Biosynthesis – Inspiration for Drug Discovery

Biosynthesis of Alkaloids

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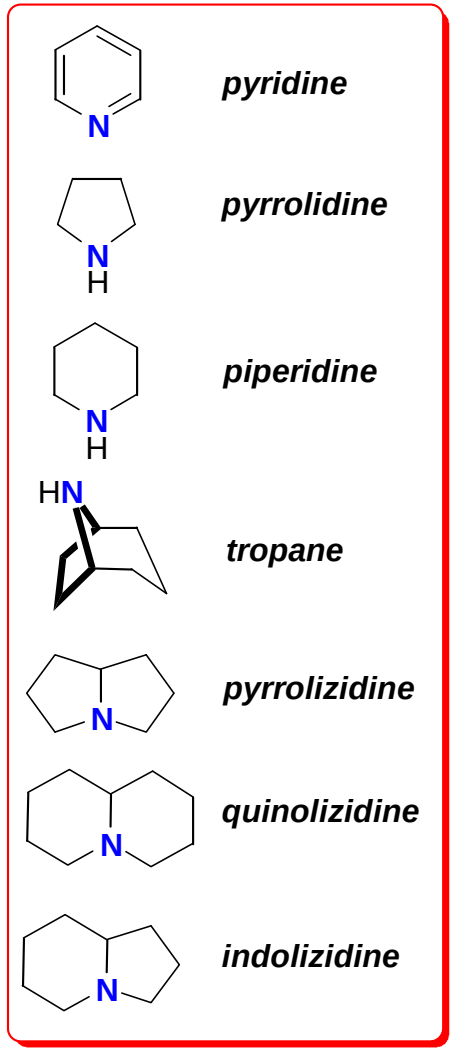
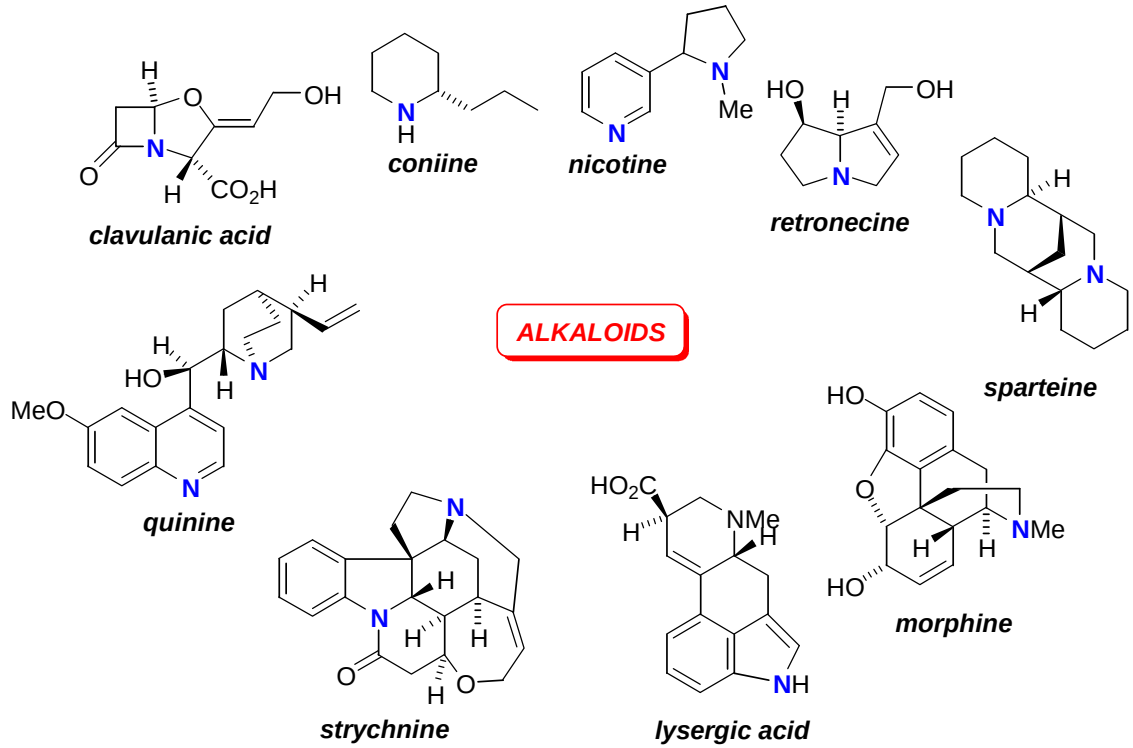
Format & Scope of Lecture

- ***What are alkaloids?***
  - definitions, 1° metabolism → α-amino acids (Lys, Orn)
    - the citric acid cycle – oxaloacetate & α-ketoglutarate
    - pyridoxal – transamination, racemisation & decarboxylation
- ***Phenylalanine & tyrosine derived alkaloids***
  - benzylisoquinolines – *e.g.* opium, aporphine & erythrina alkaloids
- ***Tryptophan derived alkaloids***
  - mixed tryptophan/mevalonate (isoprenoid) alkaloids:
    - secologanin derived: vinca-, strychnos- & quinine alkaloids *etc.*
- ***Non-ribosomal peptides & derivatives***
  - penicillins & cephalosporins

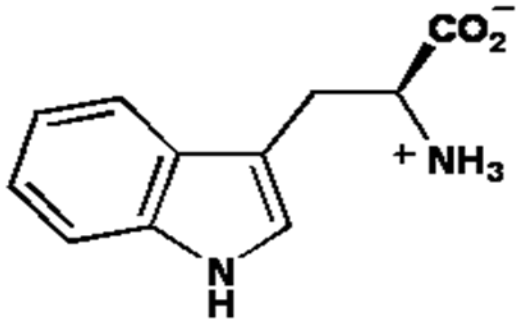


Alkaloids

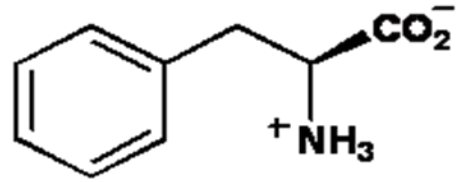
- ***Definitions:***
  - ***originally –*** '*a natural product that could be extracted out of alkaline but not acidic water*' (*i.e.* containing a basic amine function that protonated in acid)
  - ***more generally -*** '*any non-peptidic & non-nucleotide nitrogenous secondary metabolite*'



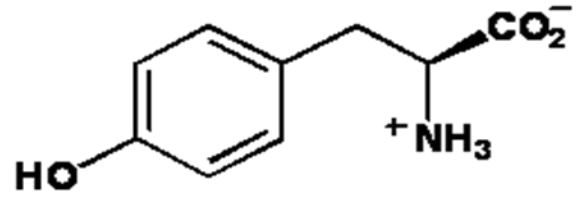
α-Amino Acids used to make Alkaloids



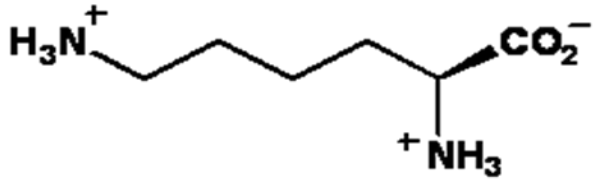
tryptophan



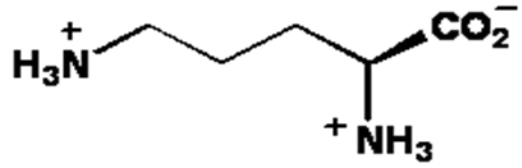
phenylalanine



tyrosine

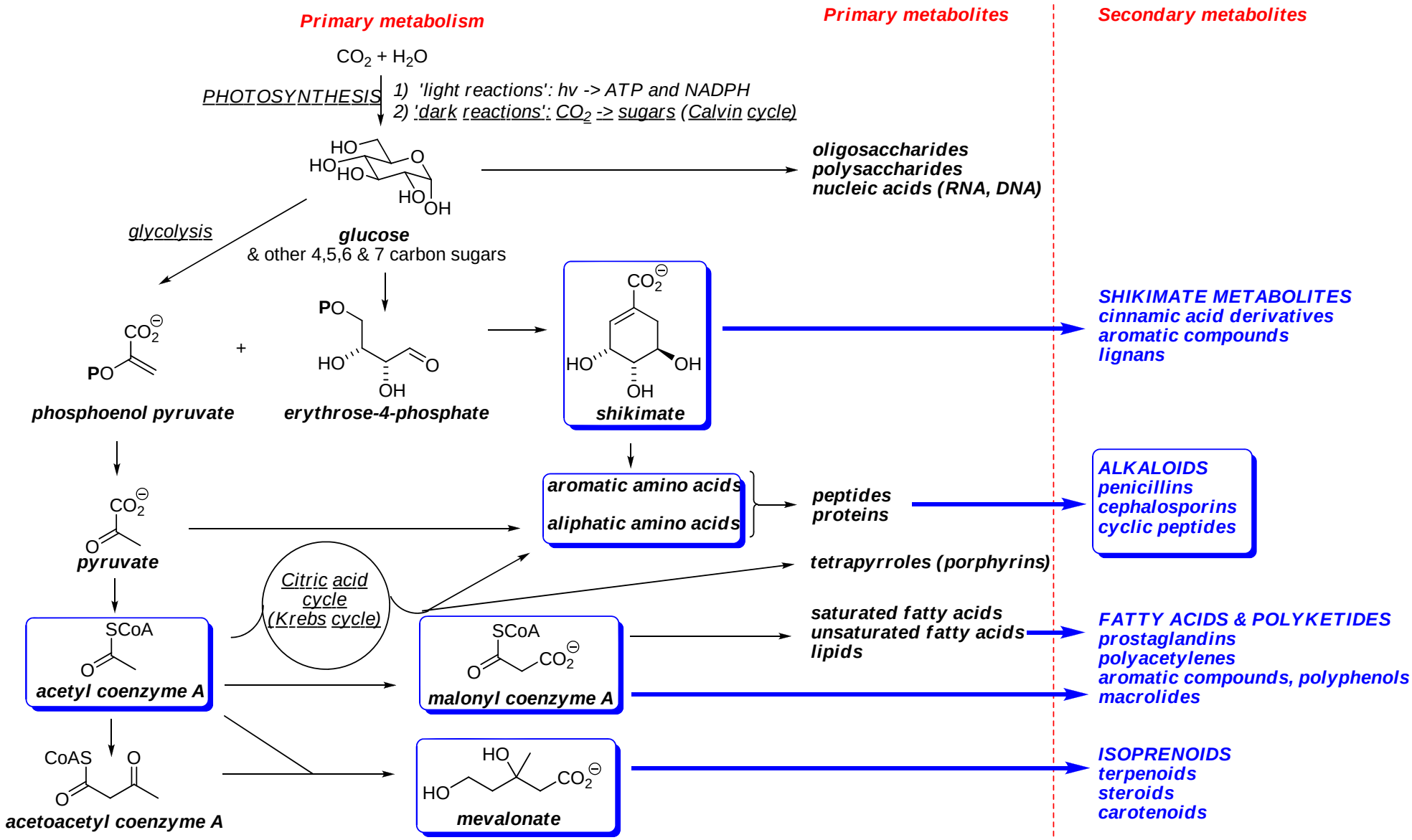


lysine



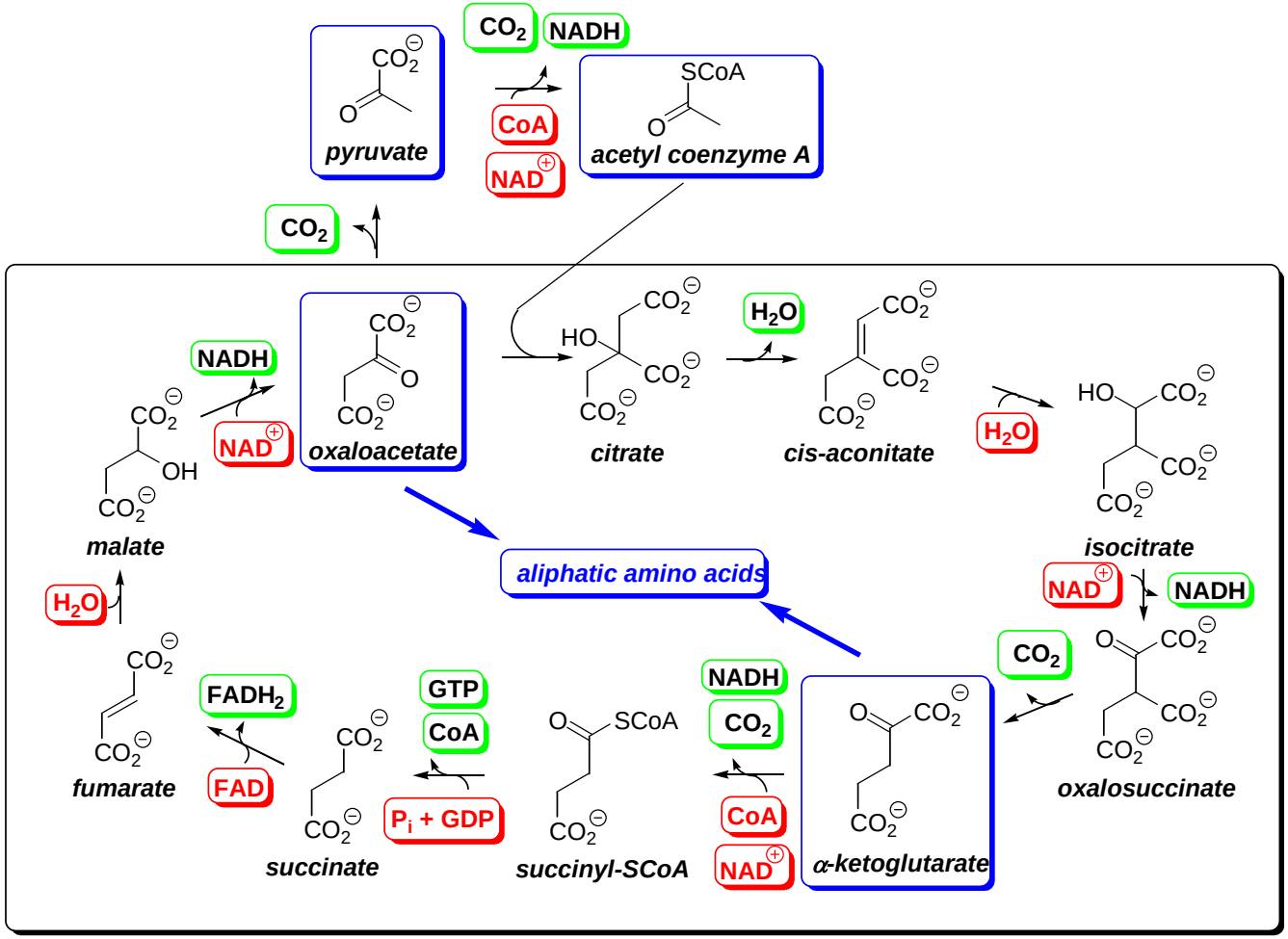
ornithine

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

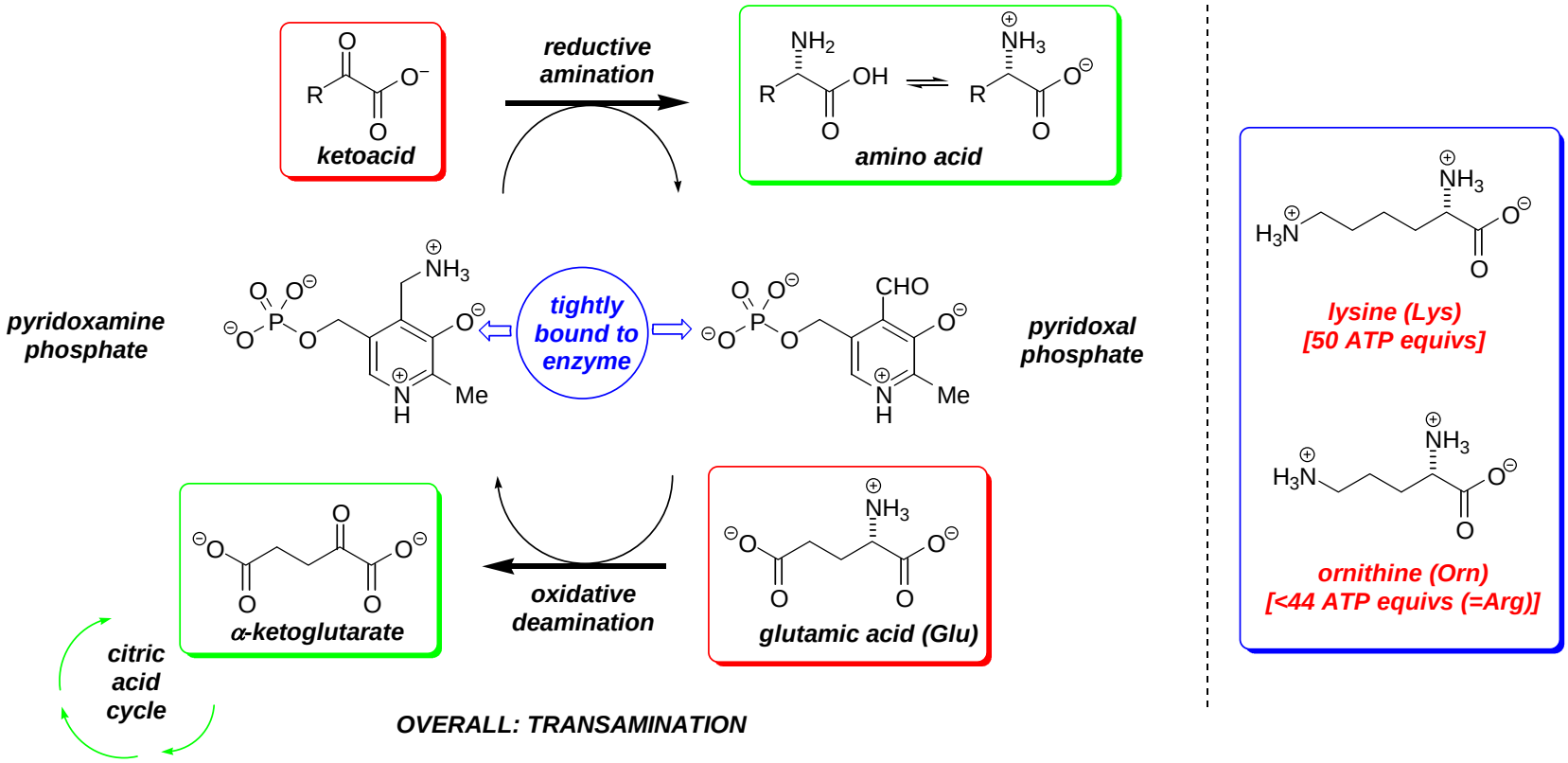
2x CO_2

+

12x ATP *energy!*

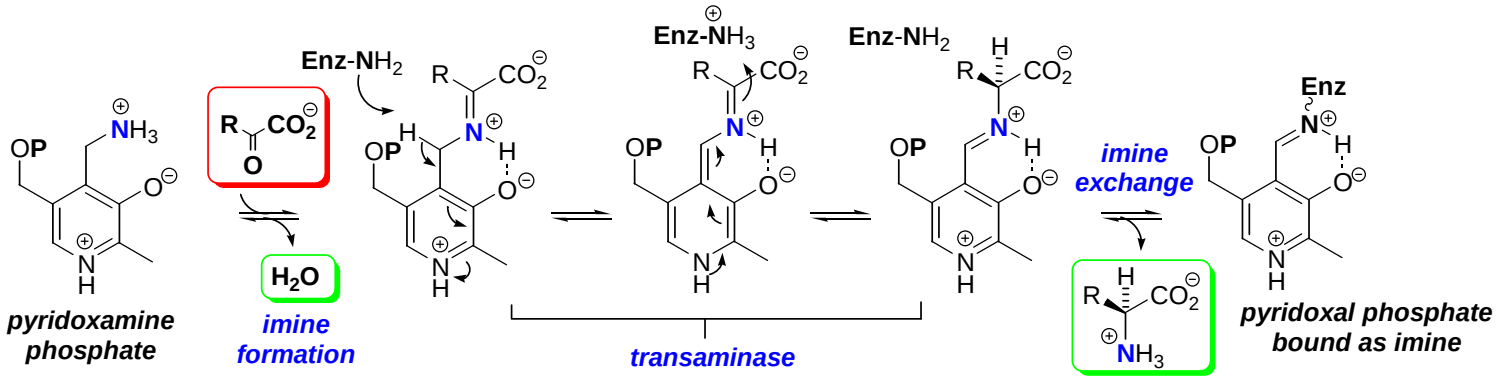
The Biosynthesis of Lysine & Ornithine

- ***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***)

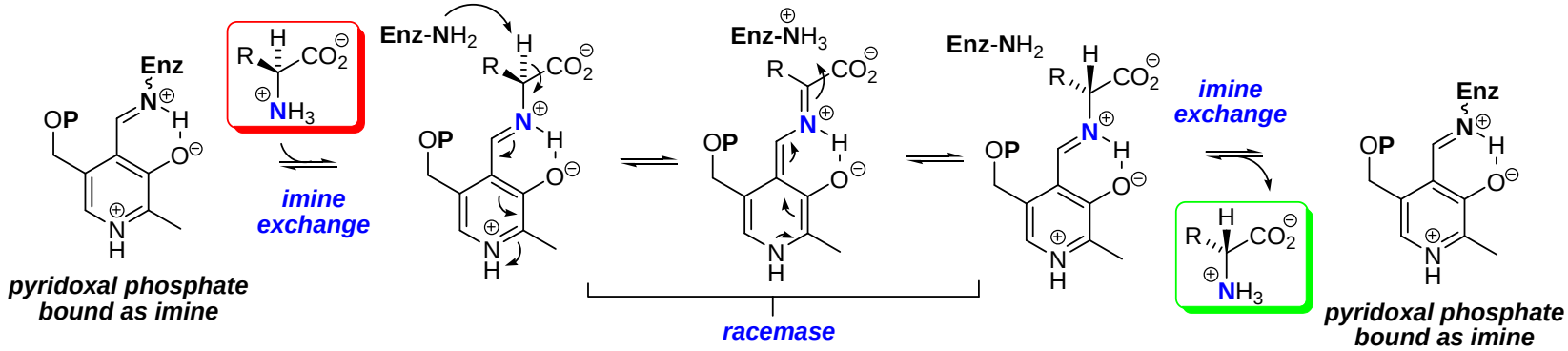


PLP Chemistry – *Transamination & Racemisation*

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

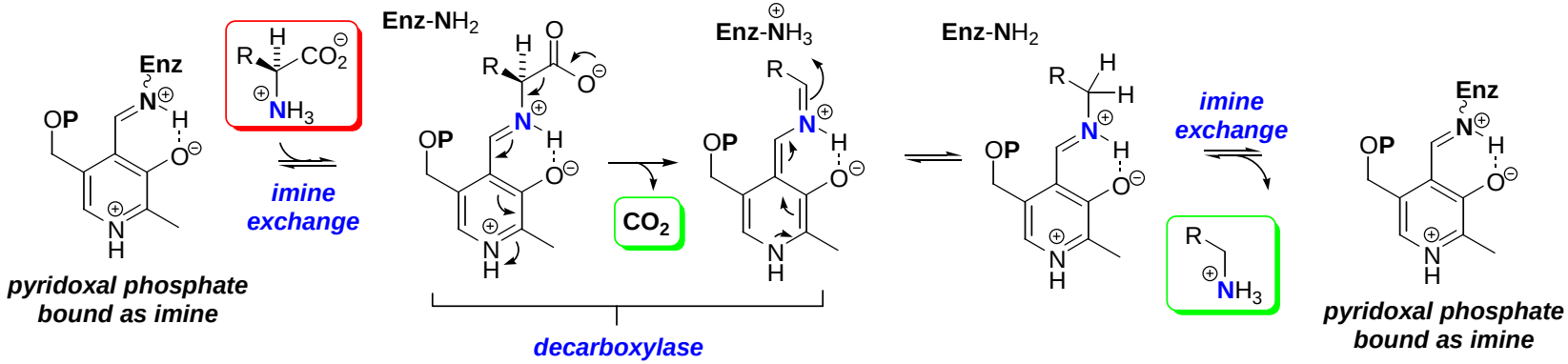


- ***Racemisation/inversion of configuration***:

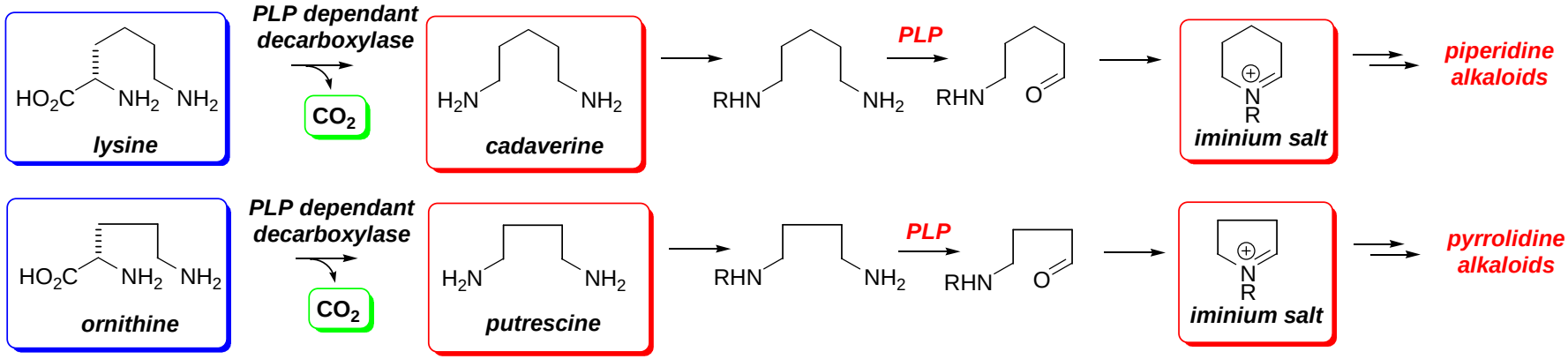


PLP Chemistry – *Decarboxylation*

- ***Decarboxylation***:

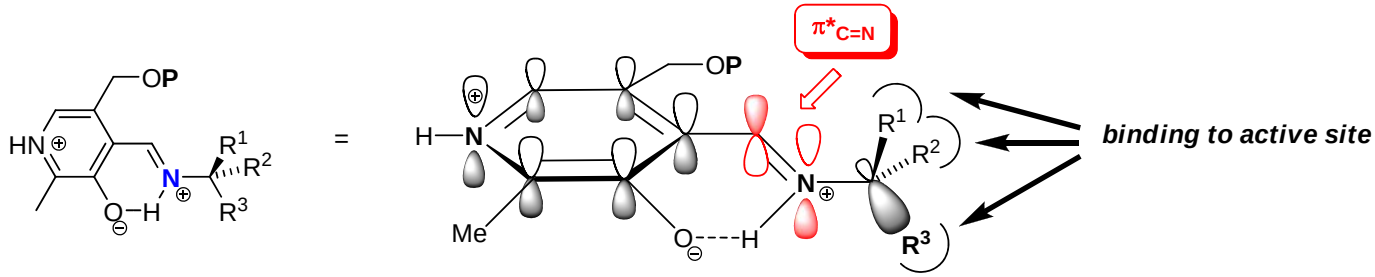


- Decarboxylation of ***lysine*** & ***ornithine***:

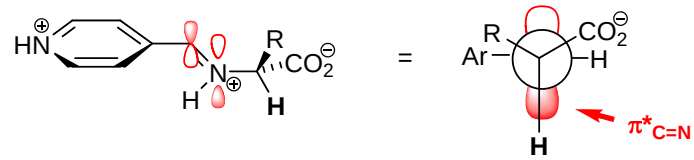


Control of PLP Activity – *Stereoelectronics*

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

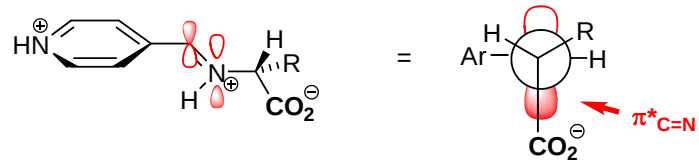


racemisation
(α proton loss)



$\sigma_{C\text{-}H}$ -> $\pi^*_{C=N}$ *anti peri planar*

decarboxylation

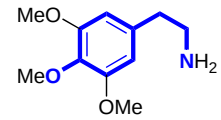


$\sigma_{C\text{-}C}$ -> $\pi^*_{C=N}$ *anti peri planar*

Phenyalanine & Tyrosine Derived Alkaloids

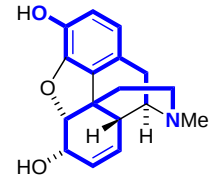
- ***Alkaloids (generally) containing an ArC_2N subunit (± ArC_2/ArC_1):***
  - Skeleta built up by ***reductive amination***, ***decarboxylation***, ***oxidation*** (*e.g.* ***phenolic coupling***, ***hydroxylation***)
  - ***Major classes:***

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



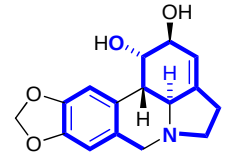
mescaline

  - ***Benzylisoquinolines***
  - opium alkaloids (*e.g.* papaverine, morphine)
  - aporphine alkaloids
  - erythrina alkaloids



morphine

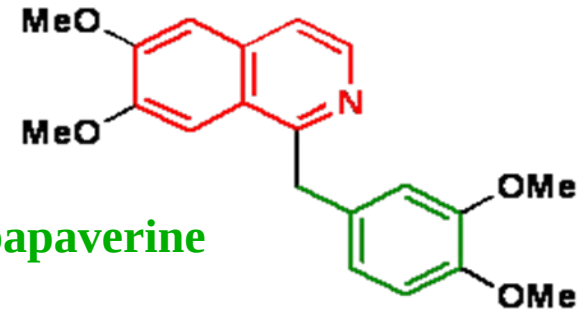
  - ***amaryllidaceae alkaloids***
  - *e.g.* lycorine, galanthamine



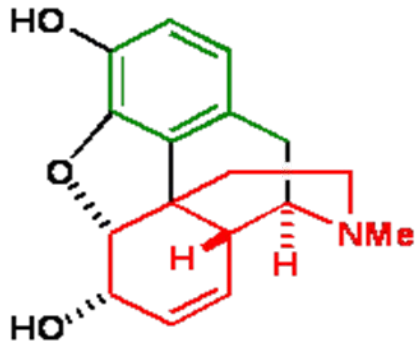
lycorine

Benzylisoquinoline Opium Alkaloids

Benzylisoquinoline Alkaloids



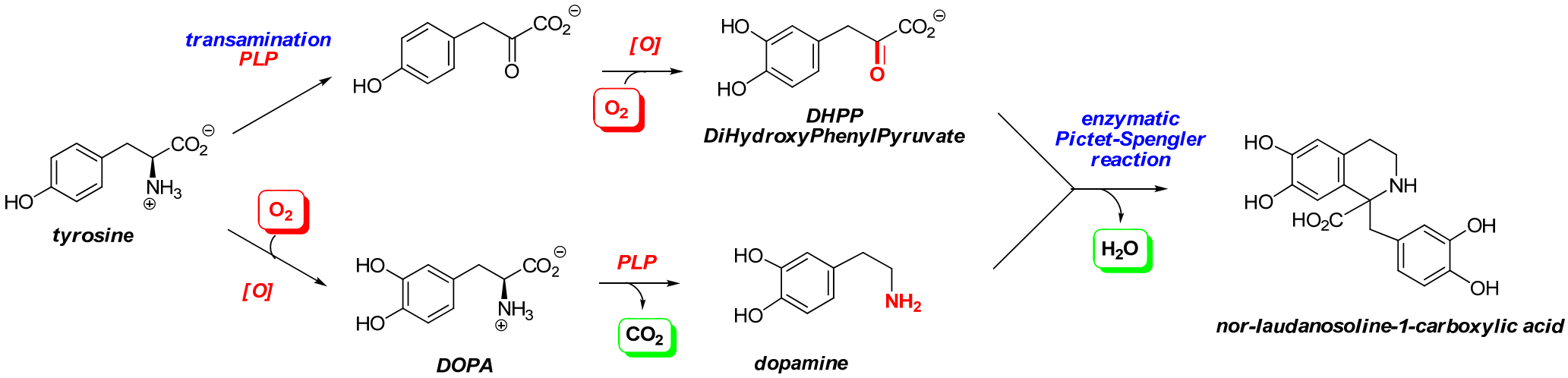
papaverine



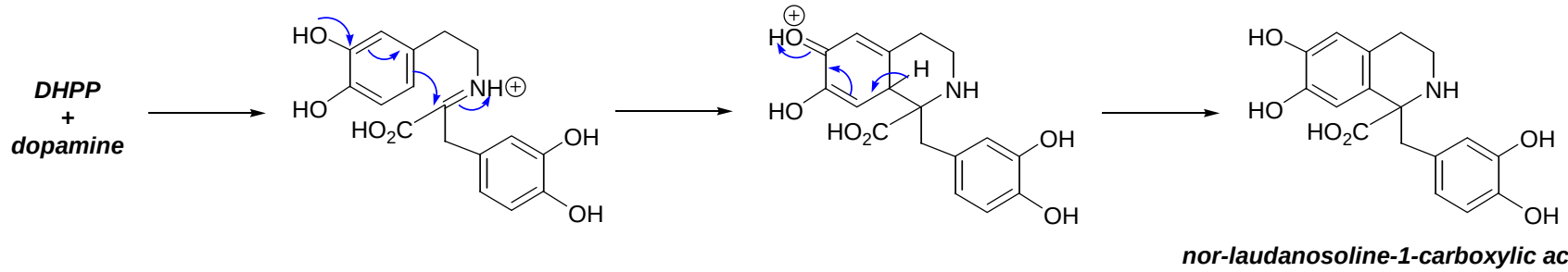
morphine

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***:

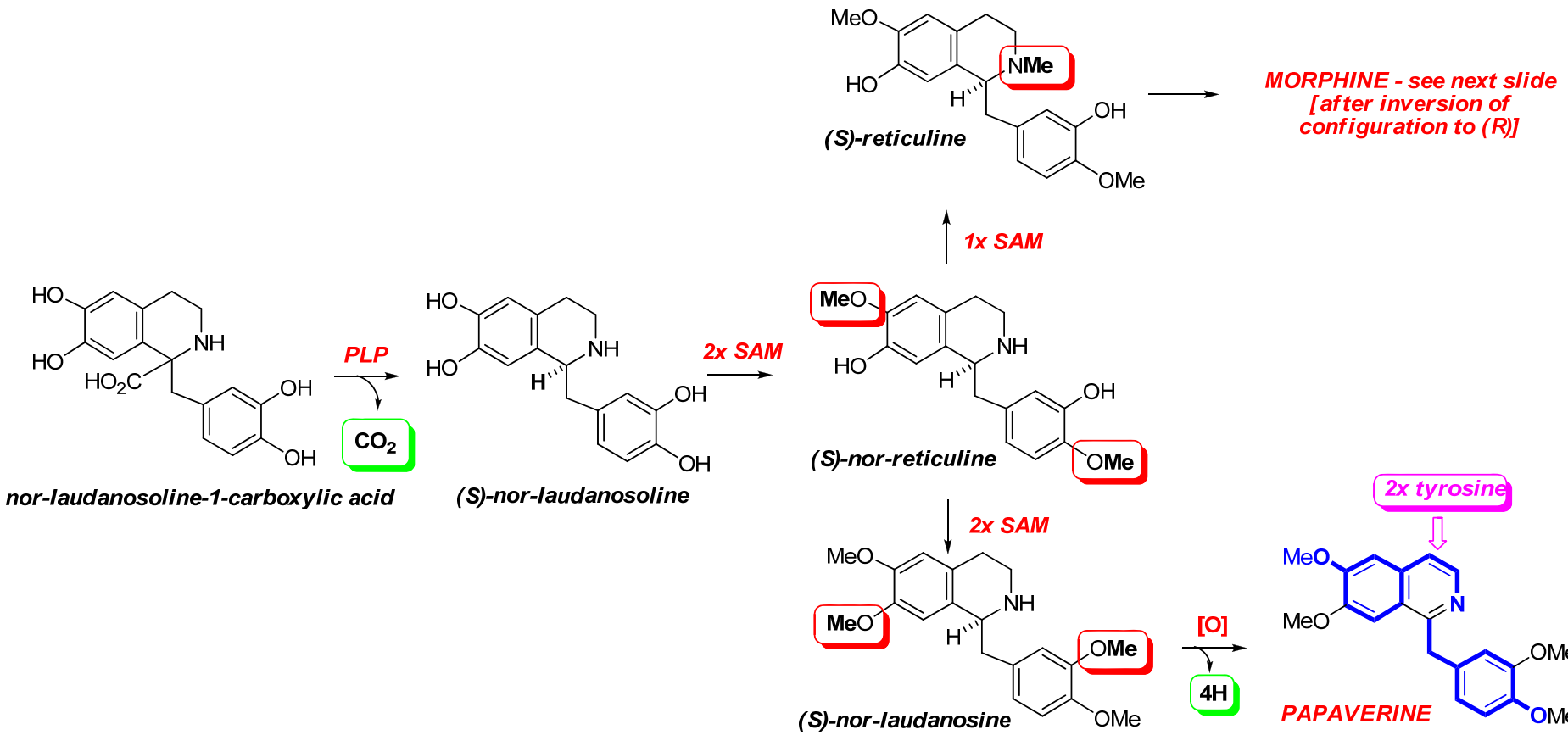


  - ***Mechanism of Pictet Spengler reaction:***



Benzylisoquinoline Alkaloids - *Papaverine*

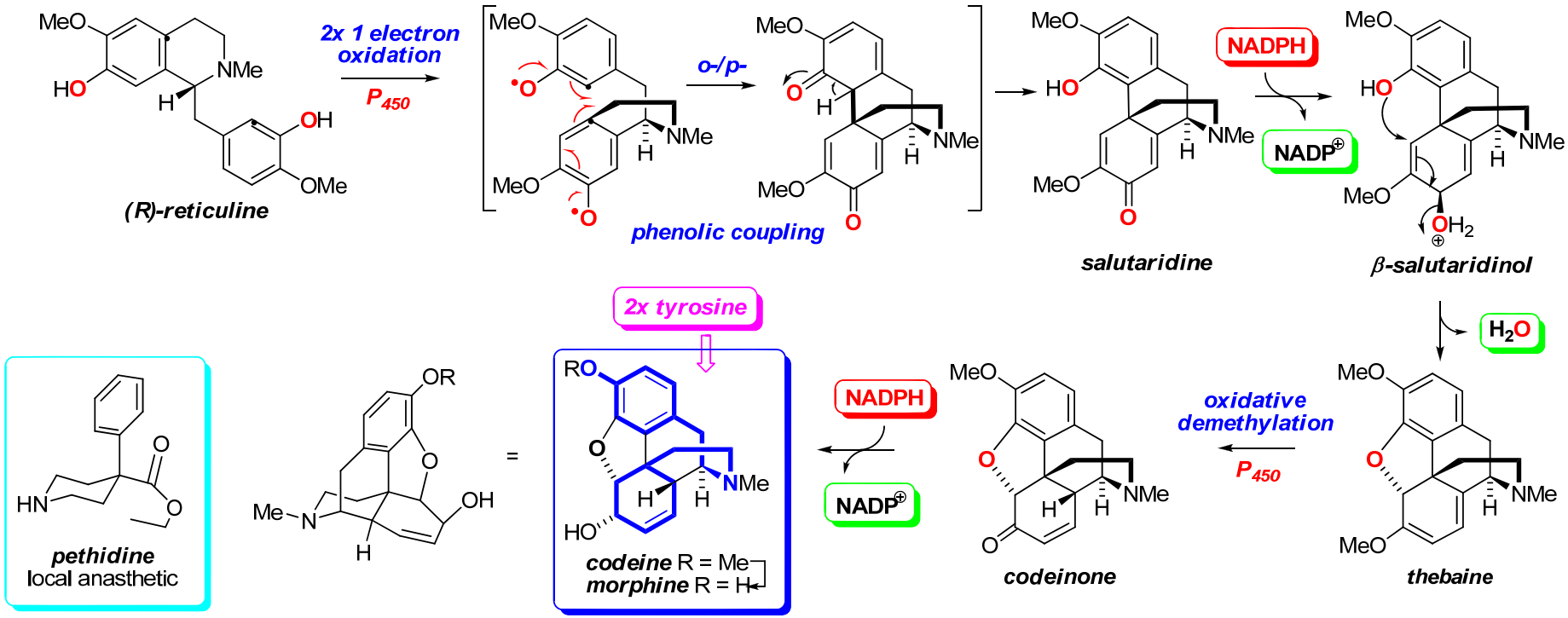
- ***Papaverine:*** analgesic contsituent of the **opium poppy** (*Papaver somniferum):*
  - ***biosynthesis:***



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

Oxidative Phenolic Coupling – *Morphine*

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

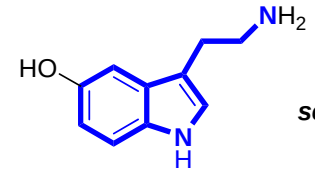


  - Morphine acts by activating the ***opiate receptors*** in the brain (IC_{50} 3 nM)
  - The natural ligands for these receptors are peptides: *e.g.* Leu-enkephalin (Tyr–Gly–Gly–Phe–Leu) (IC_{50} 12 nM)

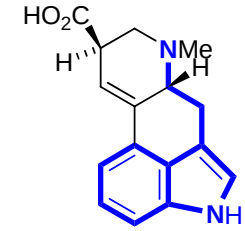
Tryptophan Derived Alkaloids

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

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

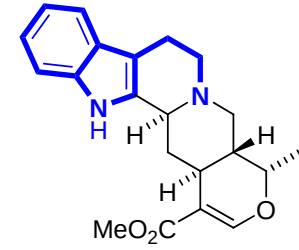


serotonin

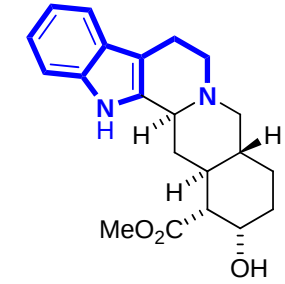


lysergic acid (ergot)

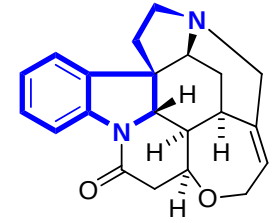
  - **mixed Trp/mevalonate alkaloids** *e.g.*
    - ***ergot*** [DMAPP derived] (*e.g.* ergoline, lysergic acid)
    - ***vinca*** [secologanin derived]
    - ***yohombine*** [secologanin derived]
    - ***strychnos*** [secologanin derived]
    - ***quinine*** [secologanin derived]



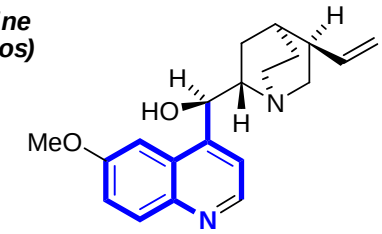
ajmalicine (vinca)



yohimbine



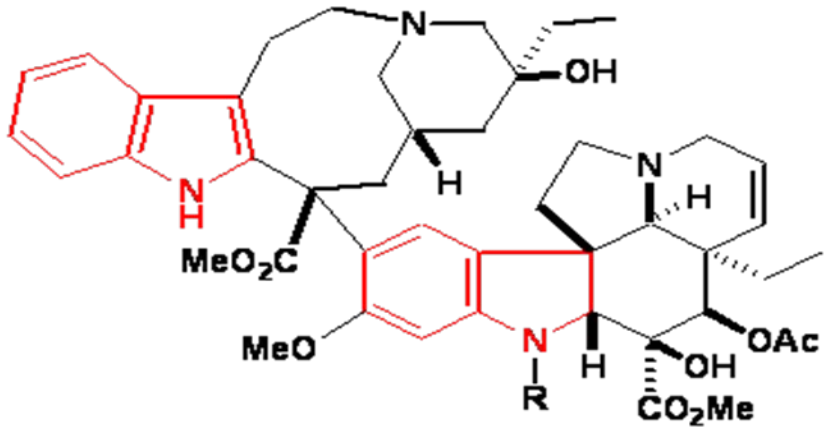
strychnine (strychnos)



quinine

Dimeric Indole Alkaloids – *Vinca extracts*

Dimeric Indole Alkaloids

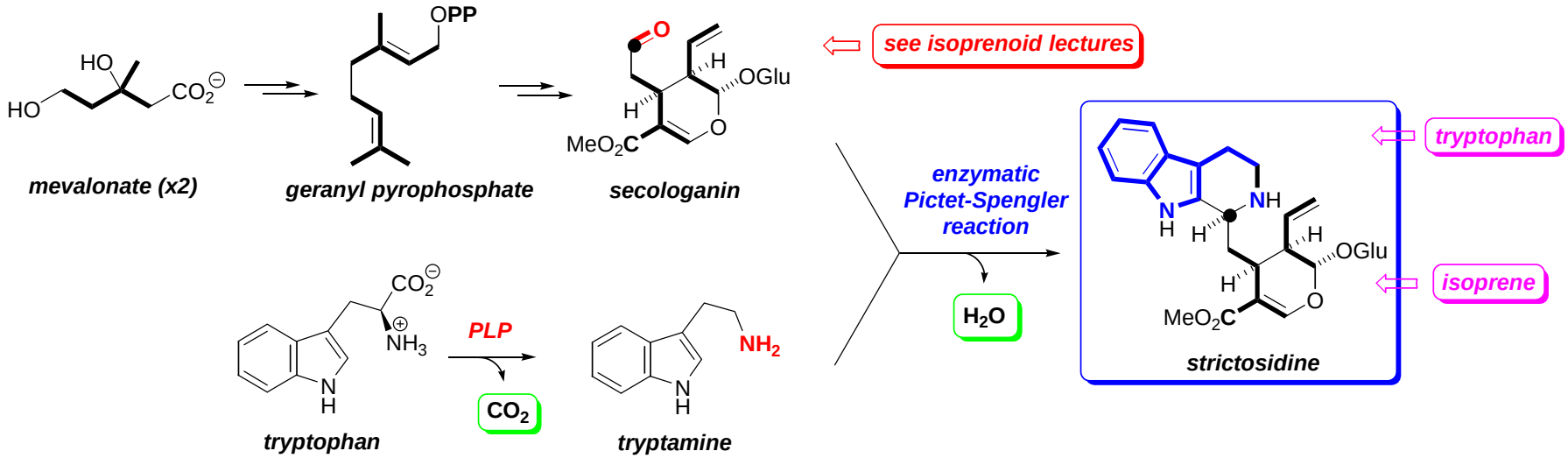


vinblastine (R = Me)
vincristine (R = CHO)

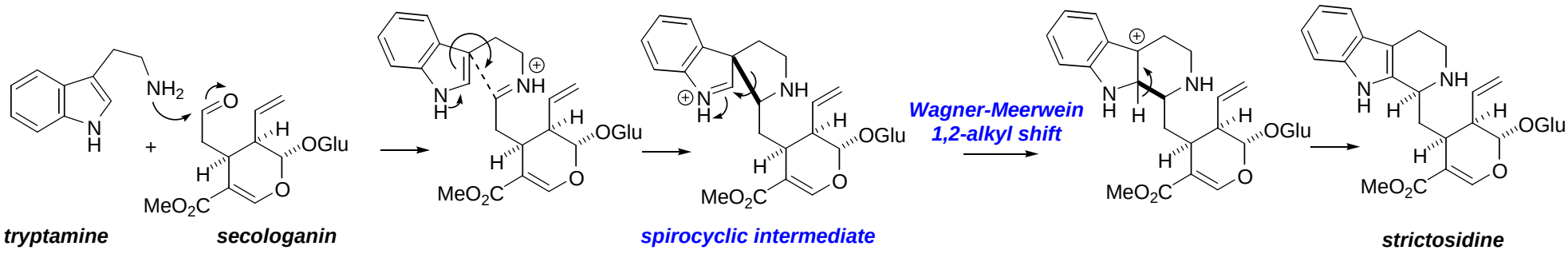
Potent **anti tumour** alkaloids used in **cancer chemotherapy**

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:***

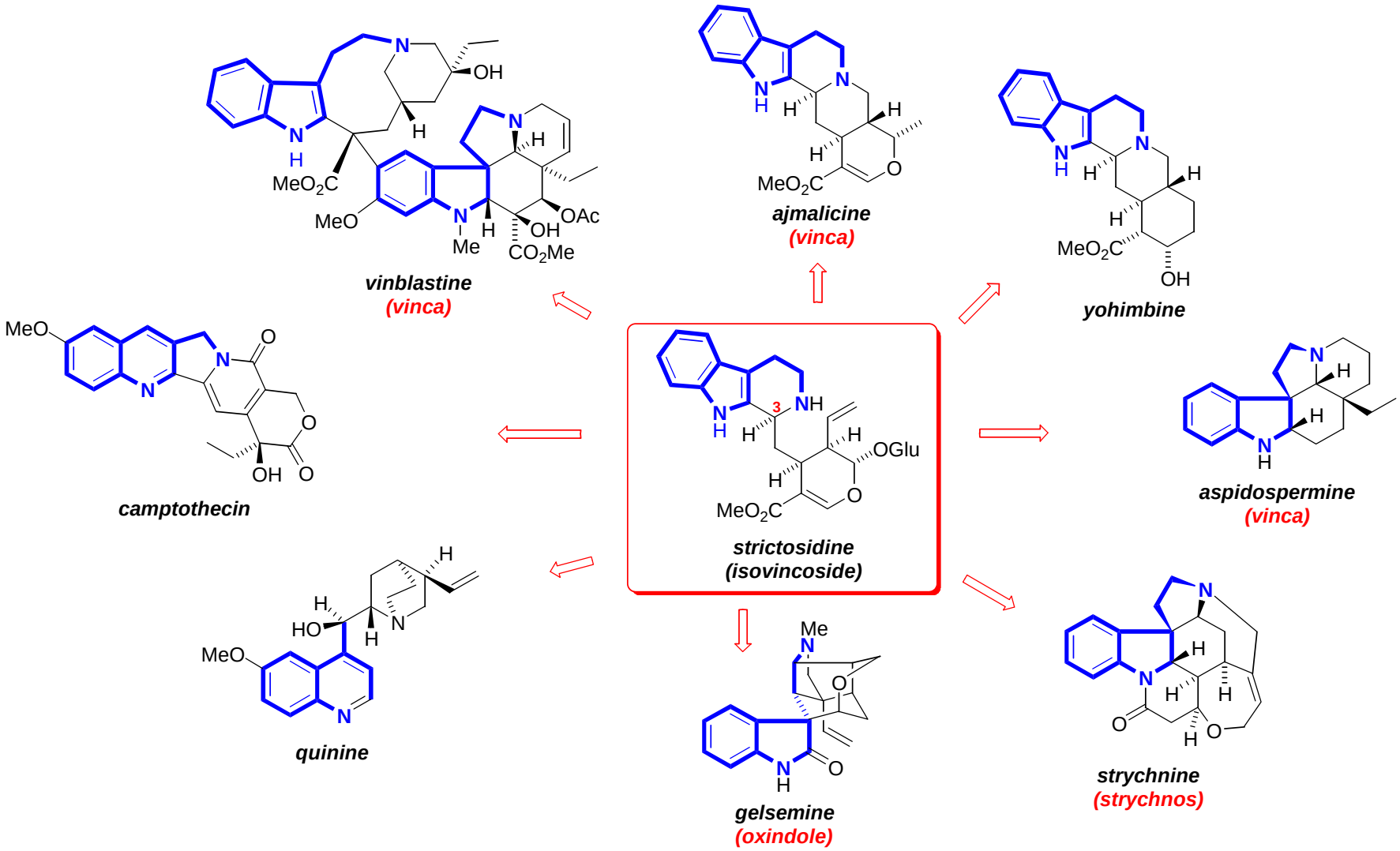


  - ***Mechanism of Pictet-Spengler reaction:***
    - *via* ***spirocyclic*** intermediate then ***Wagner-Meerwein*** 1,2-alkyl shift:



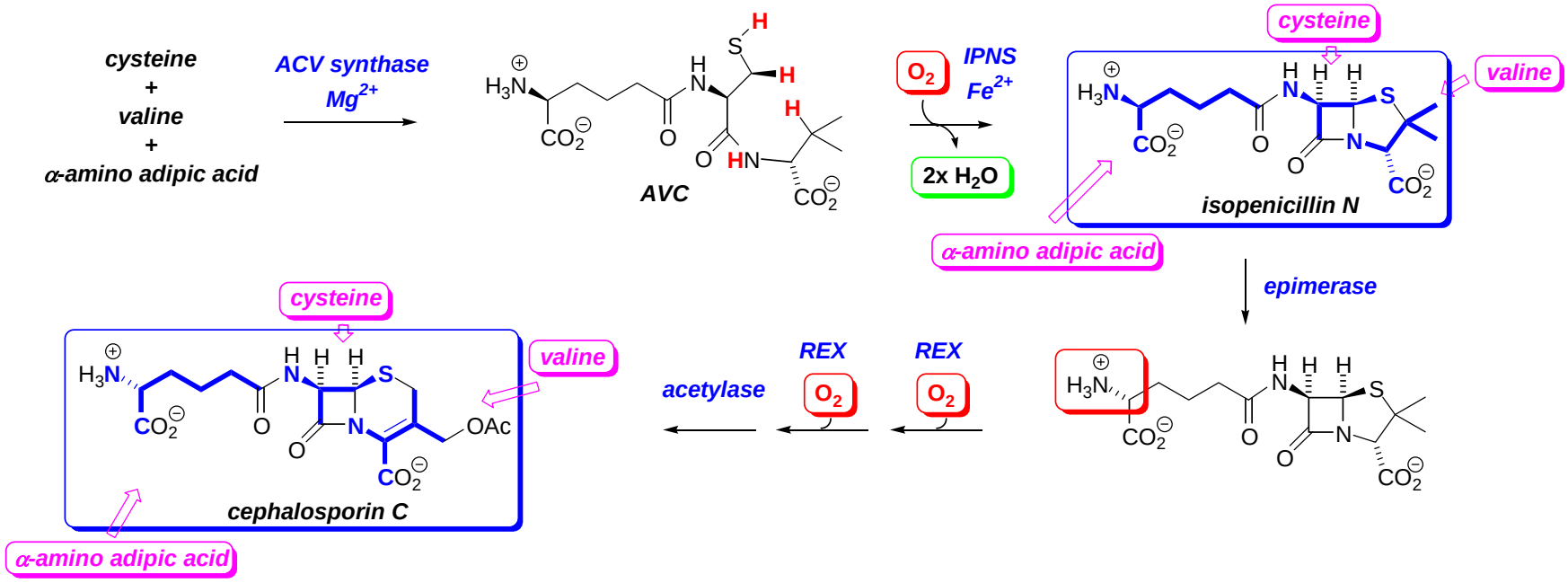
Strictosidine → *Vinca, Strychnos, Quinine etc.*

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



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



Primary Metabolism - *Overview*