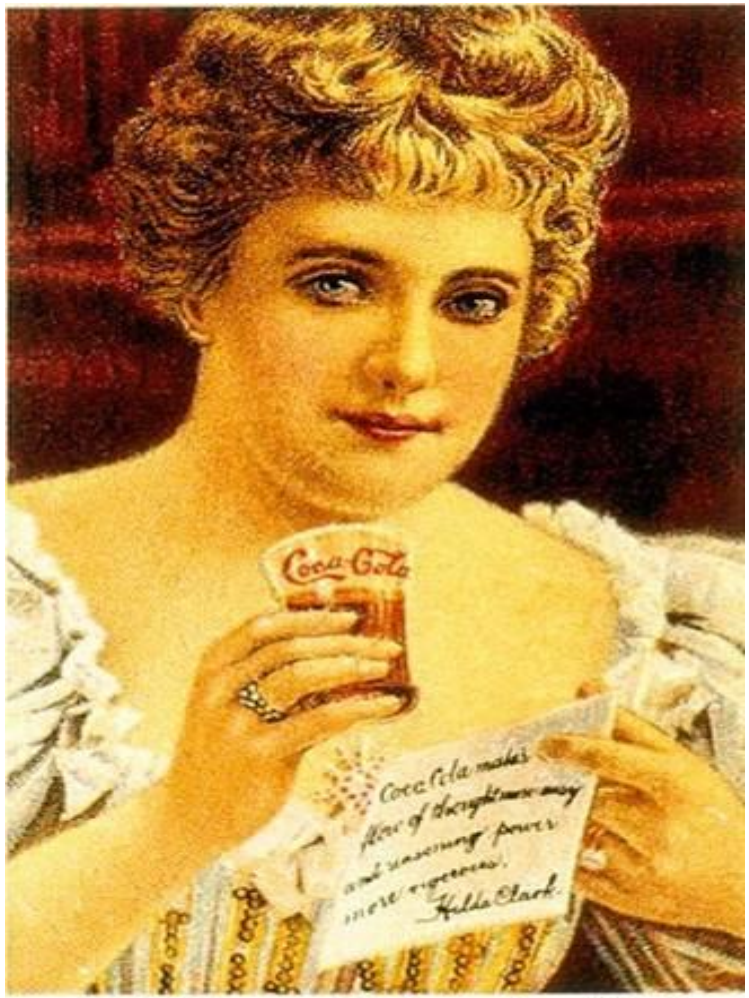
Tropane Alkaloids

Erythroxylum coca



cocaine

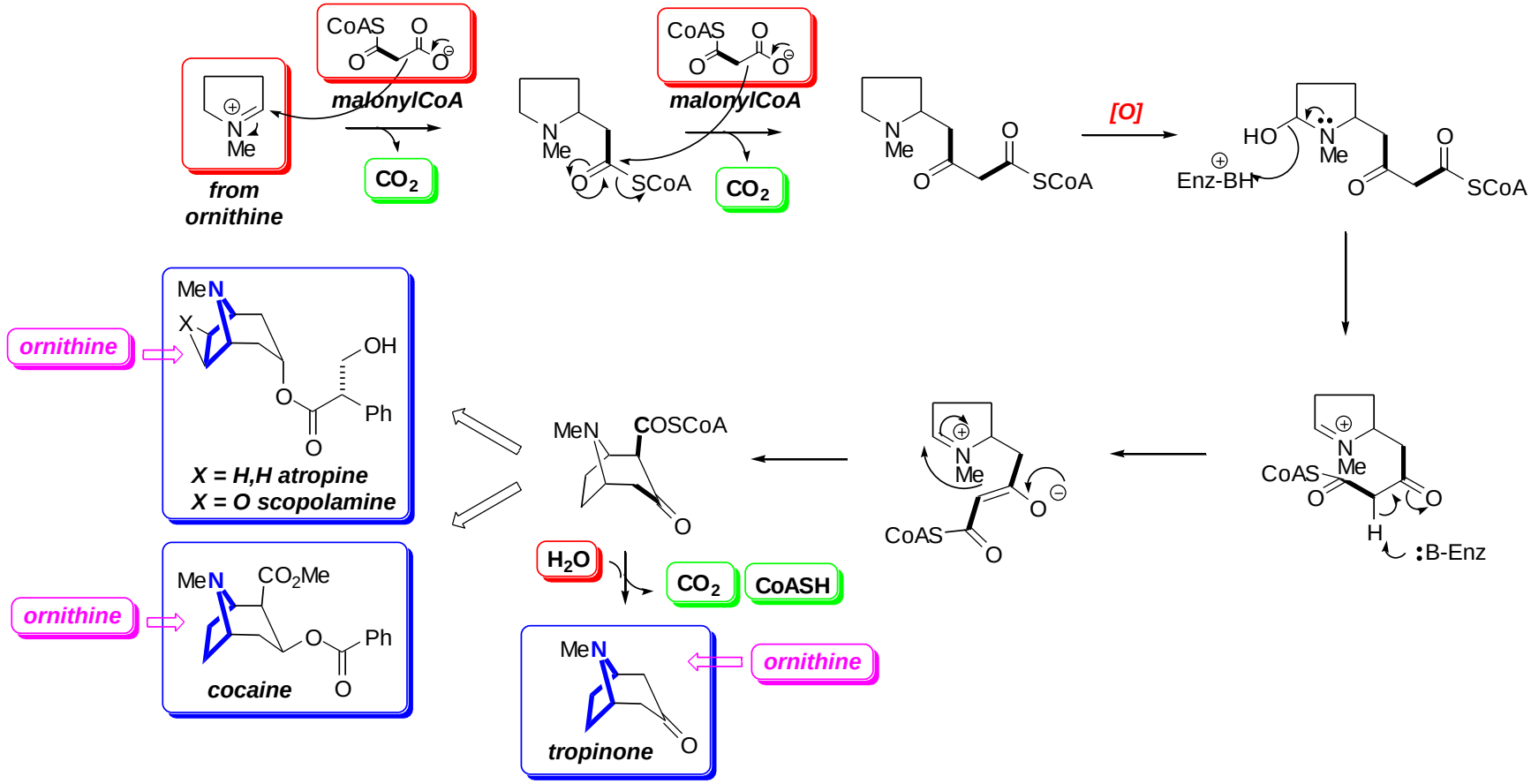
Tropane Alkaloids



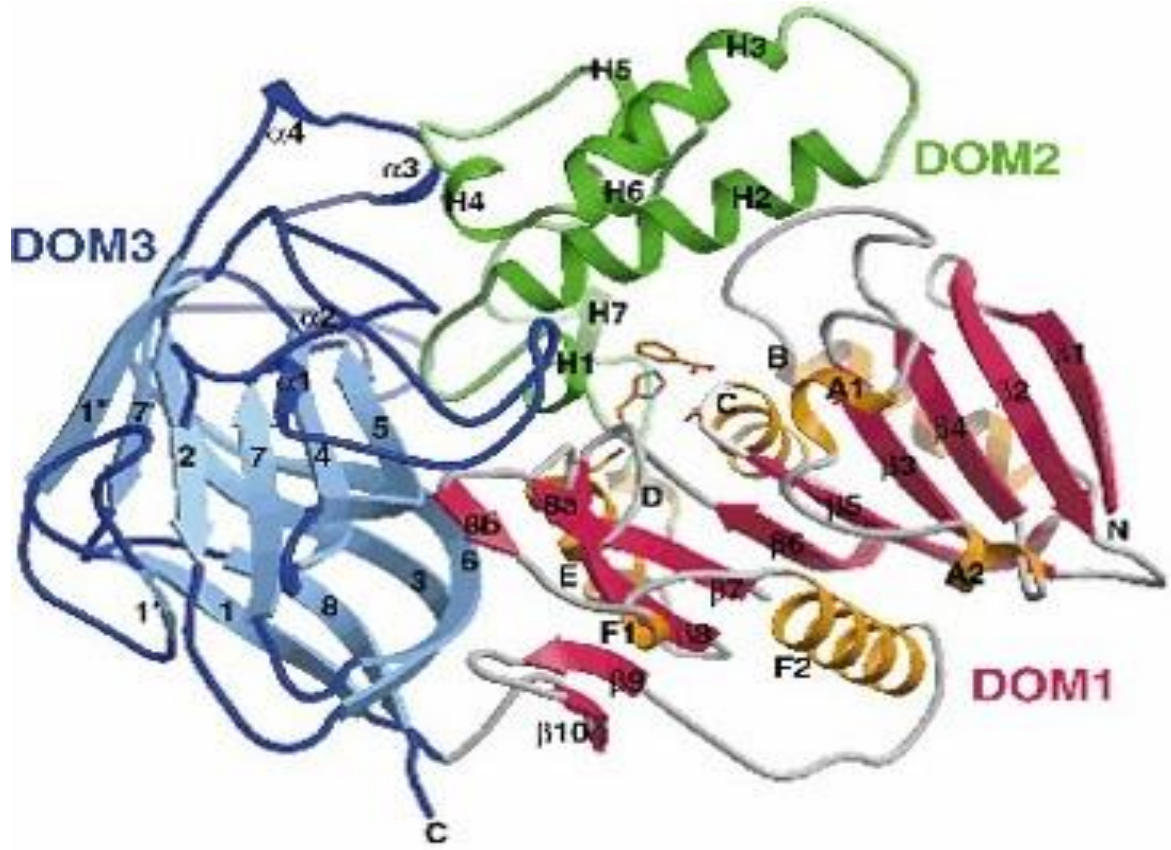
Did Coca-Cola get a kick from cocaine before 1903?

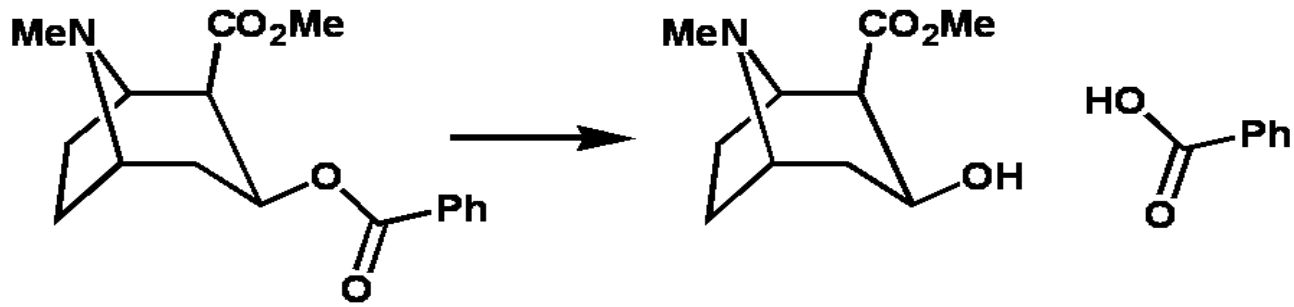
Tropane Alkaloids – *Atropine, Scopolamine & Cocaine*

- ***Atropine, scopolamine, cocaine*** & ***tropinone*** - anasthetics
  - ***Atropine*** (hyoscyamine) from Deadly nightshade (*Atropa belladonna*) – used in eye surgery (dilatory)
  - ***Scopolamine*** (hyoscine) from Thorn apple (*Datura stramonium*) – used as a surgery 'pre-med'
  - ***Cocaine*** from coca plant (*Erythroxylon coca*) – local anasthetic for ENT operations (also an hallucinogen)



Cocaine Esterase

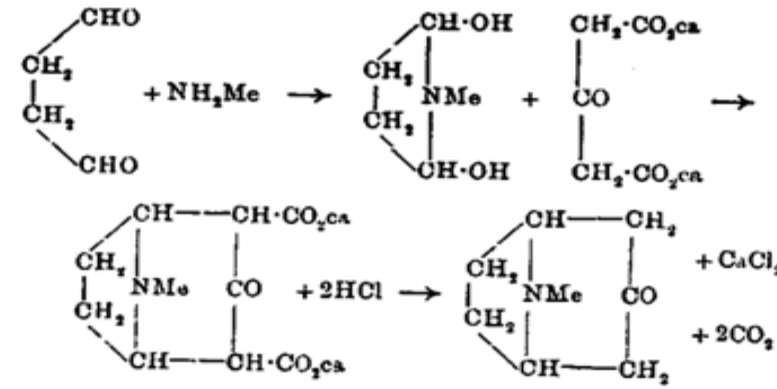




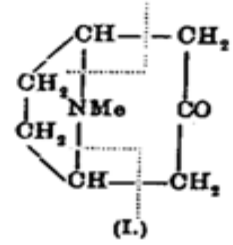
Sir Robert Robinson

JOURNAL
OF
THE CHEMICAL SOCIETY.

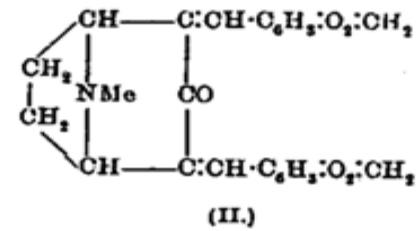
1917. Vol. CXI.



Nevertheless, an inspection of the formula of tropinone (I) discloses a degree of symmetry and an architecture which justify the hope that the base may ultimately be obtained in good yield as the product of some simple reaction and from accessible materials. By imaginary hydrolysis at the points indicated by the dotted lines, the substance may be resolved into succindialdehyde, methylamine, and acetone, and this observation suggested a line of attack of the problem which has resulted in a direct synthesis.



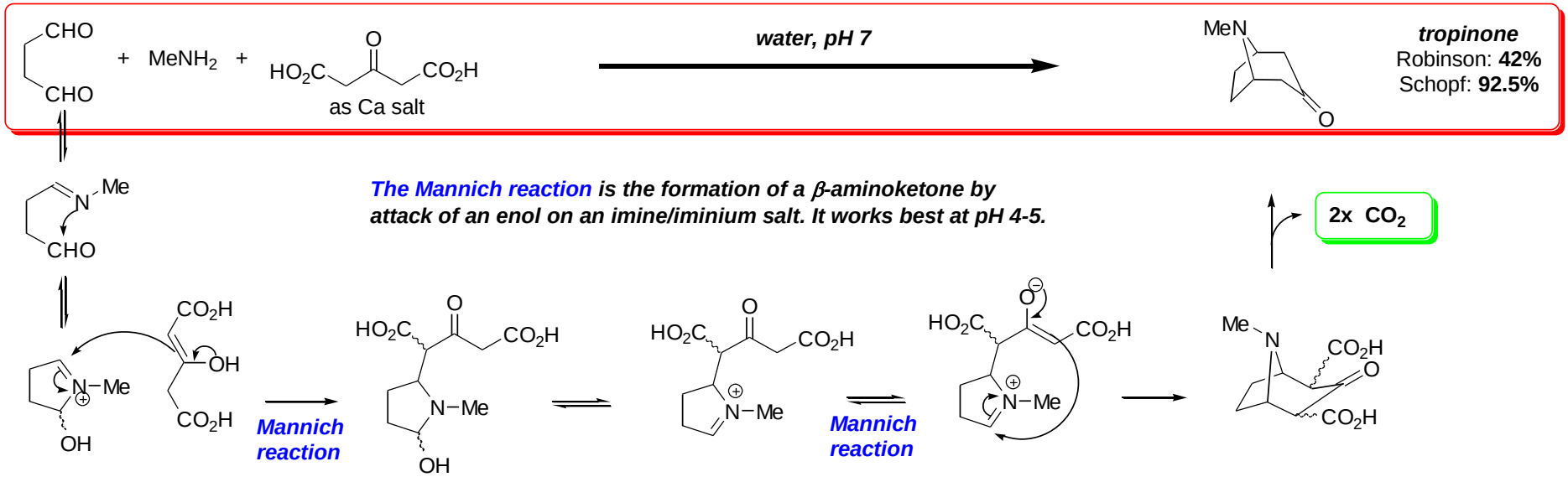
(I.)



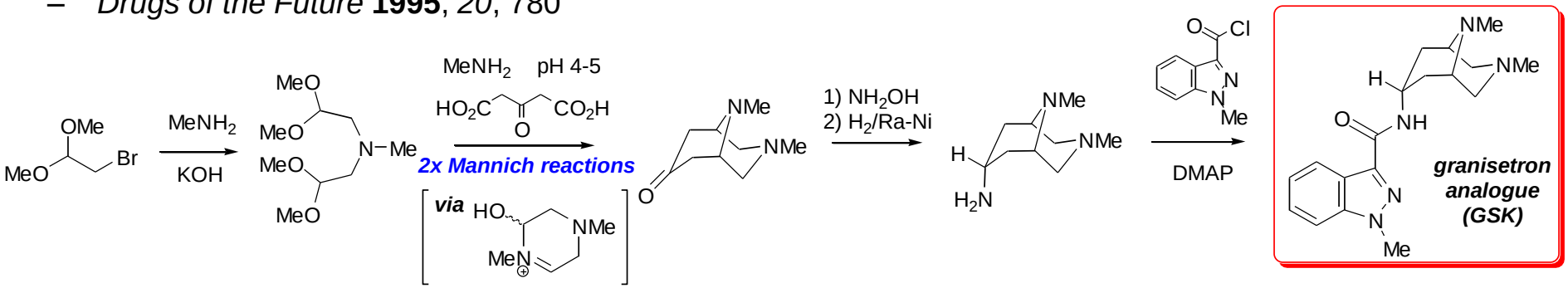
(II.)

Robinson's Biomimetic Synthesis of Tropinone

- ***Classic biomimetic laboratory synthesis – 1917!***
  - Robinson *J. Chem. Soc.* **1917**, *111*, 762 (**DOI**); Schopf & Lehmann *Justus Liebigs Ann. Chem.* **1935**, *518*, 1



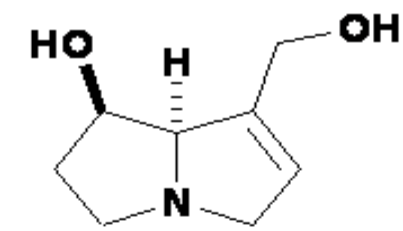
- ***...and a more recent variant:***
  - Synthesis of a 5-HT_3 receptor antagonists indicated as ***anti-emetics for cancer chemotherapy***
  - *Drugs of the Future* **1995**, *20*, 780



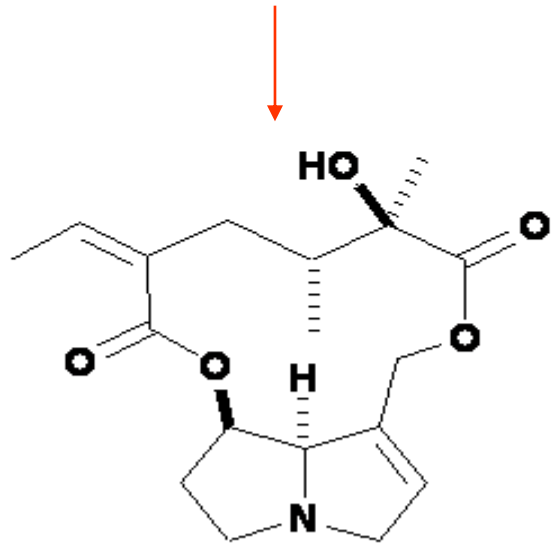
Pyrrolizidine Alkaloids

Groundsel

Ragwort



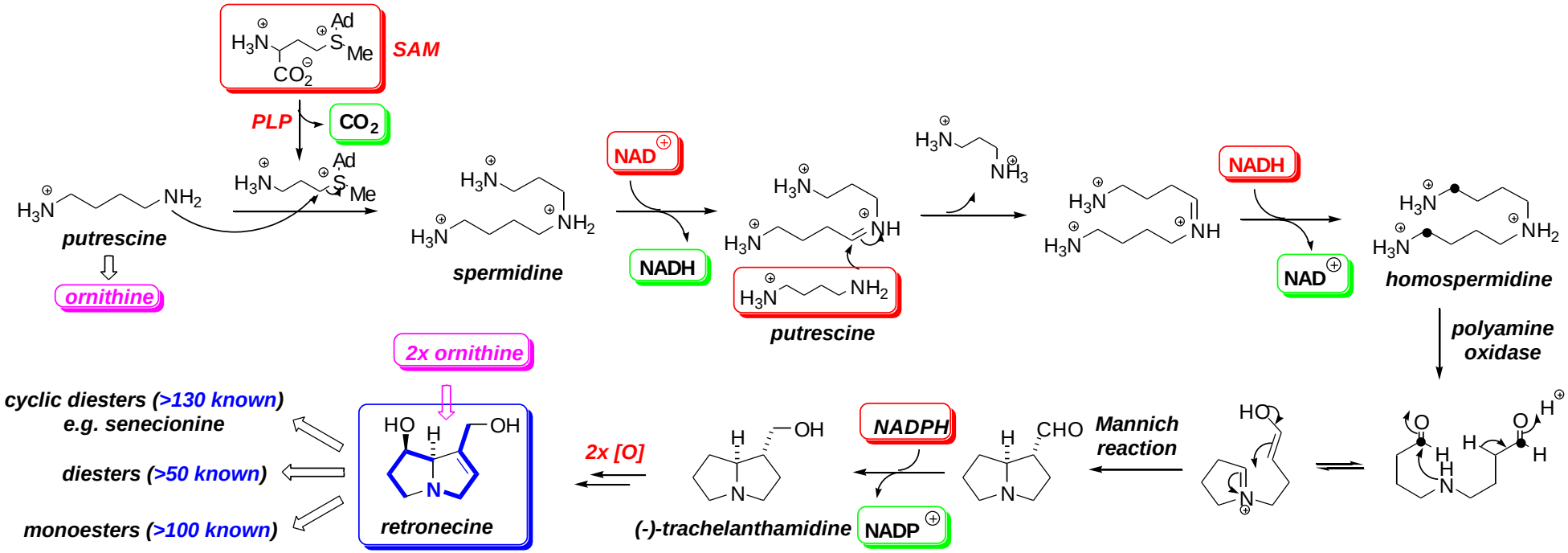
Retronecine



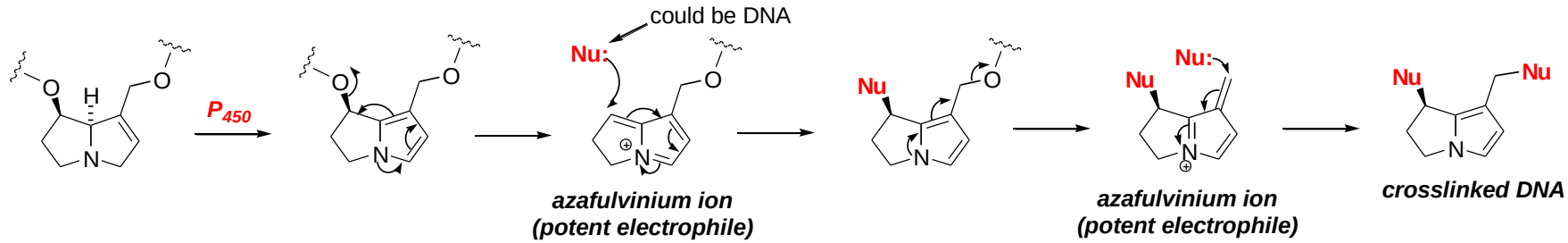
Senecionine

Pyrrolizidine Alkaloids – *Retronecine & Senecionine*

- ***Retronecine & Senecionine***:
  - biosynthesised in the roots of *senecio* plants then transported to the shoots, leaves and flowers for further processing: hydroxylations, epoxidations, *O*-acetylations *etc.*

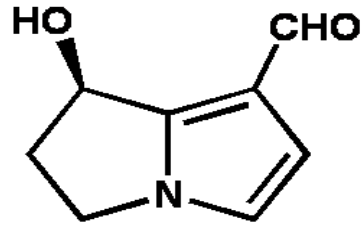


  - toxic to humans, cows & rats due to oxidation to give potent alkylating species by P_{450} enzymes in the liver:



Pyrrolizidine Alkaloids

danaidone



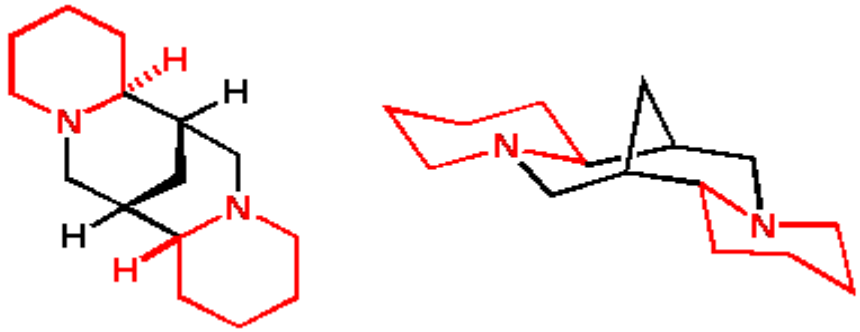
danaidal

Monarch Butterfly

For the discovery of pyrrolizidine butterfly pheromones:
Meinwald, *J. Org. Chem.* **2005**; *70,* 4903 (**DOI**)

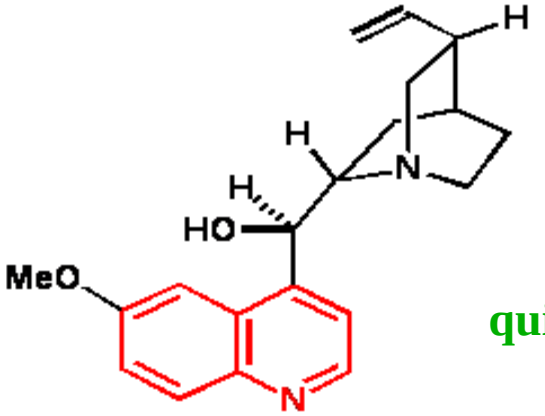
Quinolizidine & Quinoline Alkaloids

Lupin Alkaloids



sparteine

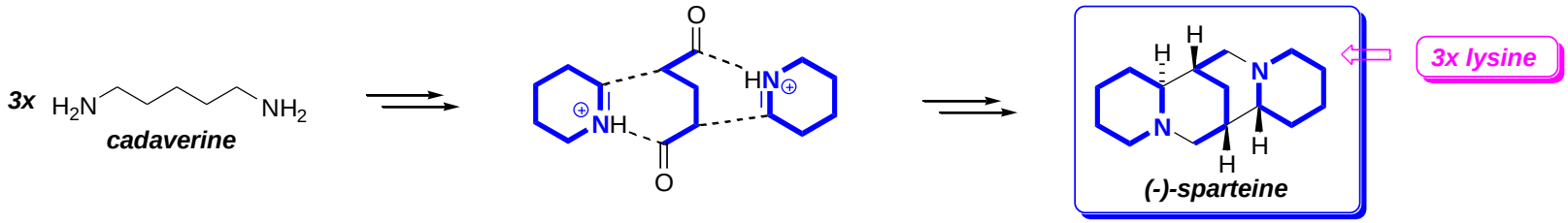
Cinchona Alkaloids



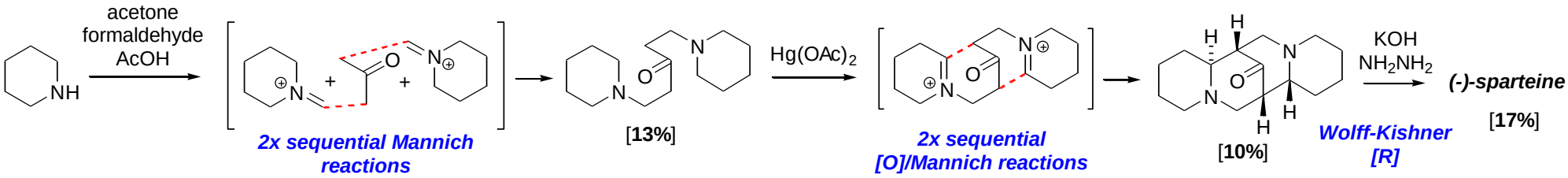
quinine

Quinolizidine Alkaloids – *Sparteine*

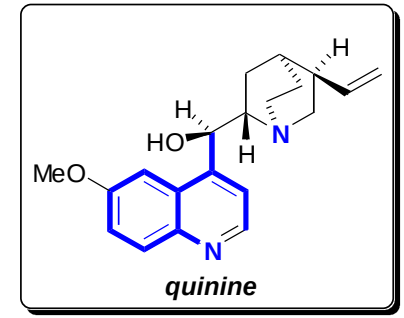
- ***Lupin alkaloids:*** (genus *Lupinus*) biogenesis *cf.* indolizidines but from ***lysine*** *via* ***cadaverine***
  - details not known



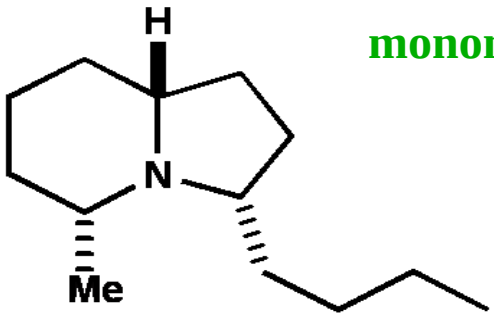
- ***Biomimetic synthesis***
  - van Tamelen & Foltz *J. Am. Chem. Soc.* **1960**, *82*, 2400 (DOI) & *ibid.* **1969**, *91*, 7372 (DOI)



- ***Cinchona alkaloids***: *e.g.* quinine (anti-malarial)
  - these are *NOT* lysine derived.
  - They are ***tryptophan***/***mevalonate*** (isoprenoid) derived alkaloids (see later)



Indolizidine Alkaloids

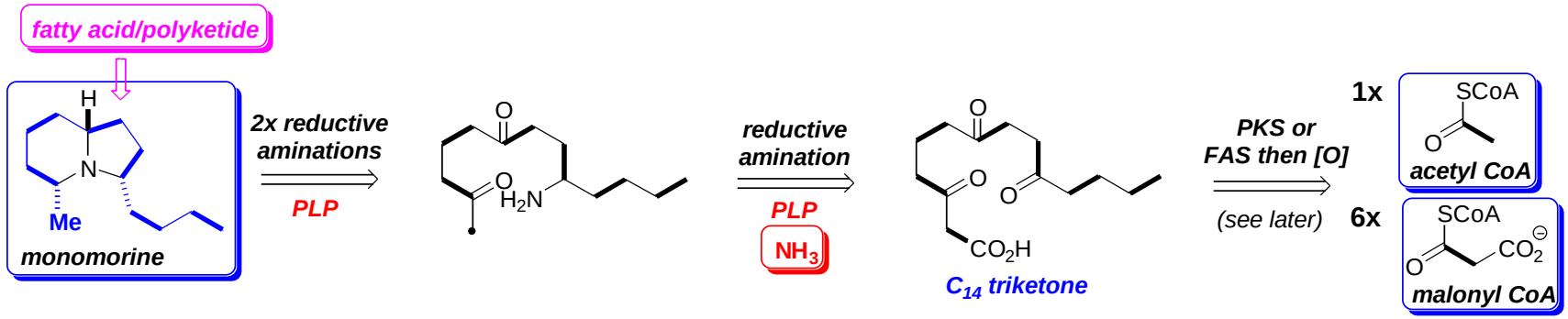


monomorine

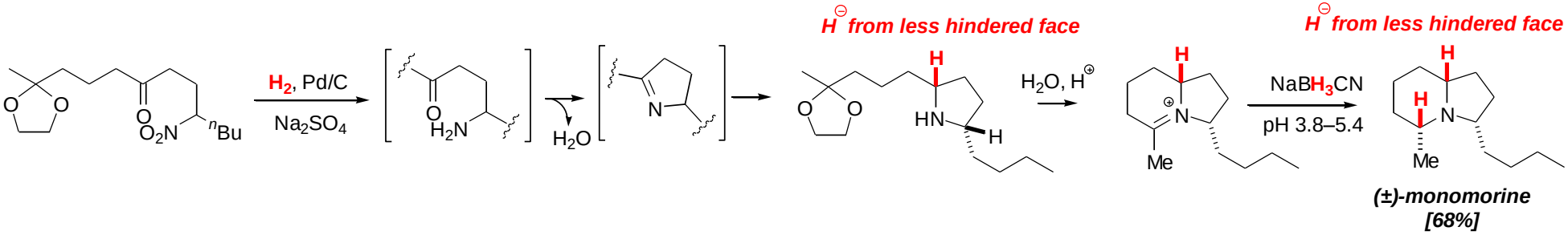
Trail pheromone of the pharoah ant
Monomorium pharaonis

Indolizidine Alkaloid – *Monomorine*

- ***Indolizidine alkaloids:*** *e.g.* monomorine (trail pheromone of the Pharaoh ant)
  - These are *NOT* lysine/ornithine derived. They are ***polyketide/fatty acid*** derived alkaloids
  - ***putative biogenesis:***



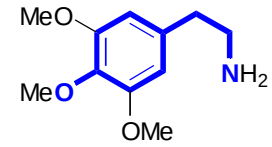
  - ***biomimetic synthesis:***
    - Stevens *et al. J. Chem. Soc., Chem. Commun.* **1982**, 102 (DOI)



Phenyalanine & Tyrosine Derived Alkaloids

- ***Alkaloids (generally) containing an ArC_2N subunit (± ArC_2/ArC_1):***
  - Skeleta built up by ***reductive amination***, ***decarboxylation***, ***oxidation*** (*e.g.* ***phenolic coupling***, ***hydroxylation***)
  - ***Major classes:***

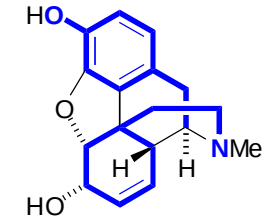
  - ***monocyclic alkaloids*** [phenethylamines (*e.g.* mescaline)]



mescaline

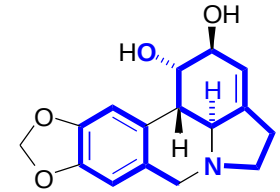
CASE STUDY
'elucidation of a biosynthetic pathway'

  - ***benzylisoquinolines*** [opium alkaloids (*e.g.* papaverine, morphine);
  - aporphine alkaloids; erythrina alkaloids]

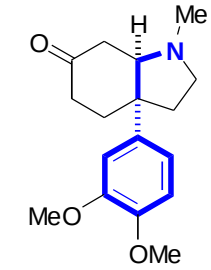


morphine

  - ***amaryllidaceae alkaloids*** (*e.g.* lycorine, galanthamine)

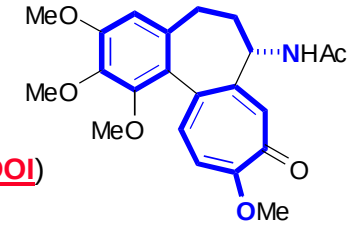


lycorine



mesembrine

  - ***mesembrine alkaloids*** (*e.g.* mesembrine)
  - Jeffs *et al. Phytochem*. **1978**, *17*, 719 (DOI)



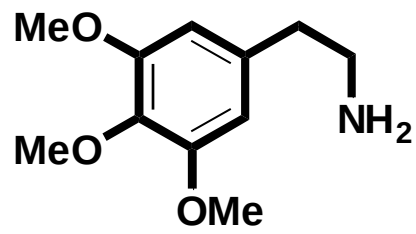
colchicine

  - ***colchicine***
  - Battersby *et al. J. Chem. Soc., Perkin Trans. 1* **1998**, 3003 (DOI)

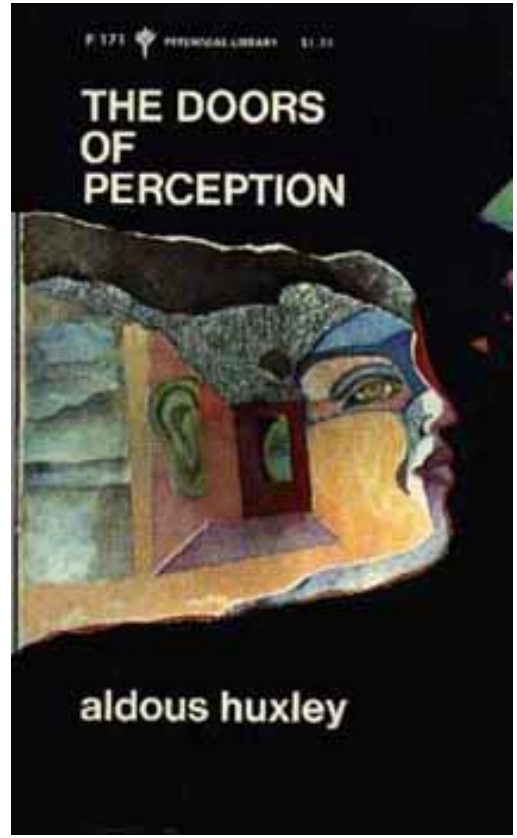
Mescaline

San Pedro

Trichocereus pachanoi



mescaline

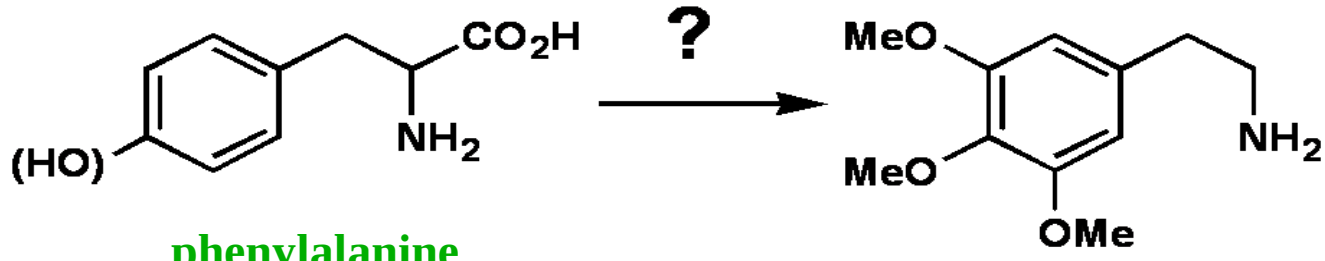


'If we could sniff or swallow something that would, for five or six hours each day, abolish our solitude as individuals, atone us with our fellows in a glowing exaltation of affection and make life in all its aspects seem not only worth living, but divinely beautiful and significant, and if this heavenly, world-transfiguring drug were of such a kind that we could wake up next morning with a clear head and an undamaged constitution-then, it seems to me, all our problems (and not merely the one small problem of discovering a novel pleasure) would be wholly solved and earth would become paradise'

Aldous Huxley

Phenylalanine or Tyrosine? - *Isotopic Labelling*

Peyote
Lophophora Williamsii

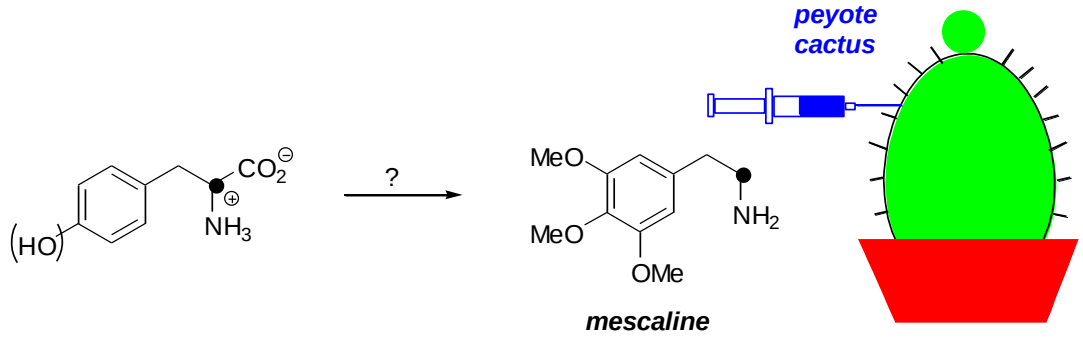


phenylalanine or tyrosine

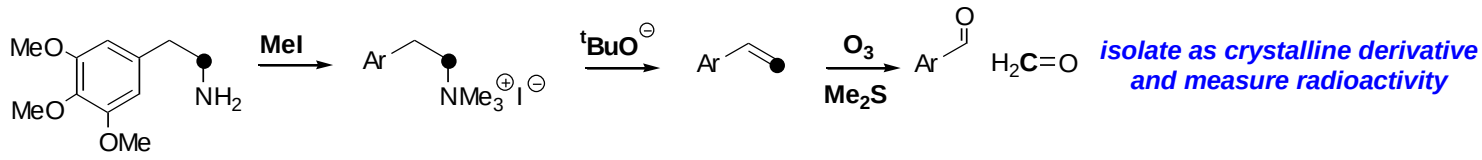
mescaline

Elucidation of a Biosynthetic Pathway – *Mescaline*

- ***Administer Labelled Precursor:***



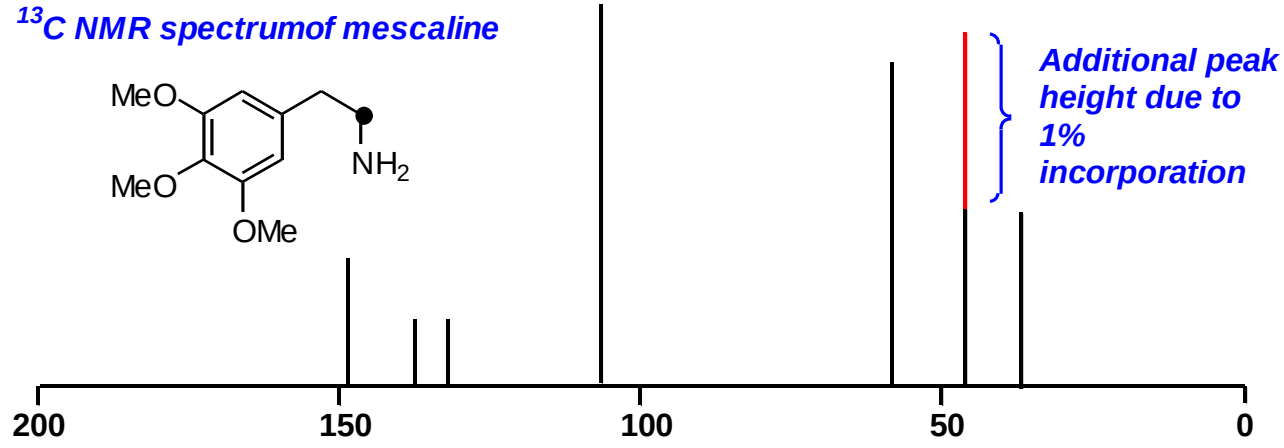
- ***Analysis of Result:***
  - ***CARBON-14 (^{14}C):***
    - ^{14}C is a β-emitter with half life of 5640 years
    - Radioactive isotopes are virtually never used at anything like 100% abundance. In fact only one molecule in a thousand or even one in a million will be labeled
    - detect presence of label by radioactivity in isolated mescaline. Position of label needs to be confirmed by degradation *e.g.*



    - ***Advantage:*** *detection of ^{14}C or ^{3}H is very sensitive and there is almost no natural abundance*
    - ***Disadvantages:*** *degradation to locate label is always long and difficult & may be impossible. Precautions needed to avoid radioactive contamination*

Elucidation of a Biosynthetic Pathway – *Mescaline*

- ***Analysis of Result:***
  - ***CARBON-13 (^{13}C):***
    - ~100% abundance usually employed (*NB.* natural abundance is ~1.1%)
    - detect presence of label by NMR. NMR spectrum needs to be assigned to confirm location of label:

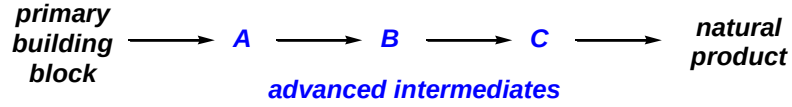


    - ***Advantage:*** *rapid determination of location of label*
    - ***Disadvantage:*** *Not very sensitive (looking for enhancement over natural abundance) - more compound needed*

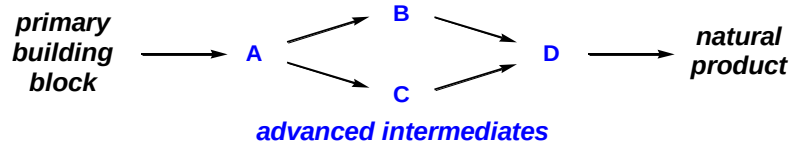
  - ***MASS SPECTROMETRY:***
    - Detection by Mass Spectrometry is also possible for any sufficiently stable isotope (^{13}C, ^{14}C *etc.*)
    - ***Advantage:*** Can be done on very small amount. Fragment ions give partial location of label
    - ***Disadvantage:*** High enrichment needed to show above the natural abundance ^{13}C peak (or incorporation of multiple isotopes to give *e.g.* M+3 peak)

Elucidation of a Biosynthetic Pathway – *Mescaline*

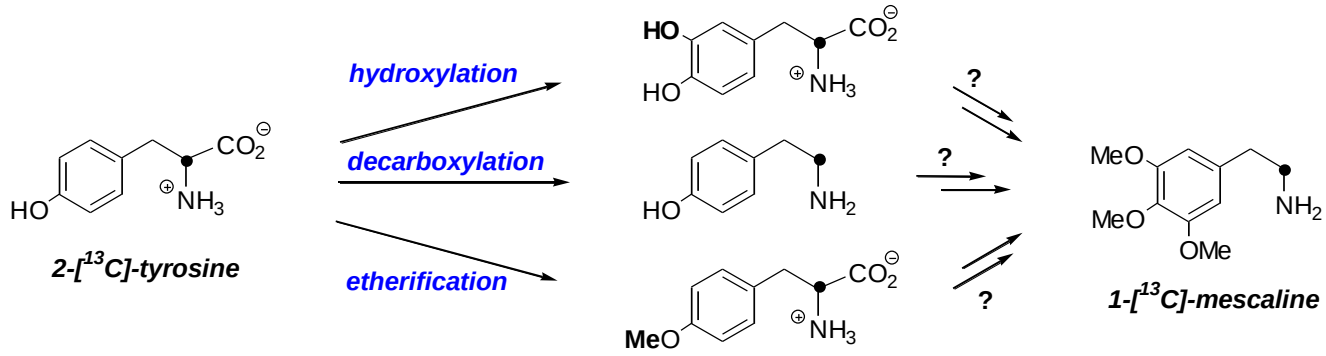
- ***Identifying intermediates:***
  - usually the biosynthesis follows a defined sequence of chemical steps with intermediates released into solution at each stage



  - less commonly alternative pathways can be followed:



  - For ***mescaline*** a number of pathways could be envisaged:

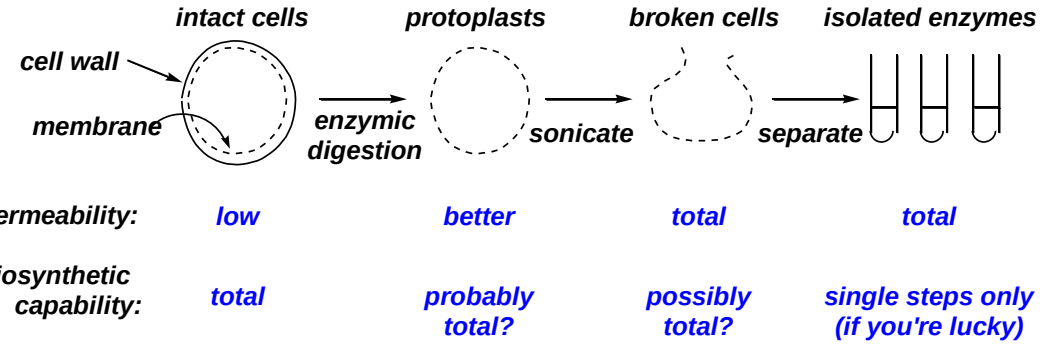


  - How do we determine the correct pathway?

Elucidation of a Biosynthetic Pathway – *Mescaline*

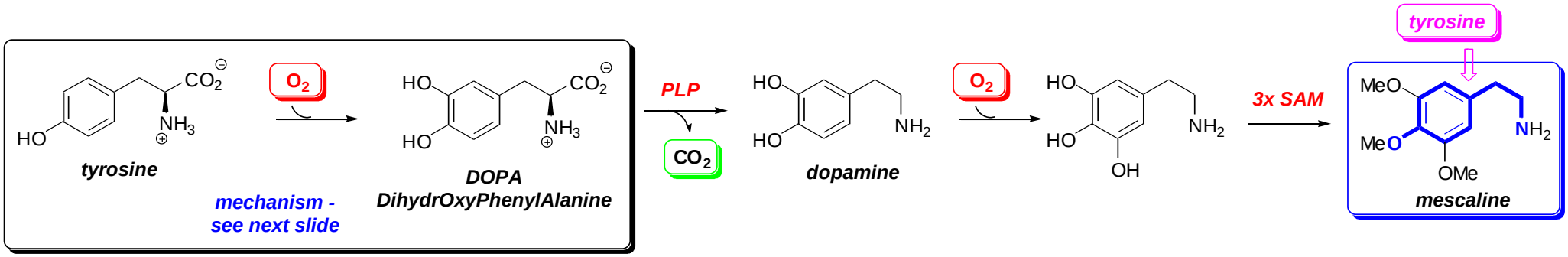
- ***Make all three in labelled form and feed them separately to the organism then isolate the natural product and see how much of the isotopic label is incorporated***
  - Incorporation of the label alone does not guarantee that the compound fed is an intermediate because:
    1. the compound may have been degraded to basic precursors such as acetate and then reincorporated
    2. the compound may fortuitously get converted to another compound which is the true intermediate
  - ***To ensure (1)*** is not happening, you have to show the label is in the expected position and would not be if degradation had occurred.
  - ***To disprove (2)*** is difficult. You need to show that the compound is in fact formed in the cell by isolating it. However, often the levels are too low for direct isolation. If this is the case, you can use dilution analysis. In this, a radioactive precursor (*e.g.* tyrosine) is fed and then after a while the organism is extracted and unlabelled putative intermediate is added to the extract. Now there is enough to allow isolation and purification. If the reisolated compound has some radioactivity then this must have been present in the organism

- ***With microorganisms feeding experiments may be done in several different ways:***

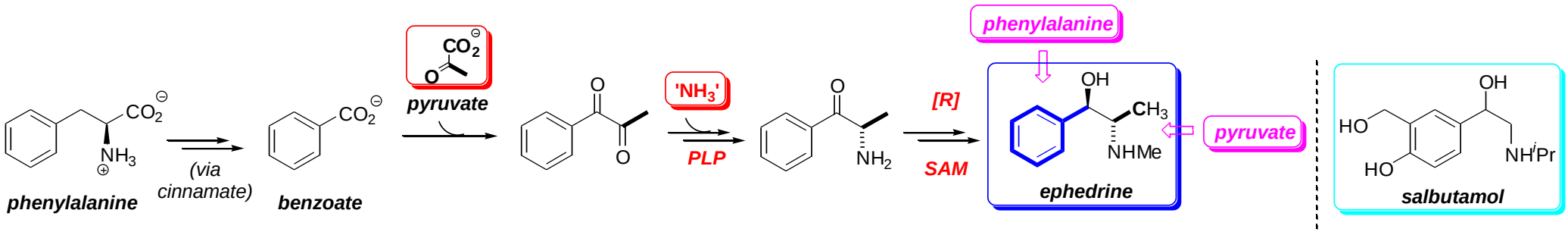


Monocyclic Alkaloids – *Mescaline & Ephedrine*

- ***Mescaline:*** psychoactive component of ***peyote cactus*** (*Lophophora williamsii*)
  - Halucinogen used in Aztec, Mayan & Inca religious ceremonies
  - ***biosynthesis from tyrosine:***

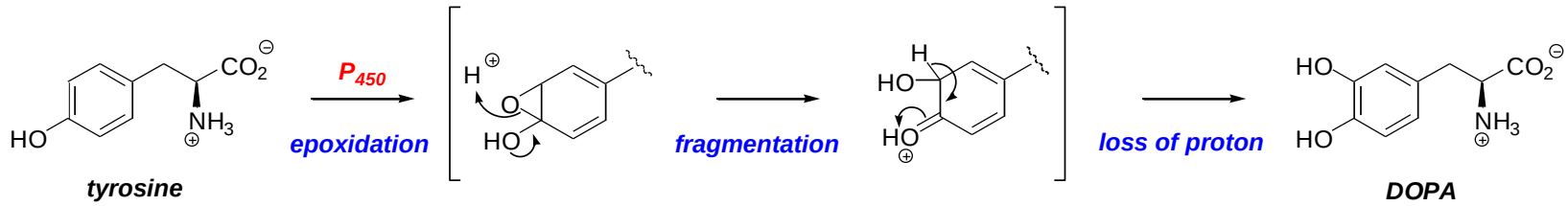


- ***Ephedrine:*** stimulant from *Ephedra* species
  - component of traditional medicines for asthma and bronchitis
  - 'inspiration' for modern bronchodilators *e.g.* salbutamol (Ventolin®)
  - ***biosynthesis from phenylalanine***, *BUT* not as directly as might have been envisaged:

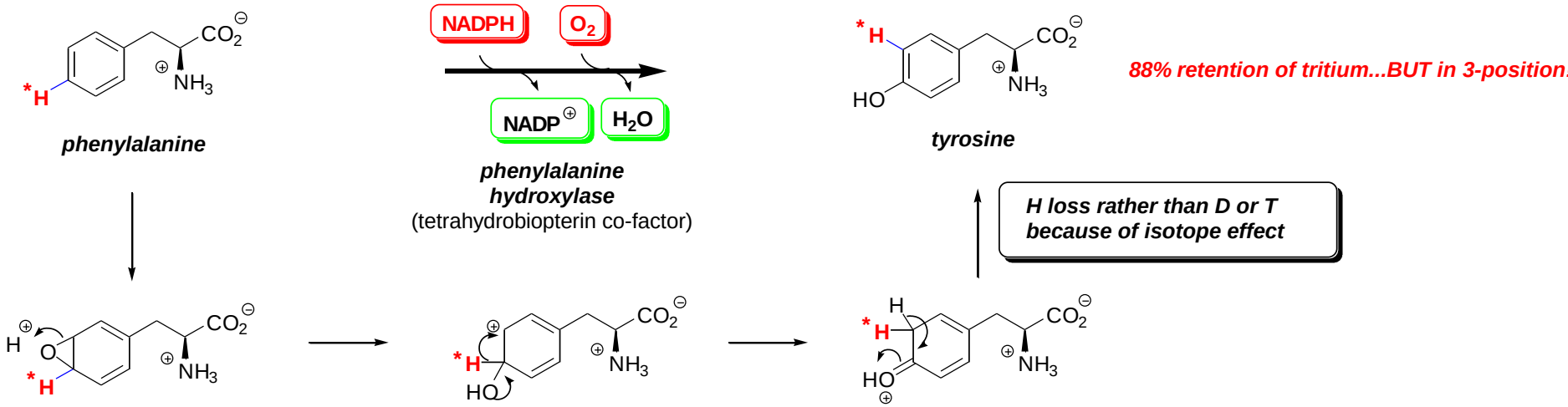


Biological Hydroxylation of Aryl Rings

- ***The basic mechanism of aromatic hydroxylation:***
  - hydroxylation can occur at any C atom carrying a H
  - the enzyme can be ***flavin/pterin-dependent*** or a ***P_{450}*** or ***non-haem iron-*** or ***Cu^{2+}-dependent***
  - *e.g.* biosynthesis of ***DOPA*** from ***tyrosine*** in ***peyote cactus*** & *Papaveraceae*:

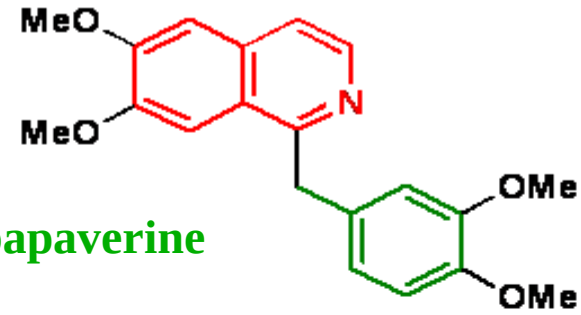


- ***If hydroxylation occurs at an 'unactivated' position - an 'NIH shift' is often observed***
  - *e.g.* biosynthesis of ***tyrosine*** from ***phenylalanine*** in barley:

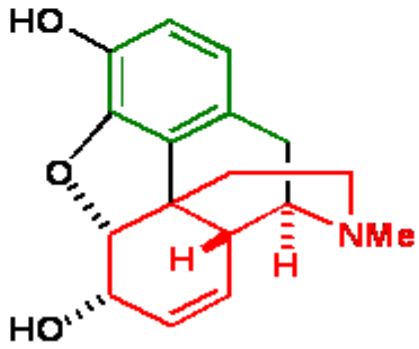


Benzylisoquinoline Opium Alkaloids

Benzylisoquinoline Alkaloids



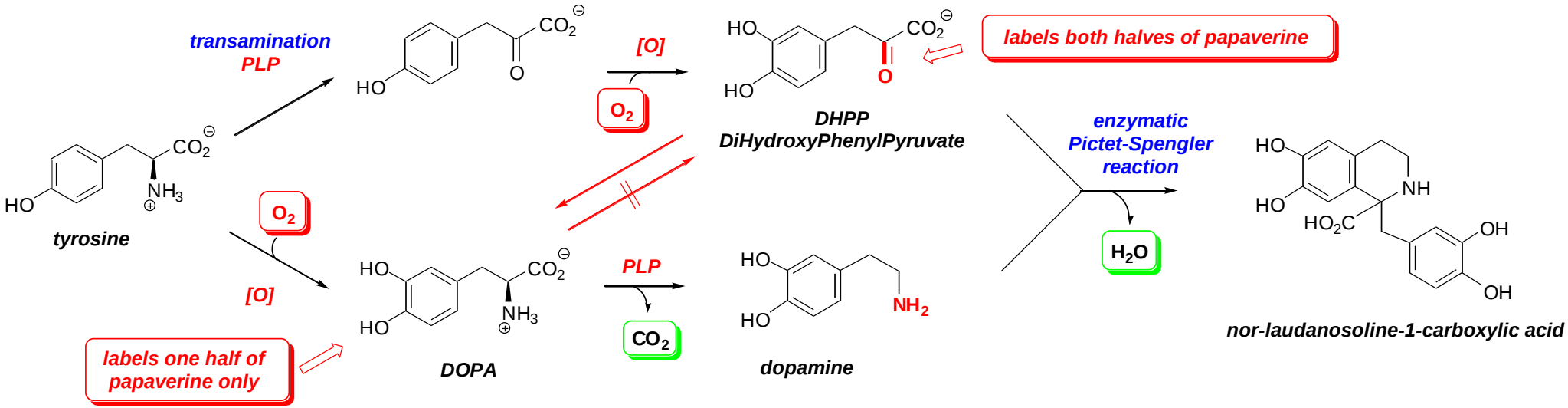
papaverine



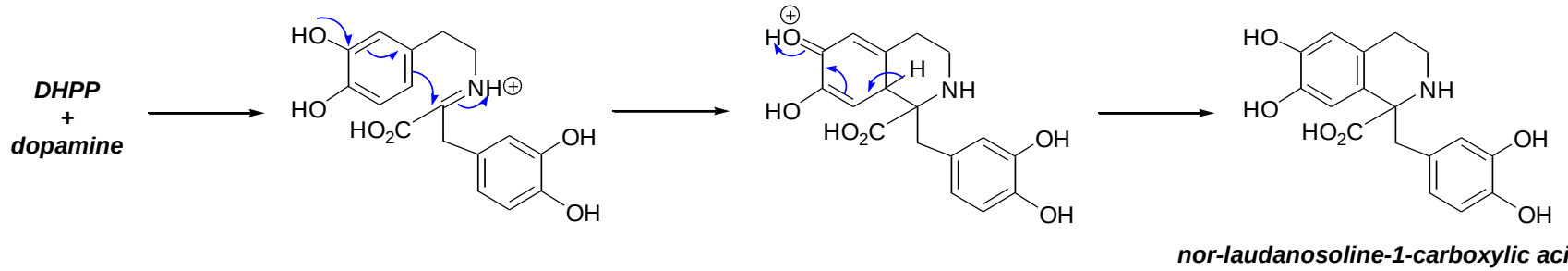
morphine

Benzylisoquinoline Alkaloids – *Ring Formation*

- ***Benzylisoquinoline alkaloids*** constitute an extremely large and varied group of alkaloids
  - many, particularly the *opium alkaloids* (*e.g.* papaverine, morphine) are ***biosynthesised*** from two molecules of tyrosine *via* ***nor-laudanosoline***:

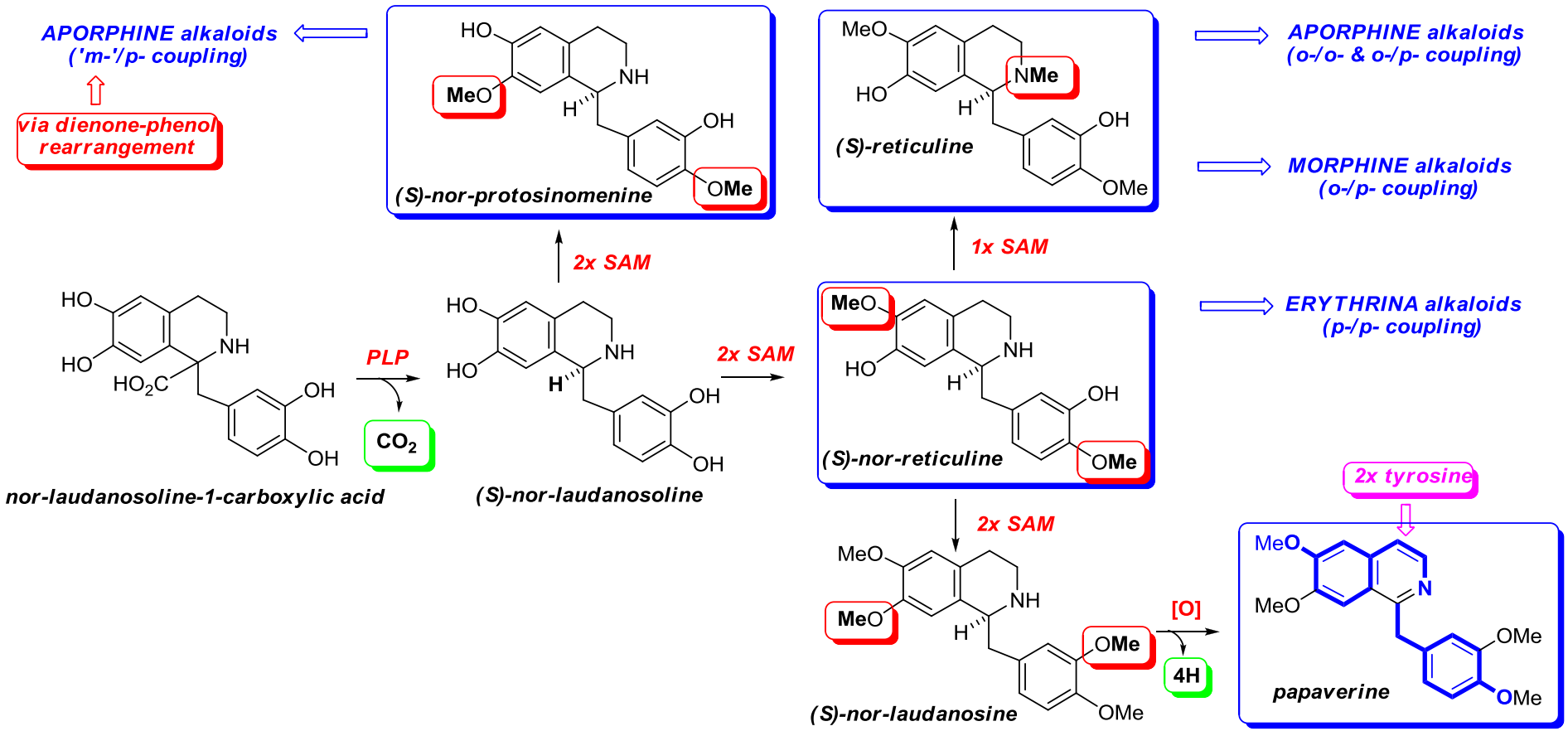


  - ***Mechanism of Pictet Spengler reaction:***



Benzylisoquinoline Alkaloids - *Papaverine*

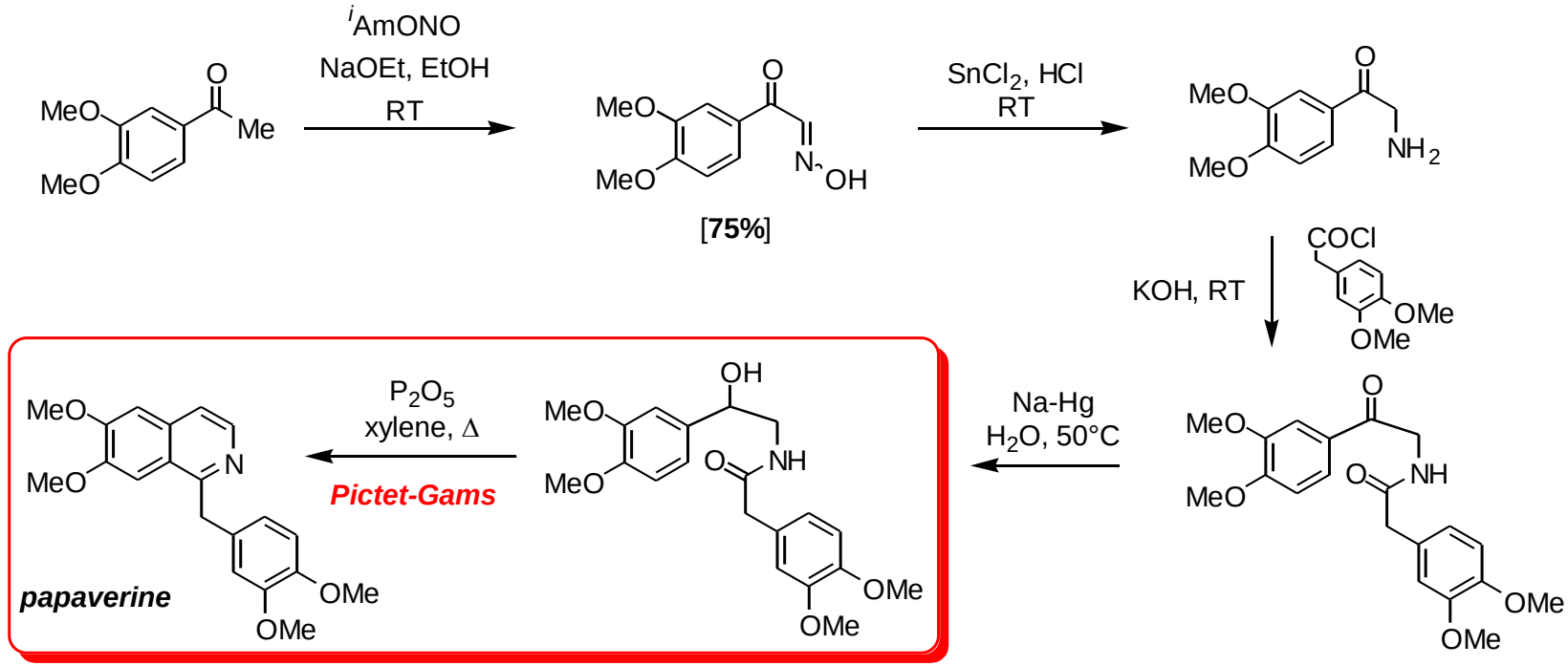
- ***Papaverine:*** analgesic contsituent of the **opium poppy** (*Papaver somniferum):*
  - ***biosynthesis:***



  - *NB.* The prefix ***nor*** means ***without a methyl group***. Laudanosoline, reticuline and laudanosine are the *N*-methyl compounds

Biomimetic Synthesis of Papaverine

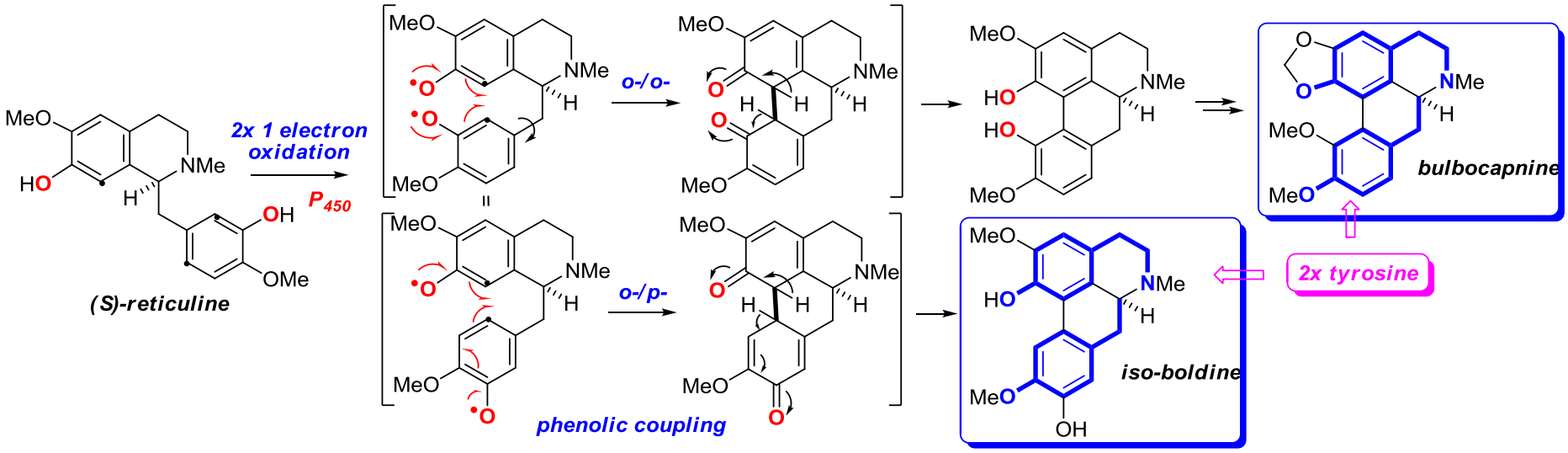
- The ***Pictet-Gams*** ring closure was developed for the synthesis of papaverine. The reaction is essentially a ***Bischler-Napieralski*** reaction which, by virtue of having a leaving group (OH) pre-installed at the benzylic position, proceeds directly to the isoquinoline (*cf.* dihydroisoquinoline)
  - Pictet & Gams *Chem. Ber.* **1909**, 42, 2943



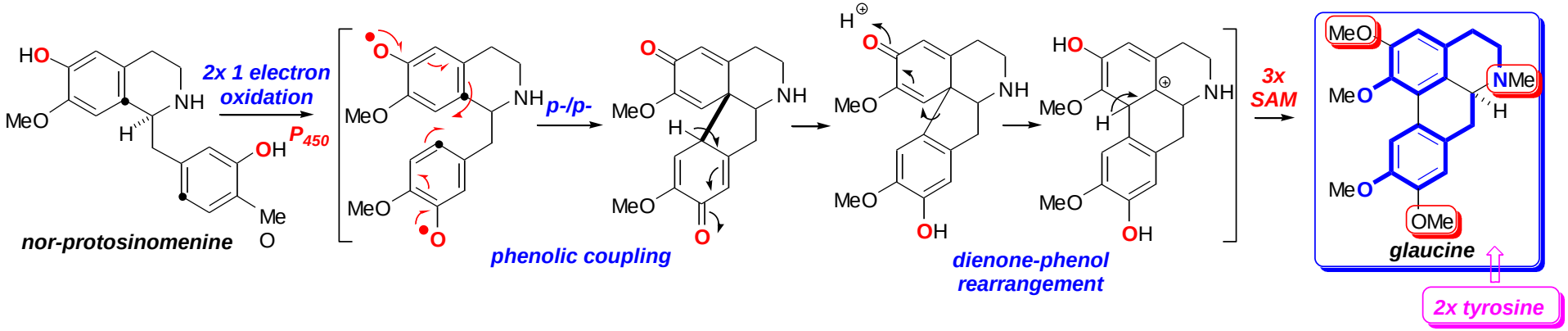
  - *NB.* A ***Pictet-Spengler*** ring closure gives a tetrahydroisoquinoline directly

Oxidative Phenolic Coupling – *Aporphines*

- ***Bulbocapnine*** & ***iso-boldine:*** *o-/o-* & *o-/p-* oxidative phenolic coupling of ***reticuline****:*

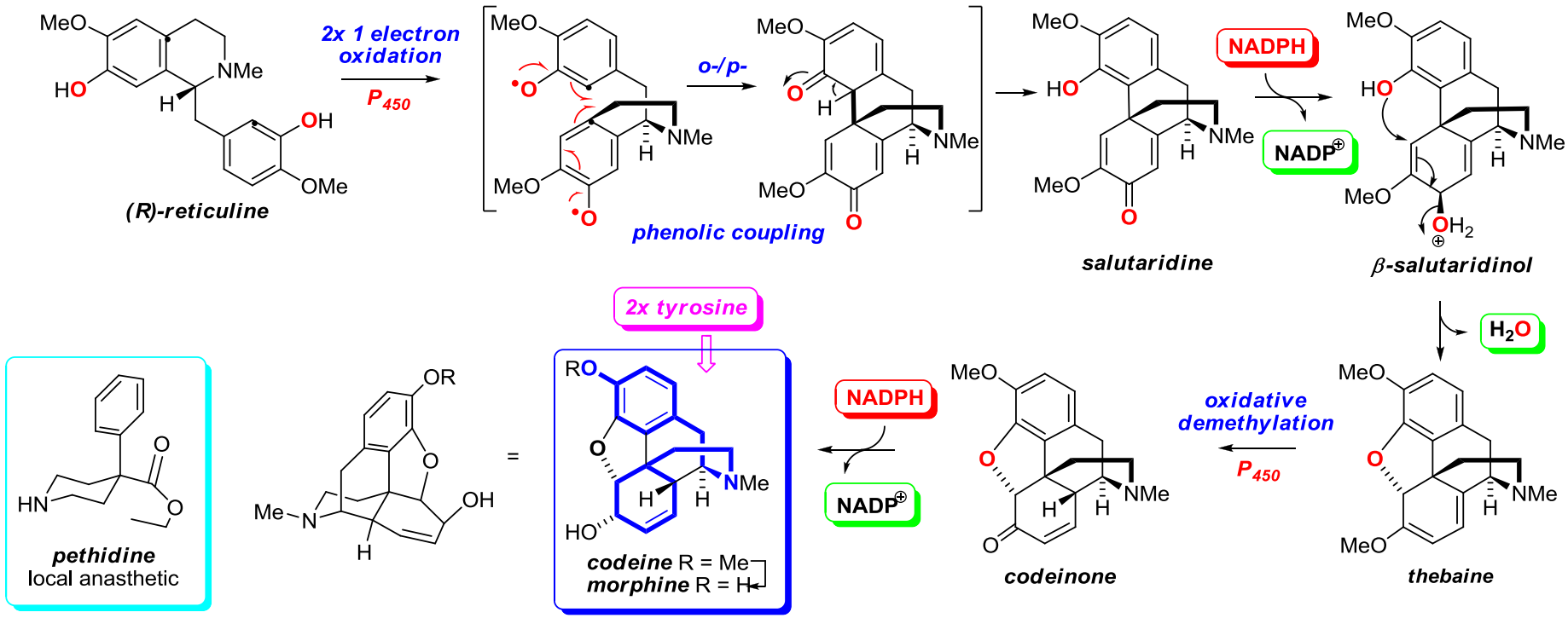


- ***Glaucine:*** *'m-'/o-* oxidative coupling of ***nor-protosinomenine*** *via* ***dienone-phenol*** rearrangement*:*



Oxidative Phenolic Coupling – *Morphine*

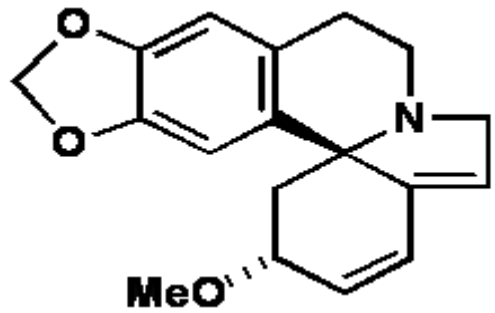
- ***Morphine:*** analgesic & sedative contsituent of the **opium poppy** (*Papaver somniferum):*
  - ***biosynthesis:*** *o-/p-* oxidative phenolic coupling of ***reticuline:***



  - Morphine acts by activating the ***opiate receptors*** in the brain (IC_{50} 3 nM)
  - The natural ligands for these receptors are peptides: *e.g.* Leu-enkephalin (Tyr–Gly–Gly–Phe–Leu) (IC_{50} 12 nM)

Erythrina Alkaloids

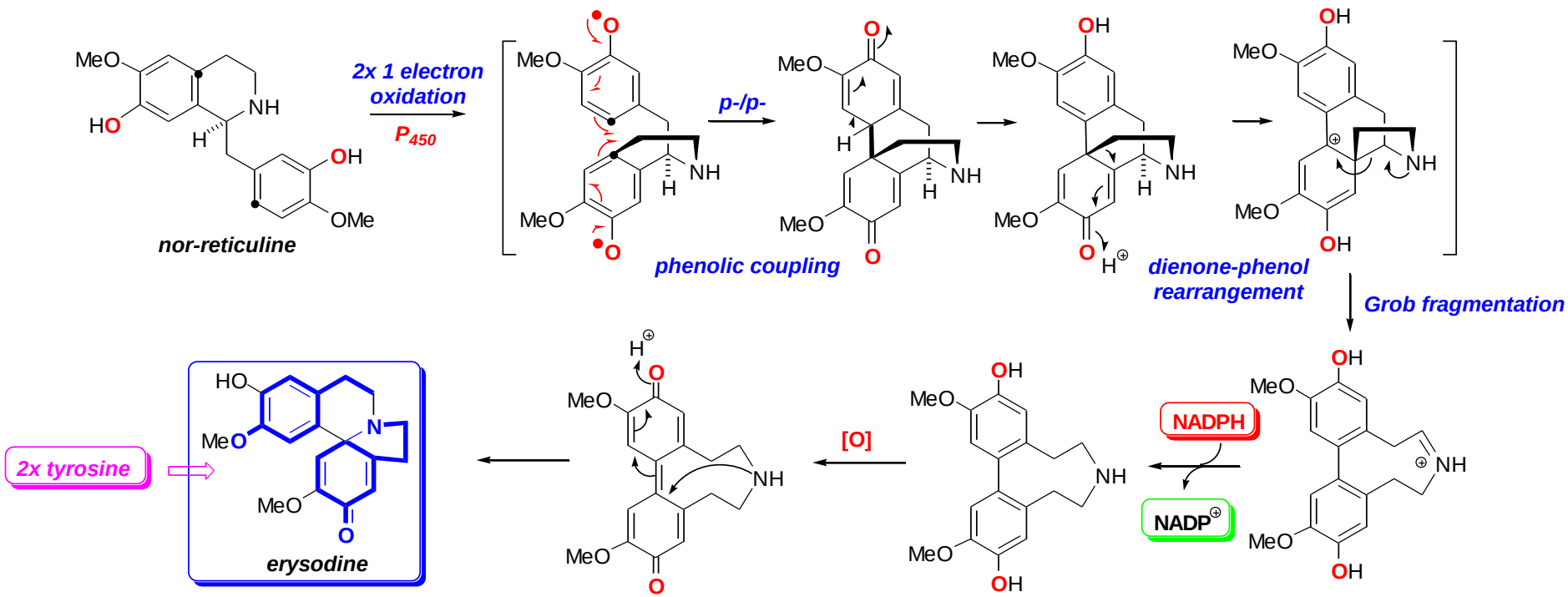
Erythrina crista-galli



erythraline

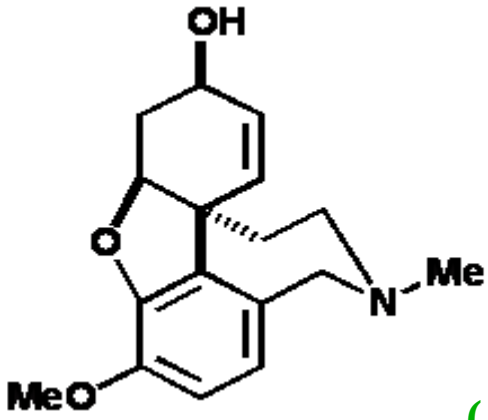
Oxidative Phenolic Coupling – *Erysodine*

- ***Erysodine:*** an ***erythrina alkaloid*** (*Erythrina crista-galli):*
  - ***biosynthesis:*** *p-/p-* oxidative phenolic coupling of ***nor-reticuline*** *via* ***dienone-phenol*** rearrangement:
    - Zenk *et al. Phytochem.* **1999**, *52*, 373 (DOI)



  - broad range of biological activity – *e.g.* constituent of curare poison arrow tips!

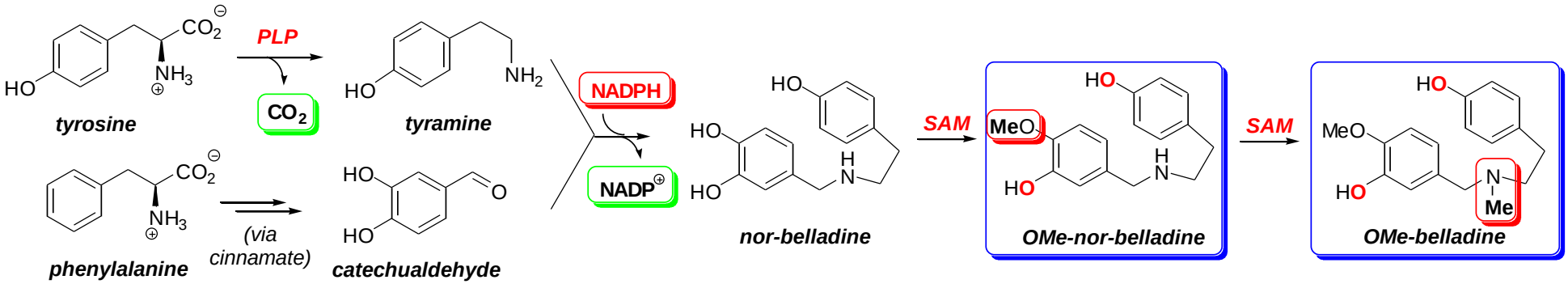
Amaryllidaceae Alkaloids



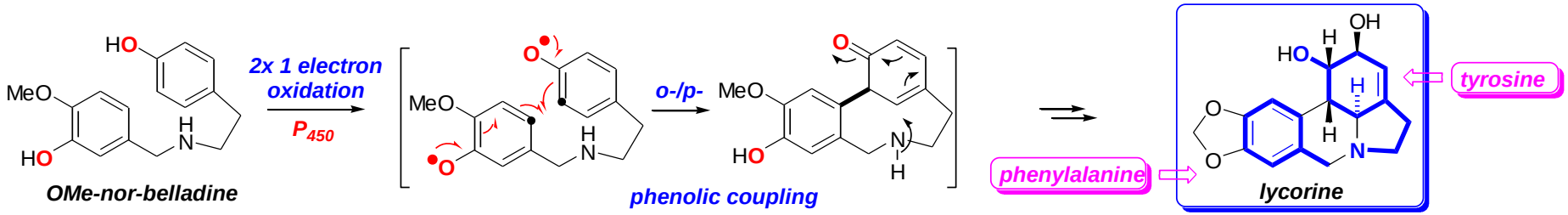
(–)-galanthamine

Amaryllidaceae Alkaloids

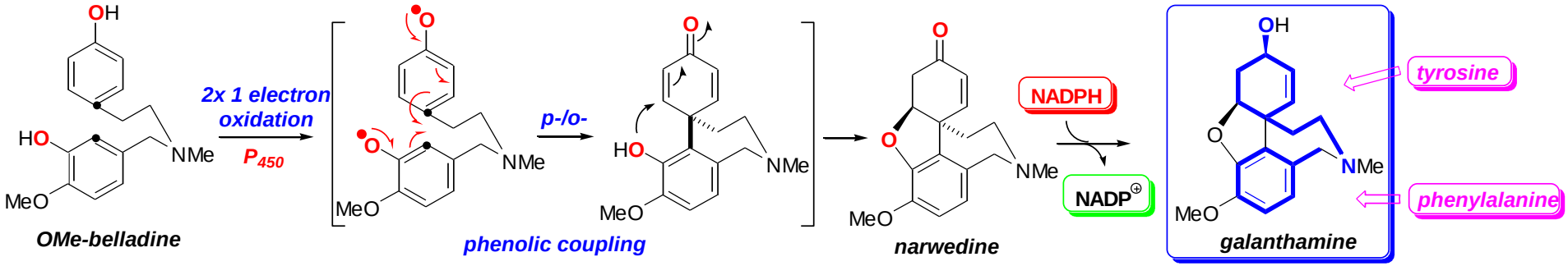
- ***Amaryllidaceae alkaloids*** are formed by ***oxidative phenolic coupling*** of ***belladine*** derivatives:



  - ***Lycorine: anti-tumour*** constituent of ***daffodils:***

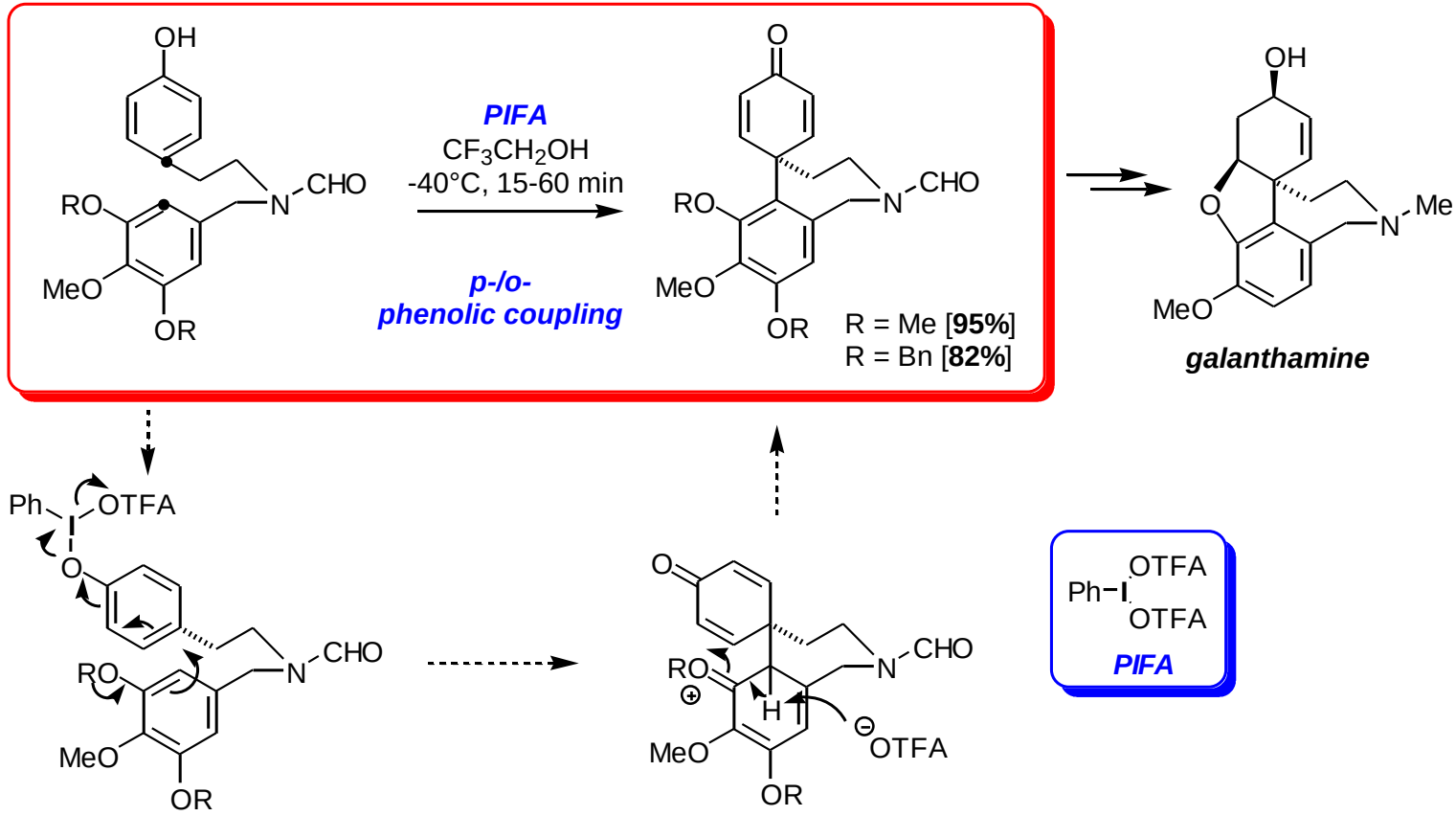


  - ***Galanthamine: anti-Alzheimer's*** constituent of ***snowdrops*** (*Galanthus nivalis*)



Biomimetic Synthesis of Galanthamine

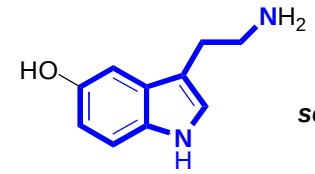
- ***Oxidative phenolic coupling*** using ***hypervalent iodine (PIFA = I^{3+}):***
  - ***review:*** Pal & Pal *Curr. Sci.* **1996**, *71*, 106-108 (DOI)
  - Node *Angew. Chem. Int. Ed.* **2004**, *43*, 2659 (DOI); Node *Angew. Chem. Int. Ed.* **2001**, *40*, 3060 (DOI); Node *Tet.* **2004**, *60*, 4901 (DOI) & Quideau *Tet. Letts.* **2001**, *42*, 7393 (DOI)
  - *cf.* 1st biomimetic route to closely related alkaloid ***narwedine***: Barton & Kirby *J. Chem. Soc.* **1962**, 806 (DOI)



Tryptophan Derived Alkaloids

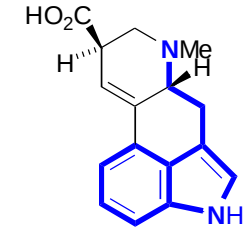
- ***Alkaloids containing an indole subunit:***
  - Skeleta built up by ***reductive amination***, ***decarboxylation*** & ***hydroxylation***)
  - ***Major classes:***

  - **simple derivatives** (*e.g.* serotonin, bufotenine)



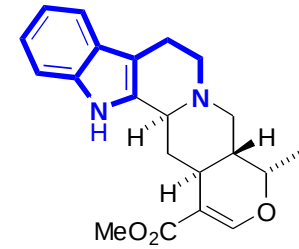
serotonin

  - **mixed Trp/mevalonate alkaloids** *e.g.*
    - ***ergot*** [DMAPP derived] (*e.g.* ergoline, lysergic acid)



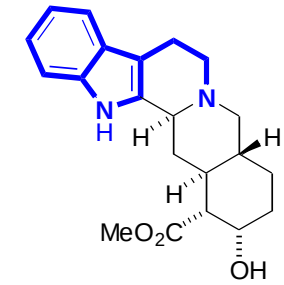
lysergic acid (ergot)

    - ***vinca*** [secologanin derived]



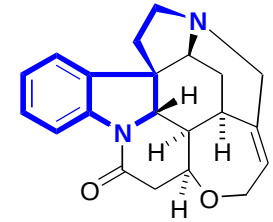
ajmalicine (vinca)

    - ***yohombine*** [secologanin derived]



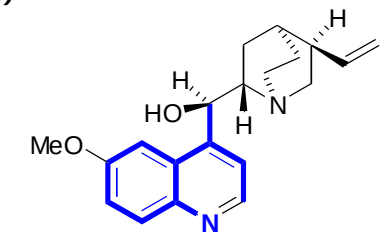
yohimbine

    - ***strychnos*** [secologanin derived]



strychnine (strychnos)

    - ***quinine*** [secologanin derived]

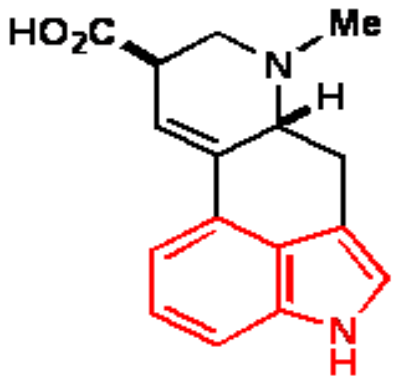


quinine

Indole Alkaloids

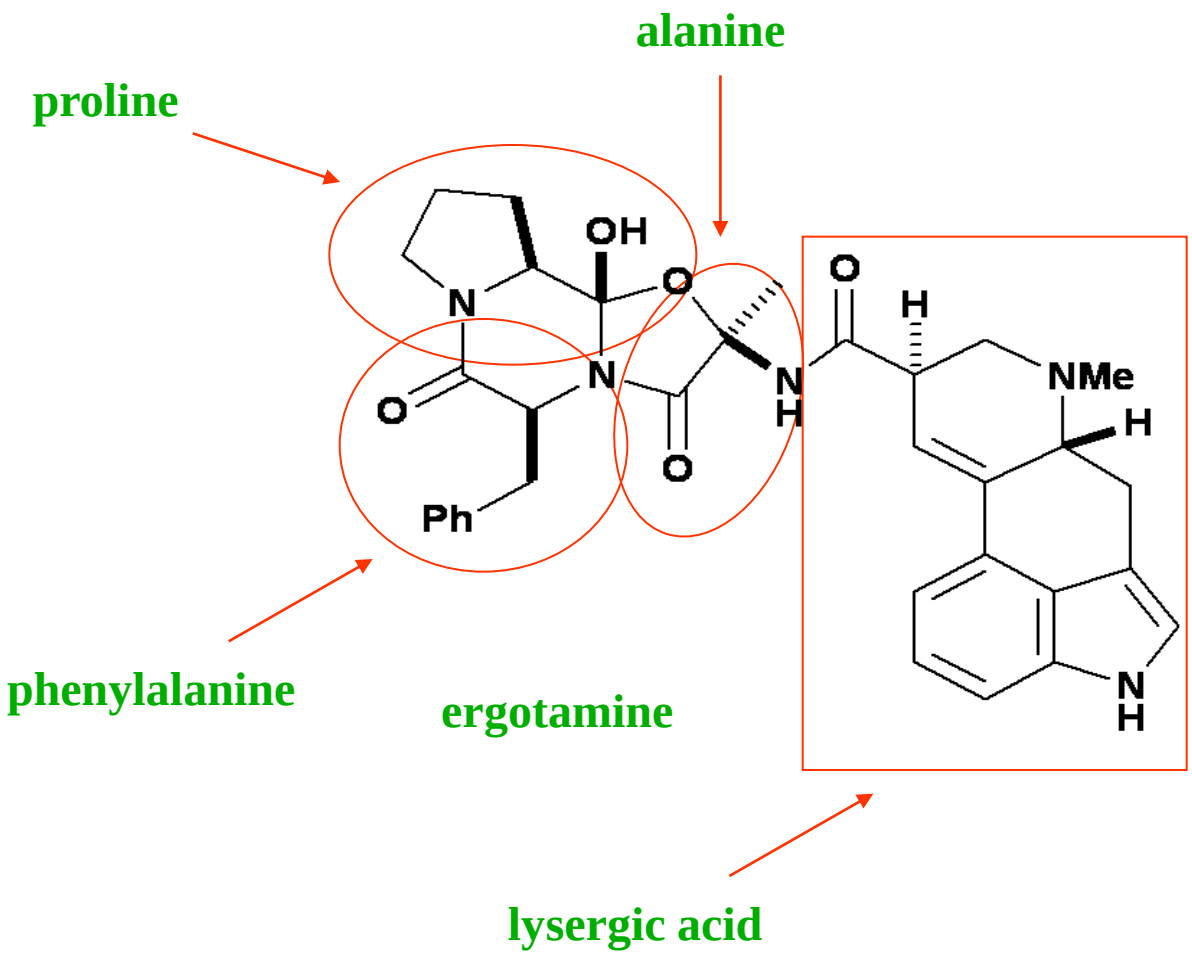
Indole Alkaloids

bufotenine



lysergic acid

Ergot alkaloids



Claviceps purpurea

Effects: burning and convulsions, hallucinations with imaginary sounds, gangrene and loss of limbs, permanent insanity, and occasionally death

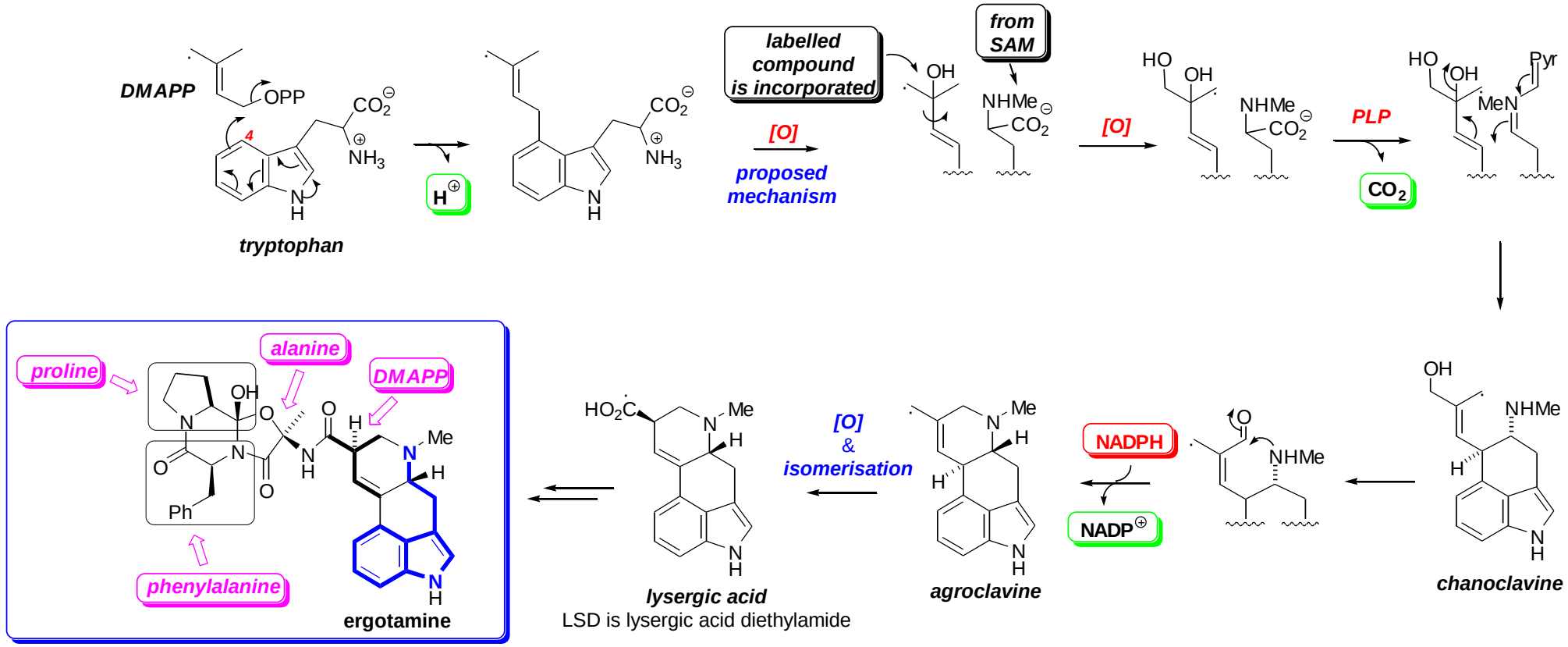
Ergot Alkaloids

Salem Witchcraft Trials
1692

Caporael ‘Ergotism: The Satan Loosed in Salem?’ *Science* **1976**, *192*, 21-26 (DOI)

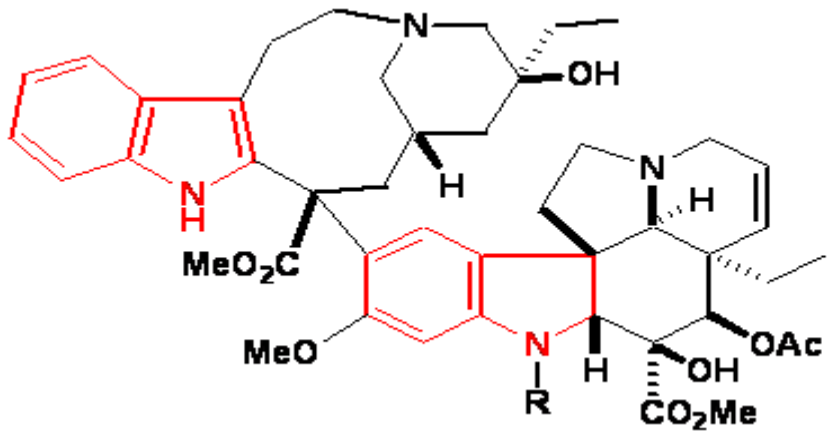
Ergot Alkaloids – *Lysergic acid*

- ***Ergot alkaloids:*** from *Claviceps purpurea* Grows on ***rye***. Eating bread made from infected rye causes hallucinations, convulsions, burning sensation (St Anthony's or Holy Fire) and in bad cases gangrene
  - ***biosynthesis of ergotamine:*** used to cause contractions of the uterus following childbirth
  - mixed ***tryptophan/DMAPP (C_5)*** metabolite:



Dimeric Indole Alkaloids – *Vinca extracts*

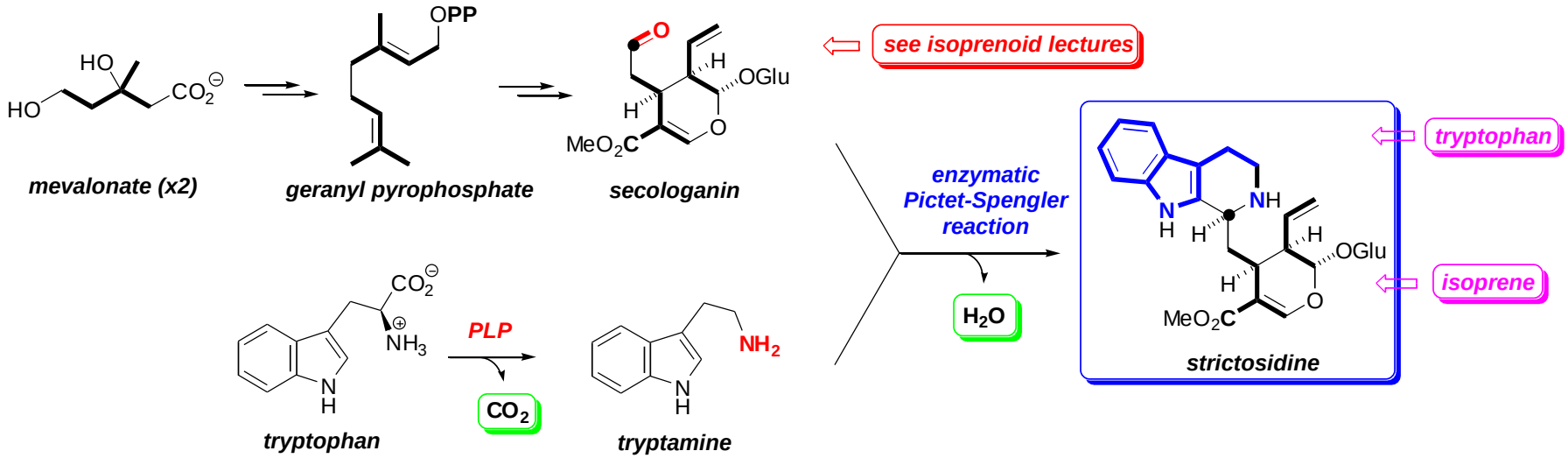
Dimeric Indole Alkaloids



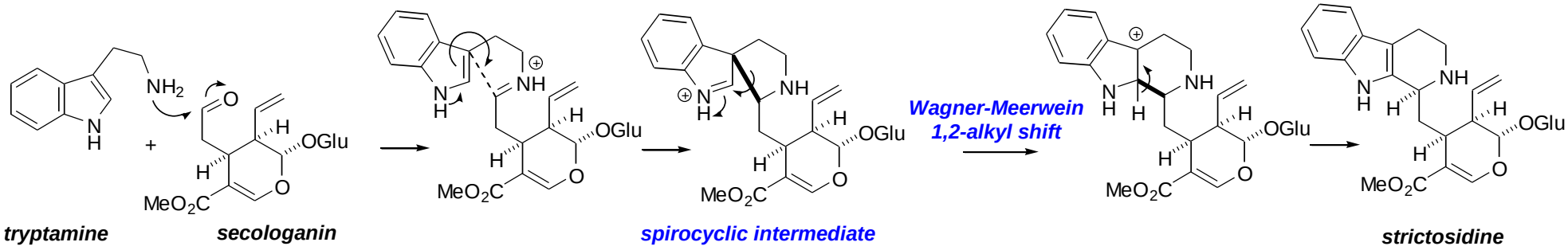
vinblastine (R = Me)
vincristine (R = CHO)

Tryptamine + Secolaganin → Strictosidine

- Most alkaloids of ***mixed Tryptophan/mevalonate biogenesis*** (>1200) are derived from ***strictosidine:***
  - ***Strictosidine*** is derived from the condensation of ***tryptamine*** with the iridoid C_{10} monoterpene ***secologanin:***

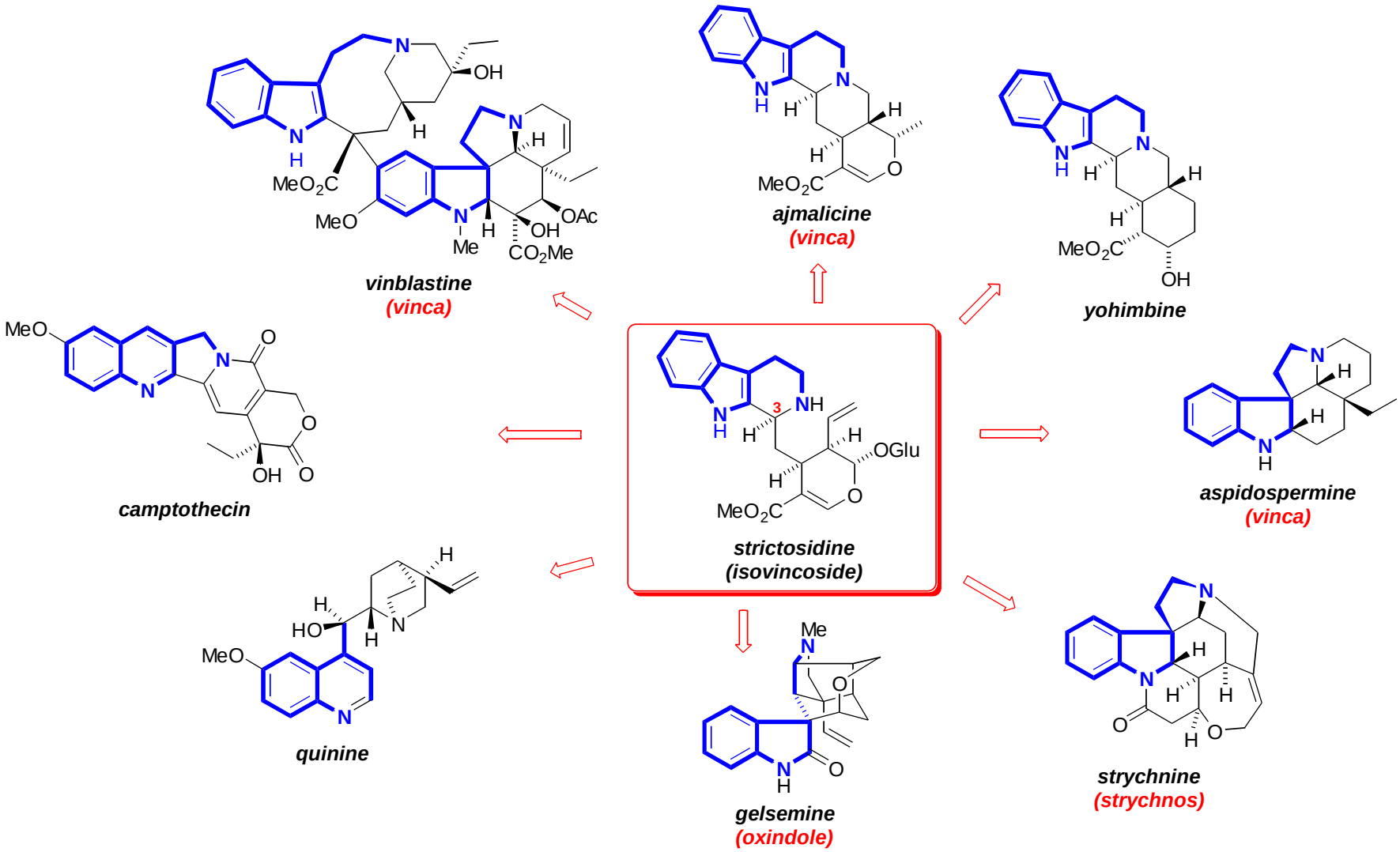


  - ***Mechanism of Pictet-Spengler reaction:***
    - *via* ***spirocyclic*** intermediate then ***Wagner-Meerwein*** 1,2-alkyl shift:



Strictosidine → *Vinca, Strychnos, Quinine etc.*

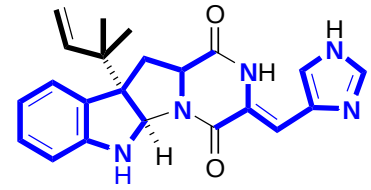
- The diversity of alkaloids derived from ***strictosidine*** is stunning and many pathways remain to be fully elucidated:



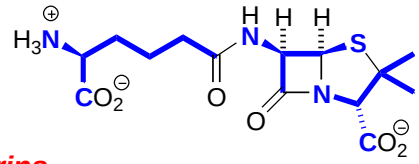
Non-Ribosomal Peptides & Derivatives

- ***Simple dipeptides & derivatives:***
  - di-peptides (diketopiperazines); penicillins & cephalosporins
- ***Cyclic polypeptides:***
  - cytokines, chemokines, siderophores *etc.*

cyclic dipeptides - diketopiperazines
(e.g. roquefortine)

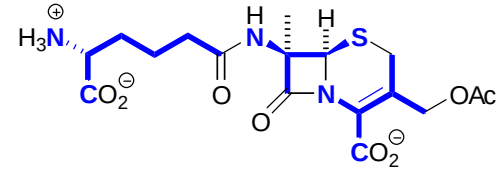


roquefortine



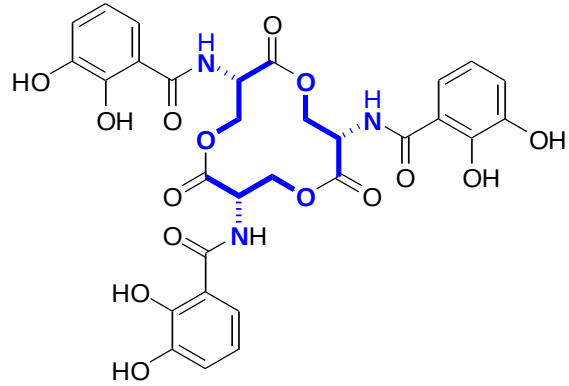
isopenicillin N

penicillins & cephalosporins



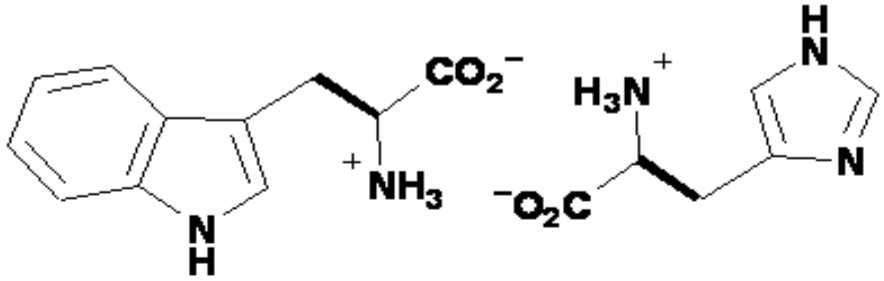
cephalosporin C

cyclic polypeptides
(e.g. enterobactin)

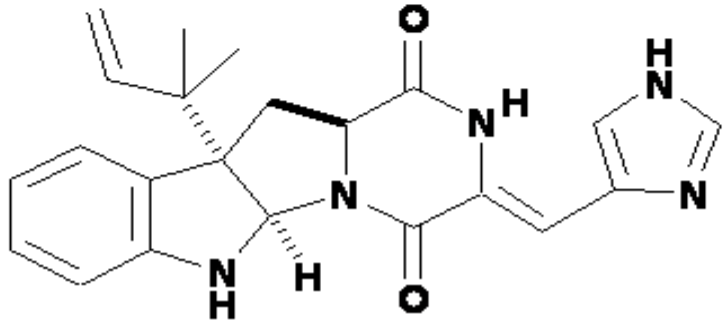


enterobactin

Roquefortine - from *Penicillium roquefortii*



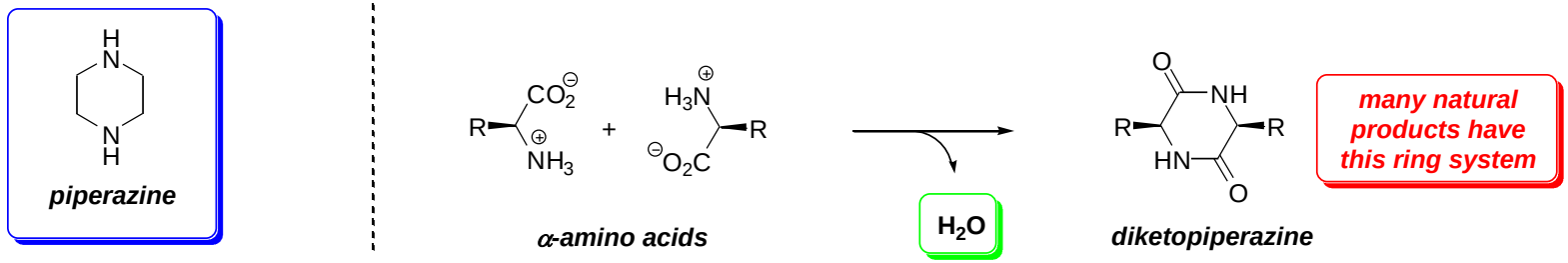
tryptophan **histidine**



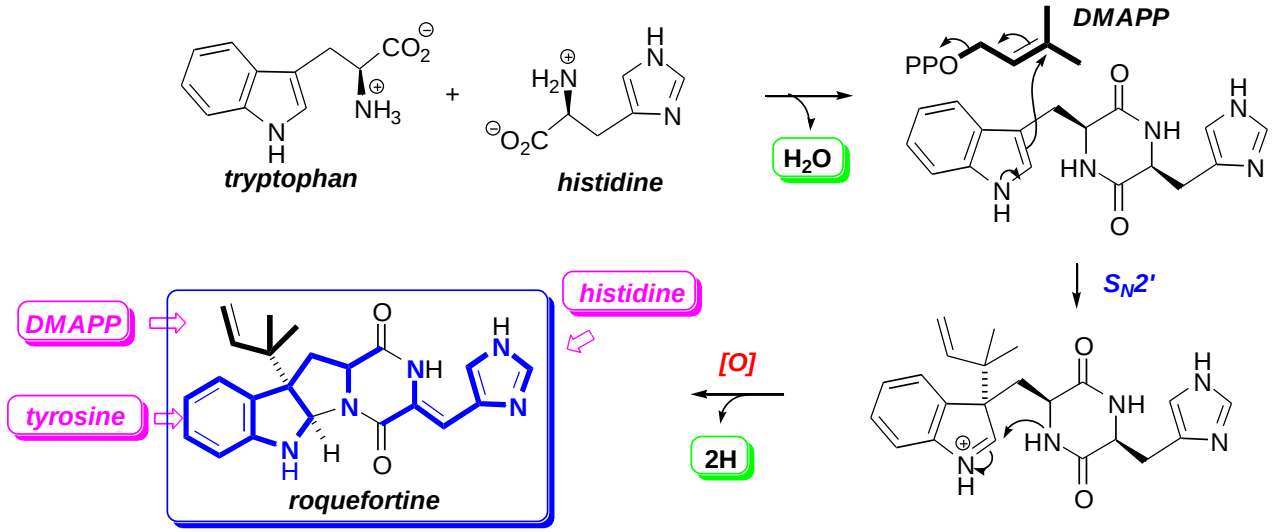
roquefortine

Cyclic Dipeptides - *Diketopiperazines*

- ***Diketopiperazines are formed by the dehydrative dimerisation of two amino acids:***
  - dimerisation does not occur at RT...but does if heated strongly or if ester or acid chloride used

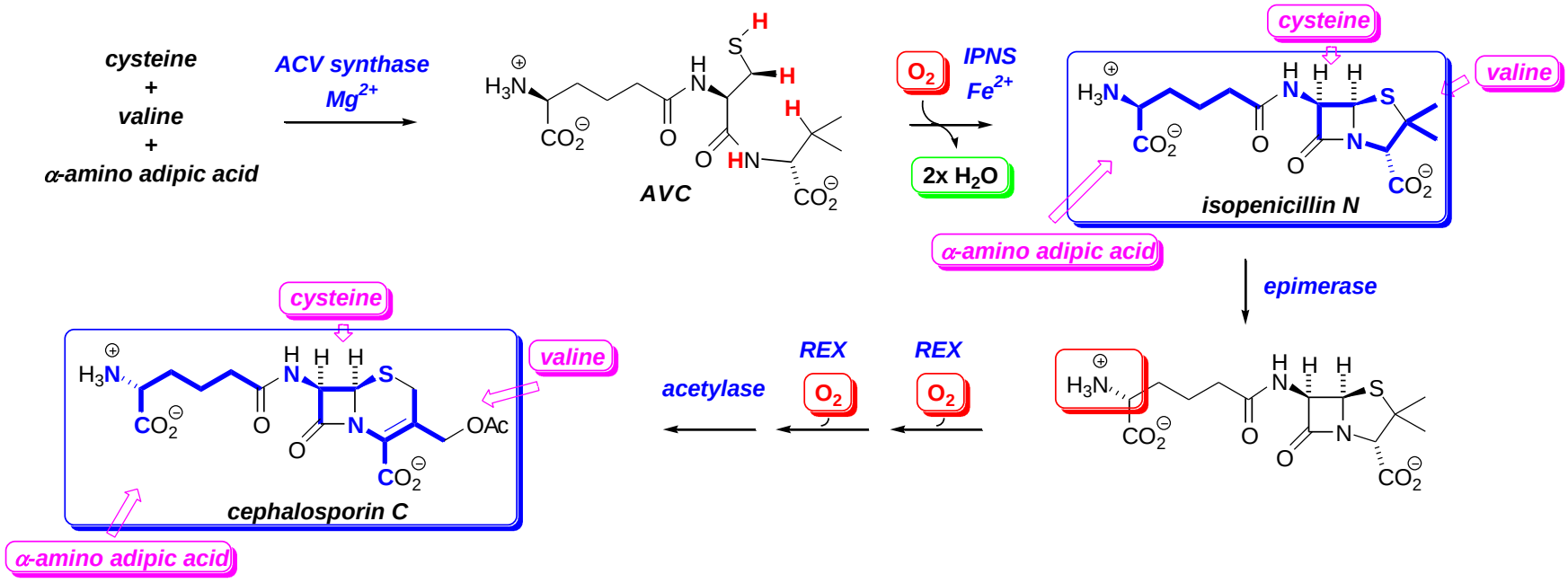


- ***Roquefortine:*** metabolite of ***blue mould*** (*Penicillium roquefortii*) in ***Roquefort cheese***
  - ***biosynthesis:***



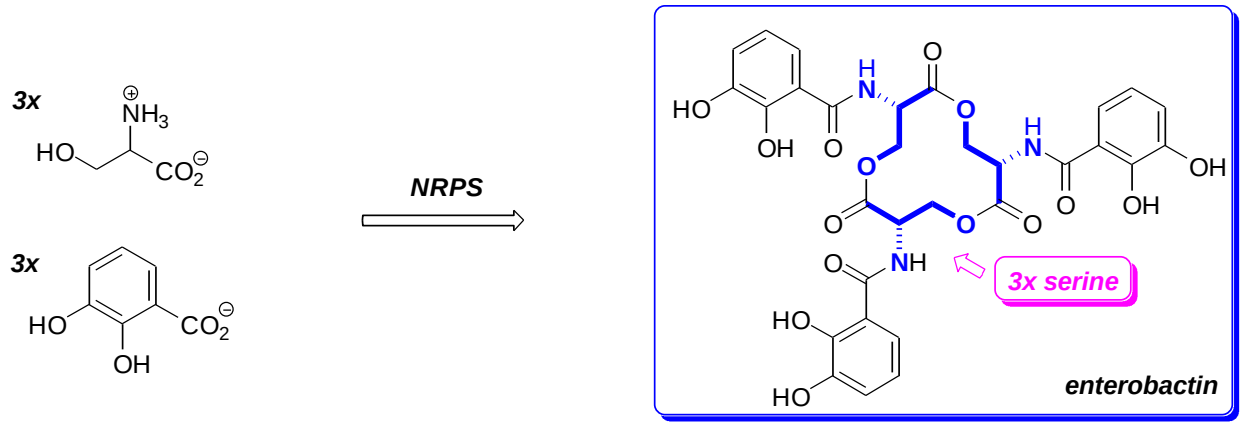
Penicillins & Cephalosporins

- Famous story of the antibiotic penicillin:
  - ***discovery*** by bacteriologist ***Alexander Fleming*** at St Mary's Hospital, London (published in ***1929***)
  - ***isolation & development*** by ***Howard Florey*** & ***Ernst Chain*** at the Dunn School of Pathology Oxford University (***1939-1945***)
    - E. Lax 'The mould in Dr Florey's coat' Little Brown & Co., **2004**, [ISBN 0316859257]
  - ***biosynthesis*** extensively studied by Baldwin:
    - Baldwin *J. Het. Chem.* **1990**, *27*, 71 & Baldwin *et al. Nature* **1995**, *375*, 700



Cyclic Polypeptides - *NRPS*

- ***Proteins*** are synthesised by the ***ribosome*** are ***templated/encoded*** by ***RNA*** (*i.e.* ***transcription***)
- However, many ***cyclic polypeptides*** are synthesised by ***Non Ribosomal Peptide Synthases*** (***NRPS***s)
  - synthesised on ***huge modular (multi-domain) proteins*** not unlike ***polyketide synthases*** (***PKS***s) & ***fatty acid synthases*** (***FAS***s)...see later lectures
  - *e.g.* ***enterobactin***: siderophore with a high affinity for Fe^{3+} ($K_D = 10^{-52}$!)



  - steps (=modules) comprise:
    - *activation*
    - *priming and loading*
    - *elongation*
    - *transfer*
    - *termination/cyclisation*
  - for further details see any recent Biochemistry text

Primary Metabolism - *Overview*