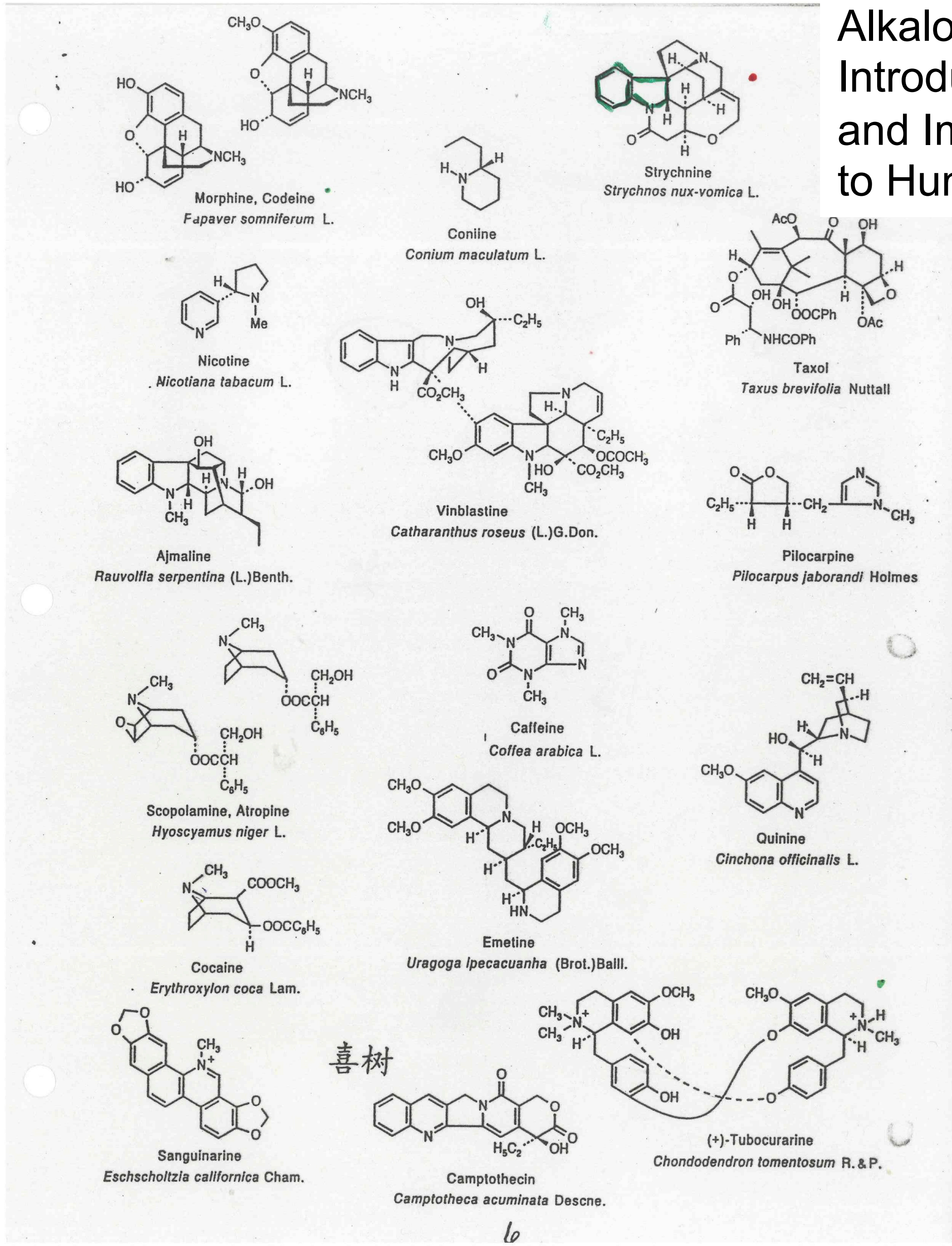
Alkaloids – Introduction and Importance to Humans



Alkaloids:

- many human physiological effects and uses

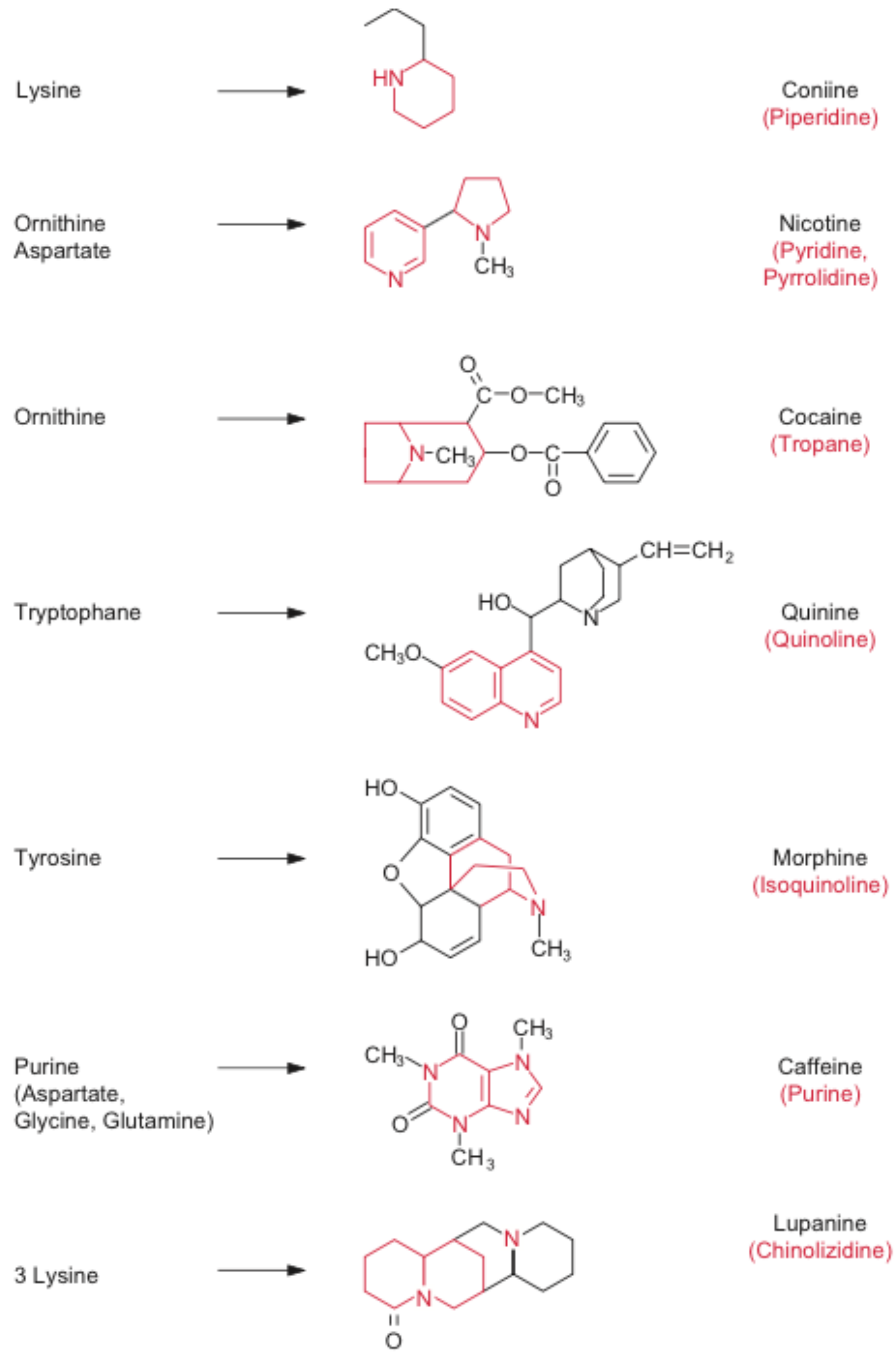
- diverse mechanisms of action

- features and characterisitics

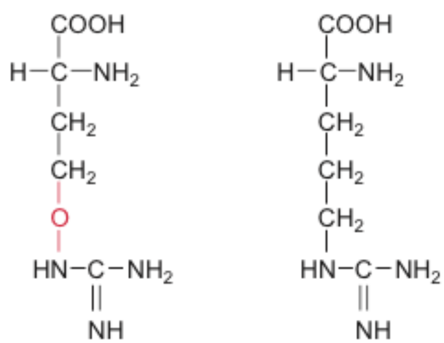
Alkaloid Families and Biosynthesis

(mostly derived from amino acids)

Heldt, Fig 16.1



Non-protein amino acids: –
not considered alkaloids but
also toxins

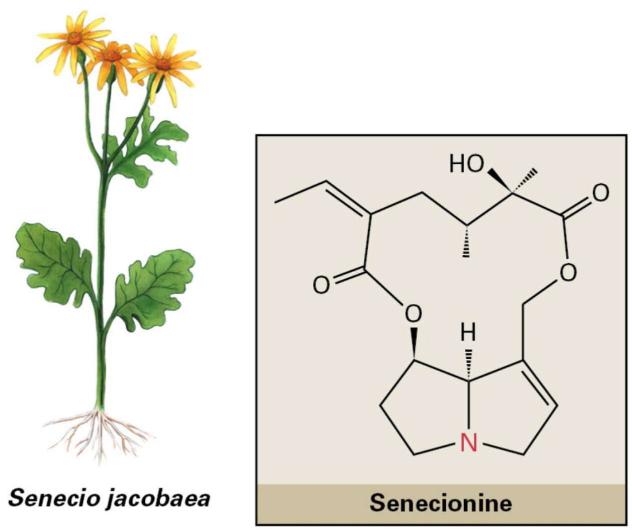


Canavanine   Arginine

Pyrrolizidine Alkaloids (Senecionine N-Oxide) in Senecio

Background

- basic structural type, but many variations
- occurrence in plants in Compositae,
  Boraginacaceae, Orchidaceae
- extremely toxic to both mammals and insects
- documented co-evolution with insects



A **B**

FIGURE 24.35 *Pyrrolizidine and quinolizidine alkaloids. (A) Structure of the pyrrolizidine alkaloid senecionine from ragwort (*Senecio jacobaea*). (B) Structure of the quinolizidine alkaloid lupanine from the bitter lupine* Lupinus polyphyllus. *Lupanine is a bitter compound that functions as a feeding deterrent.*

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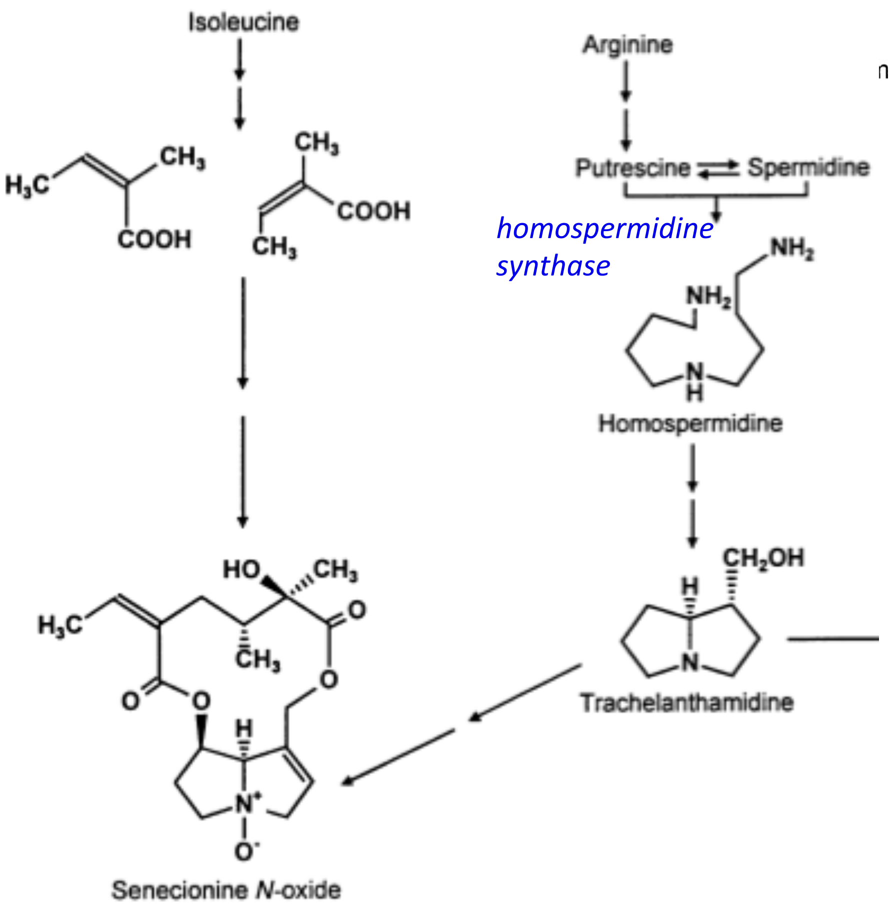
Biosynthesis of Pyrrolizidine Alkaloids (Senecionine N-Oxide) and the Major Structural Types of Pyrrolizidine Alkaloids



B

Biosynthesis

Senecionine-N-oxide synthesis

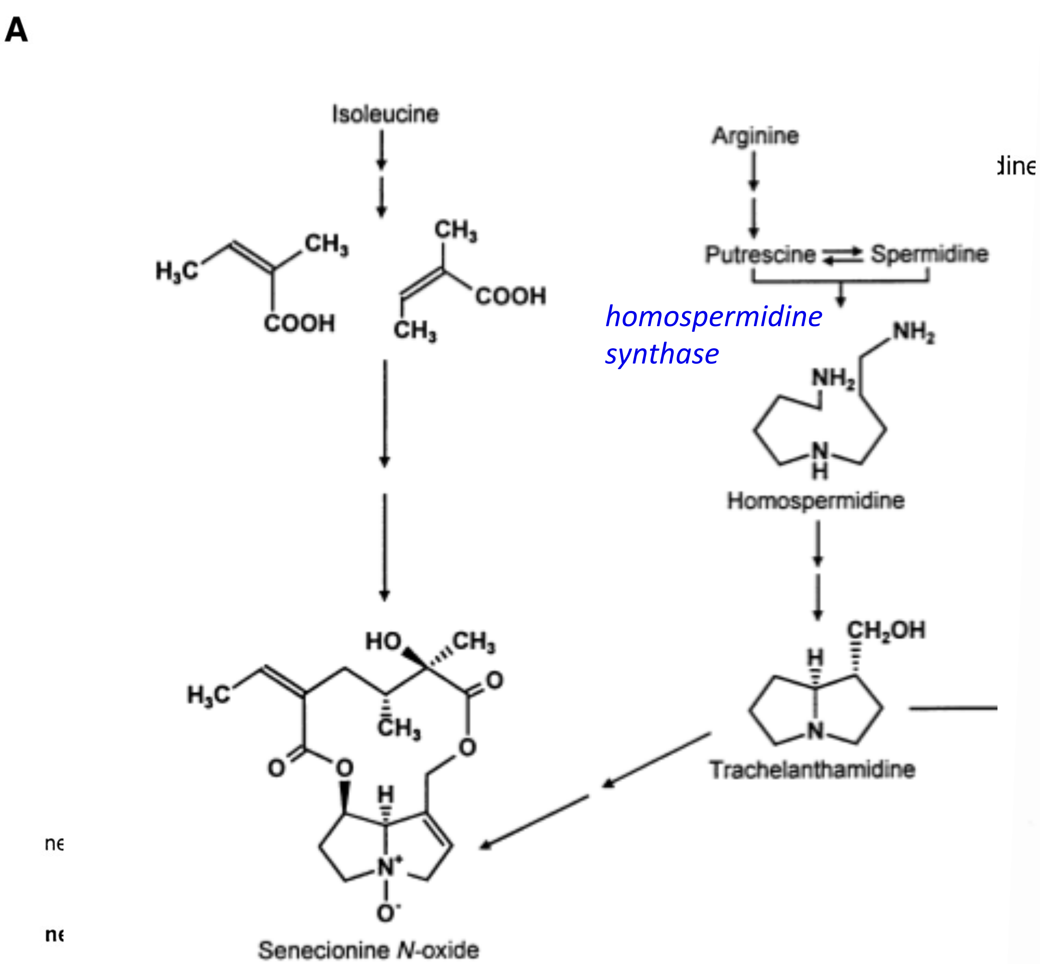
- necine moiety from **arginine** via putrescine

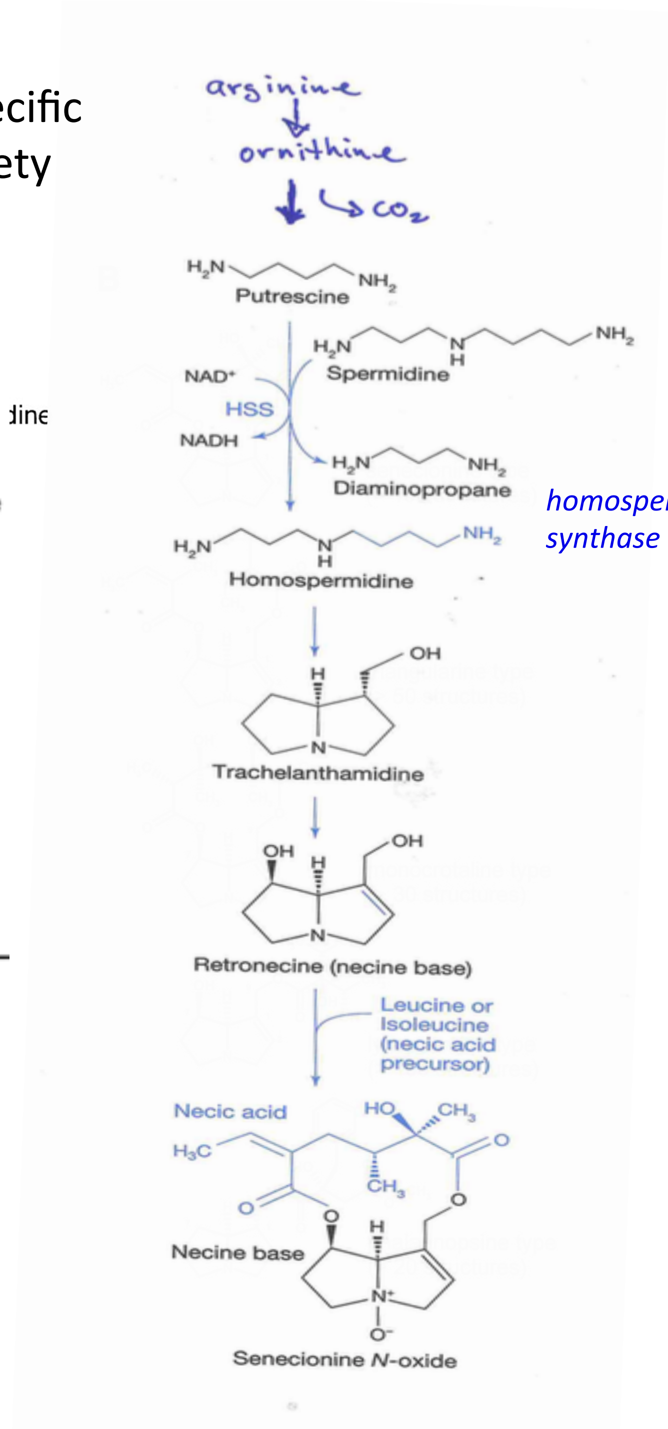
- dicarboxylic acid moiety from i**soleucine**

=> both form the macrocyclic diester

Plant Cell 2004;16:2772-2784; Planta 207:48-495

Homospermidine synthase catalyzes the first specific step of the biosynthesis of the necine base moiety



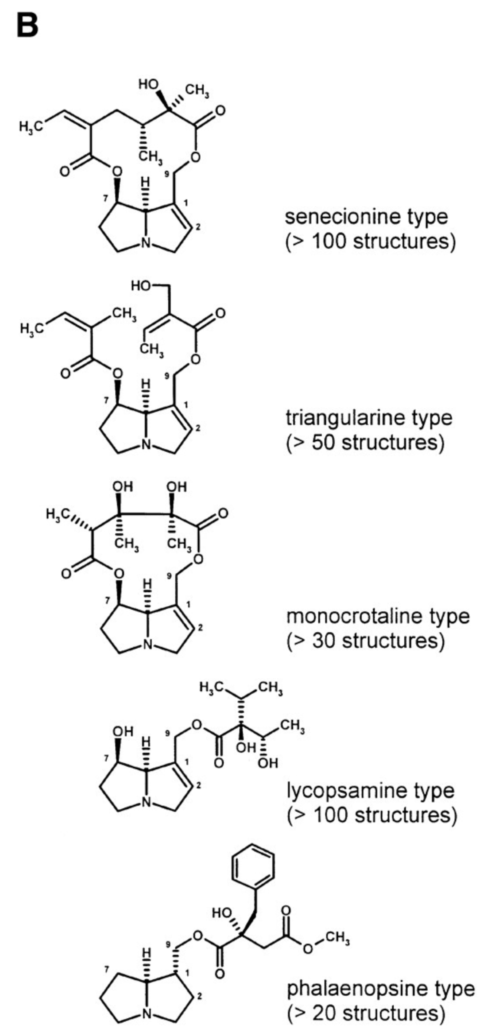


homospermidine synthase

Plant Cell 2004;16:2772-2784; Planta 207:48-495

Five structural types of pyrrolizidine alkaloids (note macrocyclic diester)

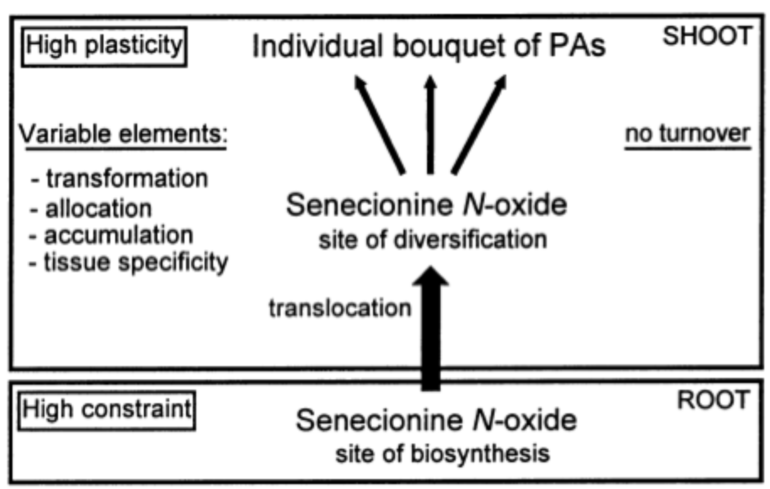
Homospermine synthase evolved independently at least four time - different plant families - from deoxyhypusine synthase, and enzyme of primary metabolism (activates eukaryotic translation initiation factor eIF5A)



Plant Cell 2004;16:2772-2784; Planta 207:48-495

Ecology of Pyrrolizidine Alkaloids

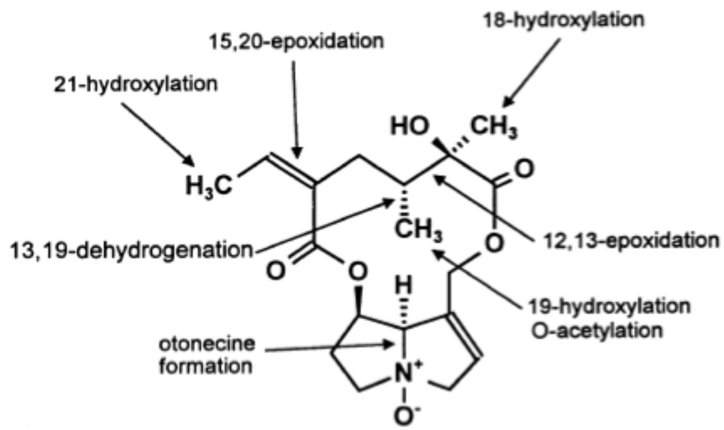
Senecio jacobaea



PA localization and transport

Hartmann, Planta 207:483 (1999)

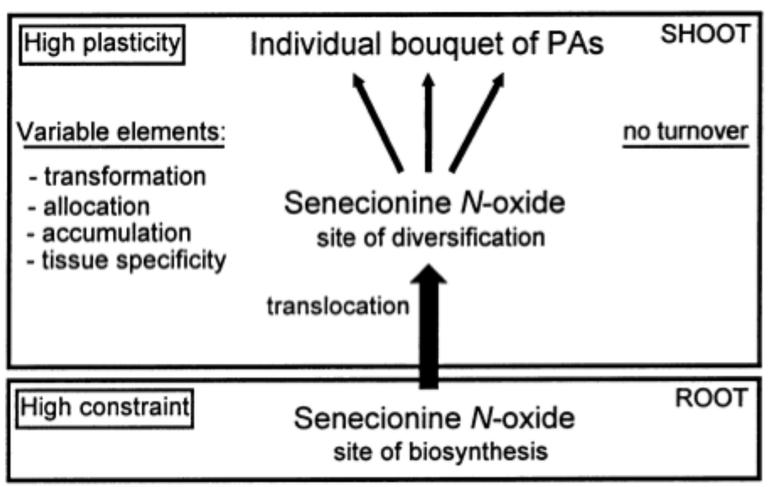
Ecology of Pyrrolizidine Alkaloids



Senecionine *N*-oxide

Fig. 4. Structural diversification of senecionine *N*-oxide documented for five *Senecio* species. The transformations are species-specific; each species produces its own PA bouquet. The transformation products are formed in a position-specific and stereoselective manner

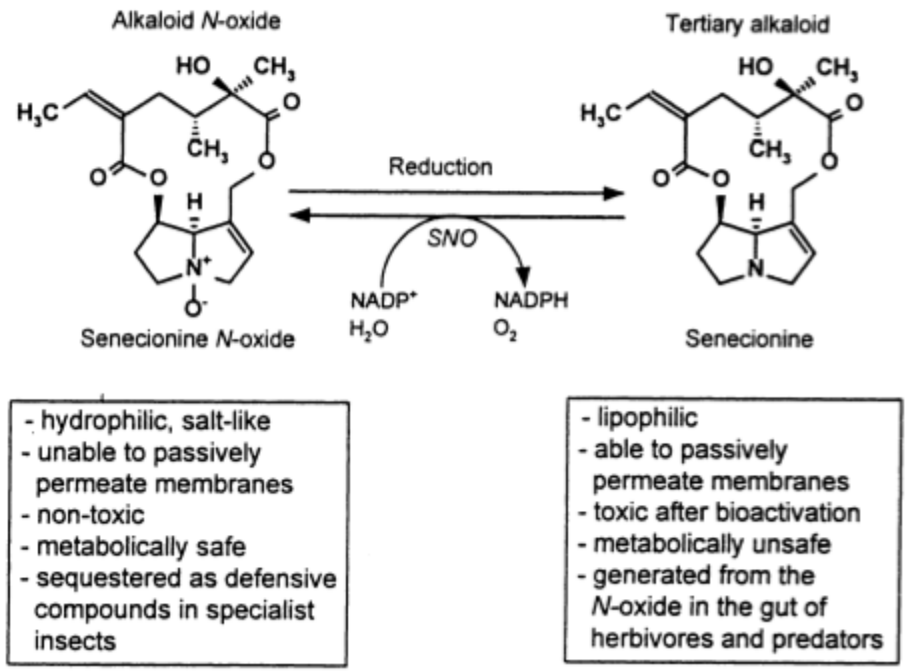
Senecio jacobaea



PA localization
and transport

Hartmann, Planta 207:483 (1999)

Toxicity of Pyrrolizidine Alkaloids and its Avoidance



Hartmann, Planta 207:483 (1999)

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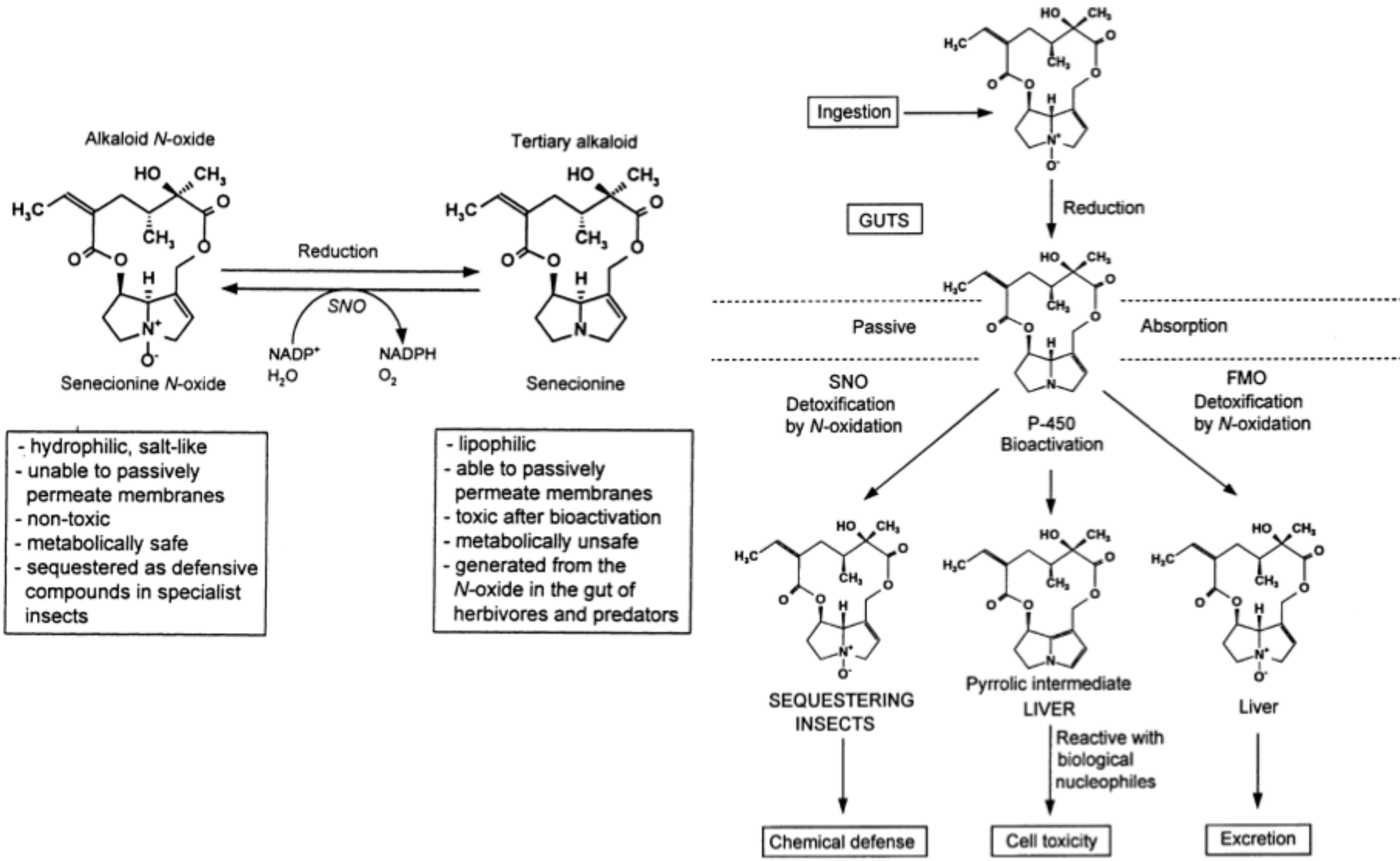


Hartmann, Planta 207:483 (1999)

Toxicity of Pyrrolizidine Alkaloids and its Avoidance

- N**-oxide** forms are stored in plant, these are the less toxic form (see figure)
- after ingestion: N-oxide are reduced to **tertiary amine** in reducing conditions in gut
- in mammals, a liver **cytochrome P450 oxidase** (general detoxification enzymes, xenobiotics) actually activates the toxin by converting to **pyrrolic** compounds (=alkylating agents)
- generalist insects are similarly susceptible to this
- however, specialist insects have a **specific** senecionine N-oxidase that keeps PA in **N-oxide form** and prevents the toxic effects

*Interestingly, guinea pigs are immune to the pyrrolizidine alkaloids. They have **non-specific oxidase** which maintains the PA as the N-oxide form.*

Additional specialist insect adaptations to the pyrrolizidine alkaloids

i) **tolerance** (see above - senecionine N-oxidase)

ii) uptake and sequestration: *(alkaloid-sequestering species are brightly colored)*

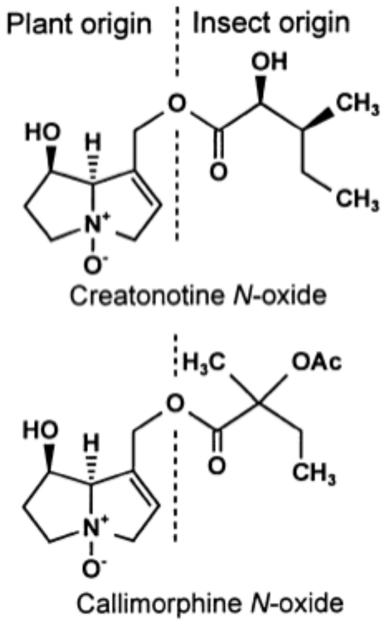
- cinnabar moths tolerate and accumulate PAs (keep in the N-oxide form)
- specialized storage and secretion by *Oreina* beetles (spray predators with PAs)
- in American arctiid moths, the PAs are sequestered, and *retained through metamorphosis* to eggs and sperm.

iii) derivatives used as pheromones

iv) **pharmacophagy** (Danainae butterflies)

Ecology of Pyrrolizidine Alkaloids

Cinnabar moth on Senecio flowers



http://www.aphotofauna.com

hair pencils

Hartmann, Planta 207:483 (1999)

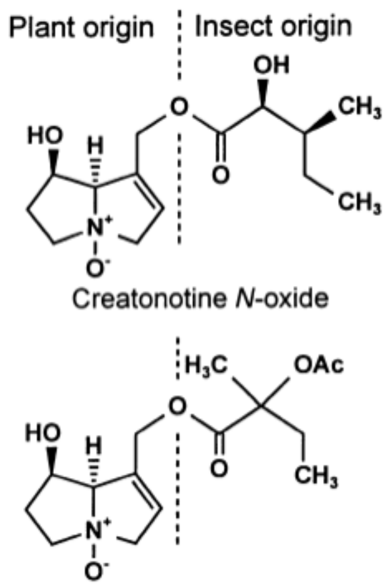
Ecology of Pyrrolizidine Alkaloids

iii) derivatives used as pheromones

hydroxydanaidal

- specialized organs (hair pencils) of male as a **courtship pheromone**

= PA content of male signaled via hydroxydanaidal



Creatonotine *N*-oxide

Callimorphine *N*-oxide

Hartmann, Planta 207:483 (1999)

Cinnabar moth on Senecio flowers

http://www.aphotofauna.com

hair pencils

Summary: Levels of adaptation of specialist insects to pyrrolizidine alkaloids (plant toxins)

i) tolerance by maintaining as N-oxides

ii) sequestration

iii) packaging PAs with eggs and sperm for protection

iv) pharmacophagy by adult butterflies to obtain PAs

Ecology of Nicotine Alkaloids

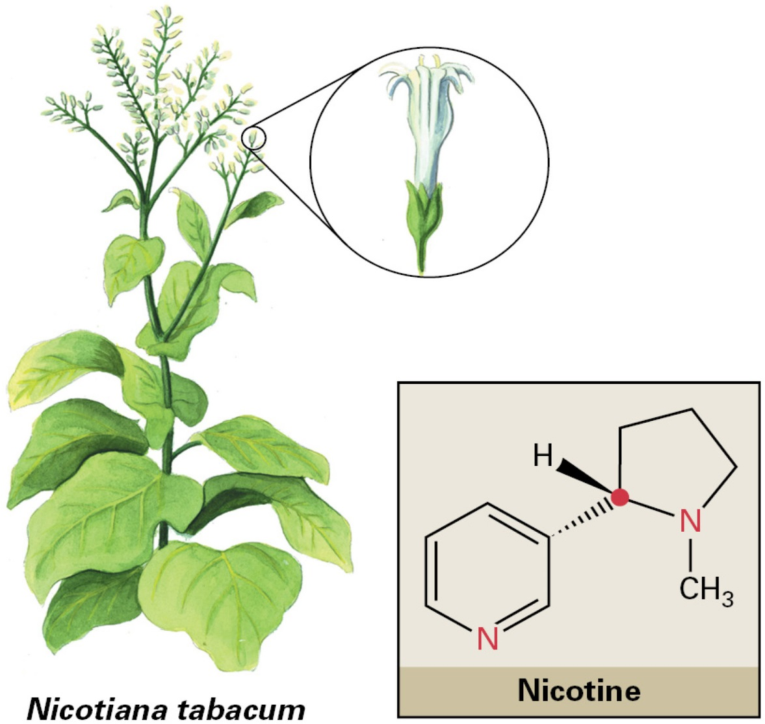


FIGURE 24.32 *Structure of nicotine from* Nicotiana tabacum.

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Ecology of Nicotine Alkaloids

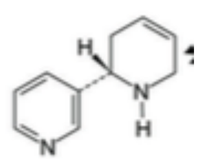
- very small structural family: nicotine, nornicotine, anatabine

- occurrence: Nicotiana *spp*. (flowers, seeds, young leaves)

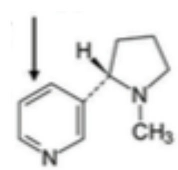
- highly toxic to mammals and insects, binds and stimulates *cholinergic synapses*

- in humans causes: "nausea, vomiting, diarrhea, mental confusion, convulsions, respiratory paralysis "

    - highly addictive (interacts w. *dopamine*)

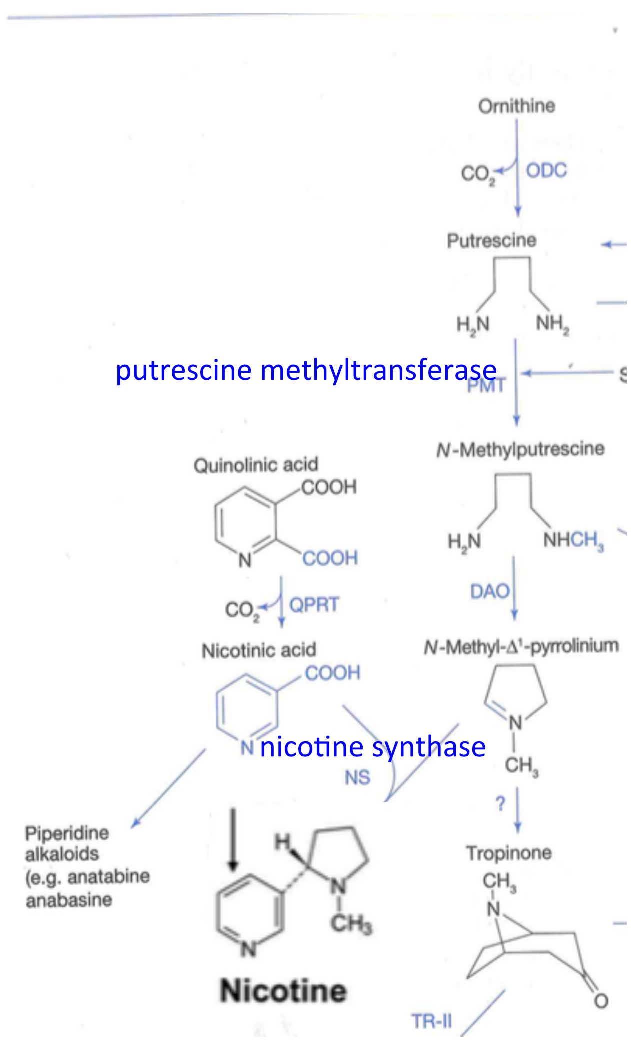


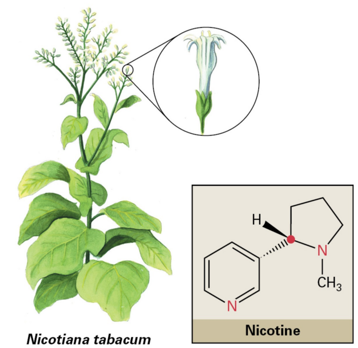
Anatabine



Nicotine

Synthesis of Nicotine Alkaloids





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Biosynthesis

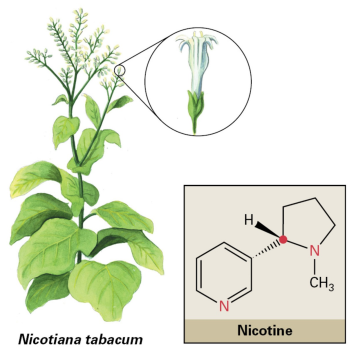
Two ring structure with

- nicotinic acid (pyrinidine)
- pyrrole (furan ring) from arg
- note: decarboxylation first
- *putrescine methyltransferase (PMT),*
- *nicotine synthase*
- synthesis occurs root

Synthesis of Nicotine Alkaloids

Nicotine synthesis is induced by wounding

- this is somewhat unusual for an alkaloid
- 'topping' of commercial tobacco plantations is known to increase nicotine synthesis
- putrescine methyltransferase is wound-induced in root. → Defensive function (?)
- a good example of **systemic signaling**. It is now known to require *methyl jasmonate*



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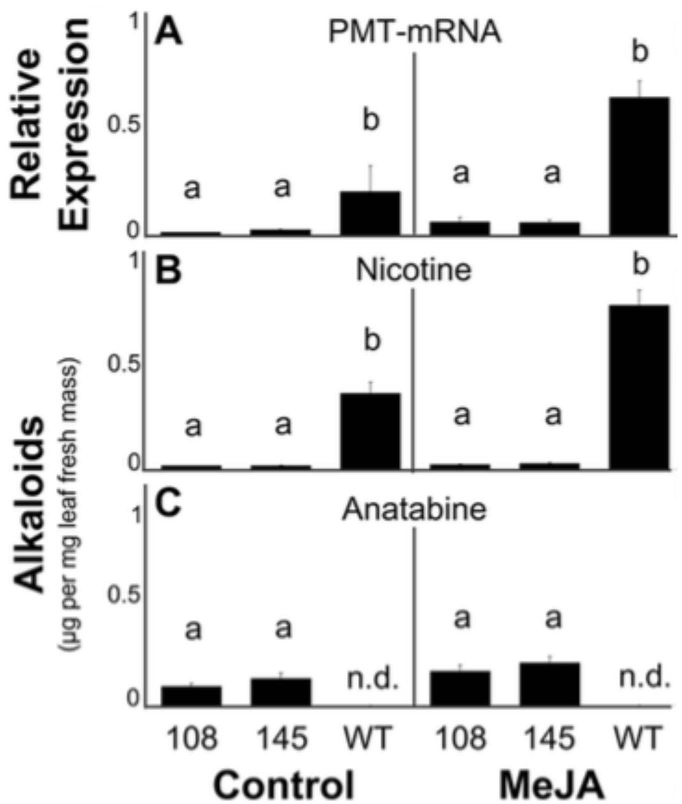
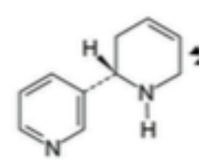
Chemical Ecology of Nicotine Alkaloids



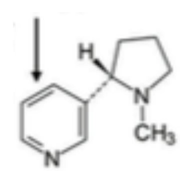
Reduction in nicotine via RNAi

Steppuhn & Baldwin,PLoS 2: e217 (2004)

- both constitutive and induced nicotine levels are affected.

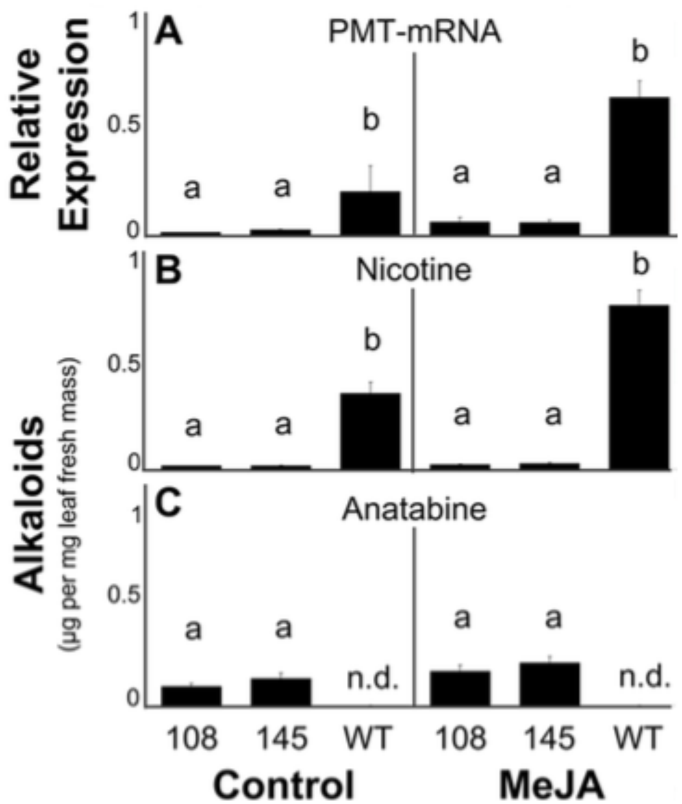


Anatabine



Nicotine

Chemical Ecology of Nicotine Alkaloids



Reduction in nicotine via RNAi or antisense

Steppuhn & Baldwin,PLoS 2: e217 (2004)

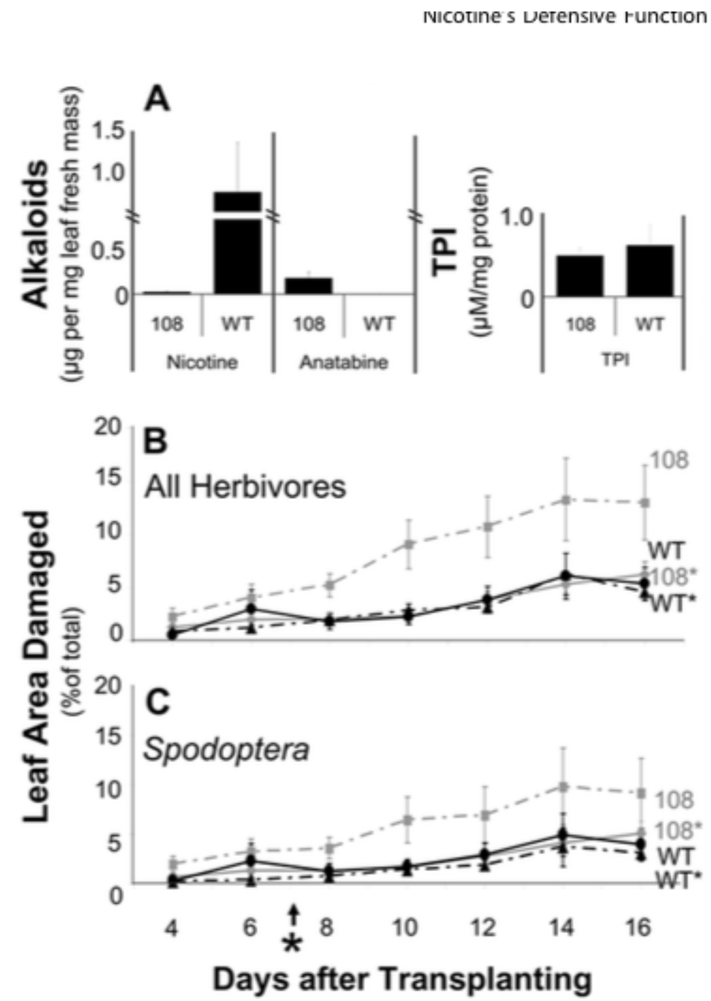
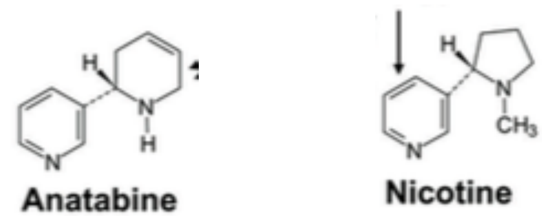


Figure 4. Herbivore Damage to IR*pmt* and WT *N. attenuata* Plants in Natu



List experimental evidence of nicotine for defense function against insect pests

- wounding induces greater nicotine accumulation
- RNAi-mediated suppression & lower nicotine leads to faster growth of insect pests

Interesting counteradaptation by specialist insects

i) *Manduca sexta* (tobacco hornworm) tolerates otherwise lethal doses of nicotine (metabolism / excretion)

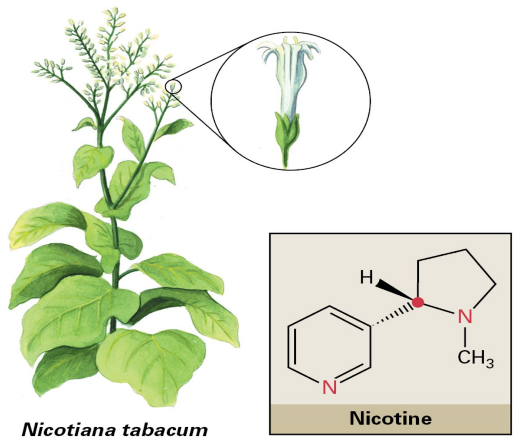
- larvae can at least partially suppresses *nicotine induction* during feeding
- nicotine in larvae helps defend against parasitoid wasp predators of *Manduca*

ii) Nicotine in nectar prevents overconsumption and promotes shorter feeding times (data not shown)

iii) Nicotine is used by tobacco hornworm to repel predators (Kumar et al., PNAS 111: 1245 (2014)

Parasitoid pupae on lepidopteran pest

Ecology of Nicotine Alkaloids
(*Nicotiana attenuata* system, Ian Baldwin)



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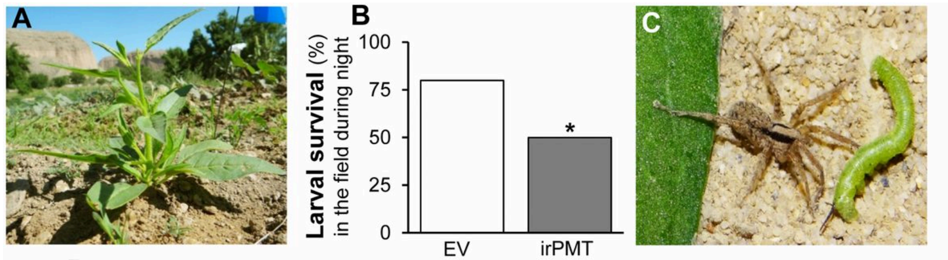
Field study site Utah
(Max Planck Institute for Chemical Ecology, Jena)

Wolf spiders are deterred by nicotine-fed Manduca larvae.



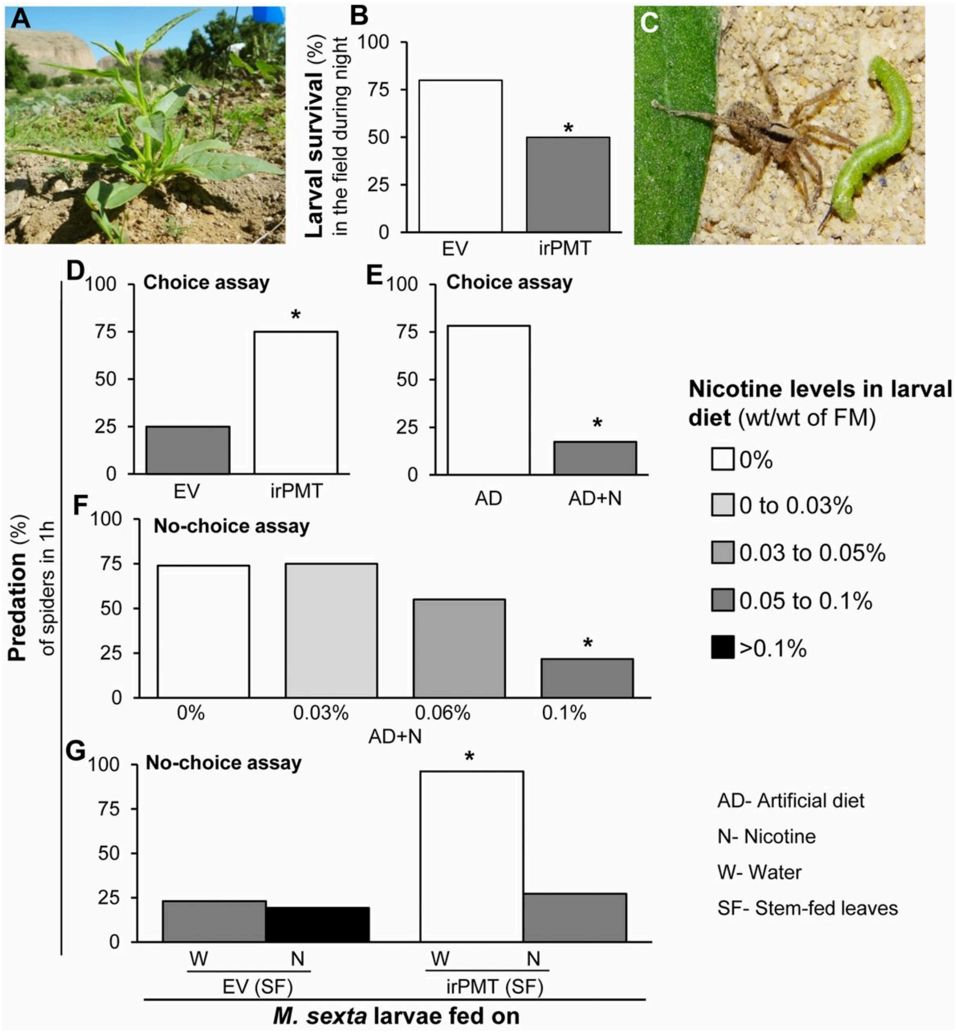
https://www.youtube.com/watch?v=5RYidP4PROQ

https://www.youtube.com/watch?v=5U_OL8IGnBs

Pavan Kumar et al. PNAS 2014;111:1245-1252

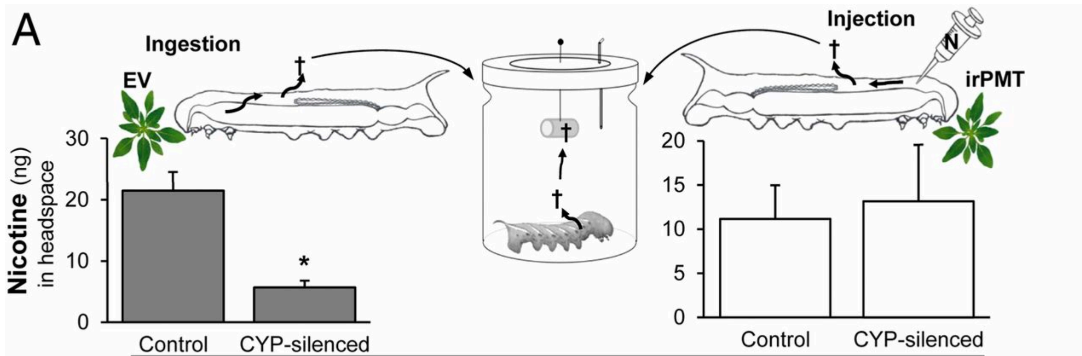
PNAS

Wolf spiders are deterred by Manduca larvae fed on high and low nicotine leaves(EV vs irPMT) or artificial diet (AD +/- N)



Pavan Kumar et al. PNAS 2014;111:1245-1252

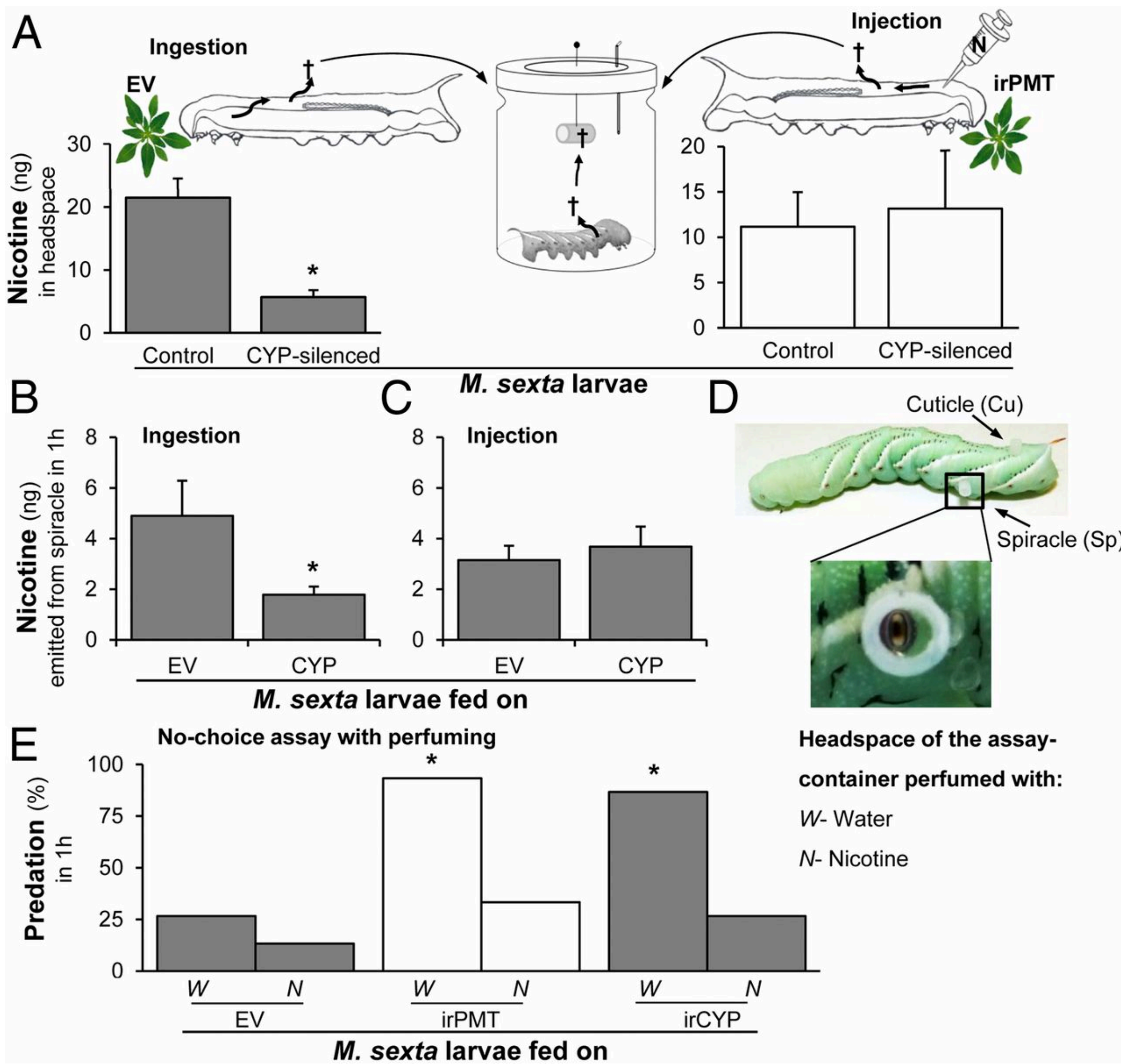
CYP6B46 silencing in transgenic N. attenuata reduces larval nicotine emission and increases spider predation



Why inject larvae with nicotine?

Pavan Kumar et al. PNAS 2014;111:1245-1252

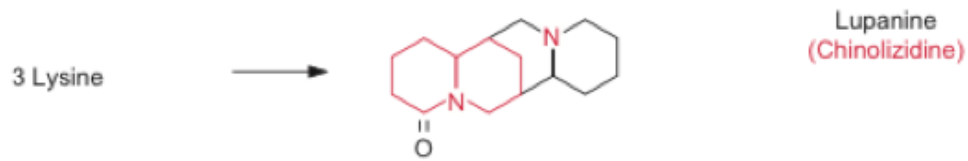
CYP6B46 silencing in transgenic N. attenuata reduces larval **nicotine emission** and increases spider predation



Pavan Kumar et al. PNAS 2014;111:1245-1252

iii) Nicotine is used by tobacco hornworm to repel wolf spiders *(Kumar et al., PNAS 111: 1245 (2014)*

- larvae feeding on low nicotine N. attenuata (PMT-RNAi silenced) are preferred by nocturnal spiders
- observations and loss of nicotine suggest that **nicotine is emitted** from **spiracles** of larvae ("***defensive halitosis"***)
- larvae feeding on plants expressing a **Manduca CYPB46 RNAi gene** show reduced nicotine emission



Example 3. Quinolizidine alkaloids (lupines - *Lupinus* sp.) [lupanine]

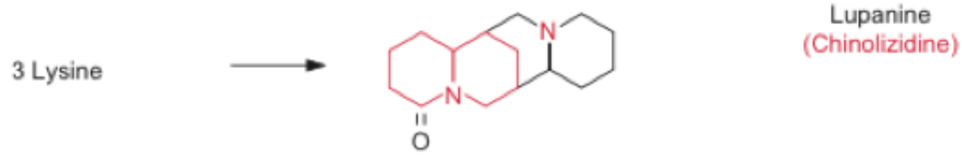
Lupines (legume family) are common native species, highly toxic due to alkaloids, only eaten by herbivores if starving.

- strong biological effects in mammals: "loss of coordination, convulsions, liver damage, .. crooked calf disease (teratogen)"

- cattle poisoning, especially in autumn, from high alkaloid seeds of lupines on range.

- wild hares learn to avoid high-alkaloid lupine (feeding deterrent)

Quinolizidine Alkaloid-Containing Plants



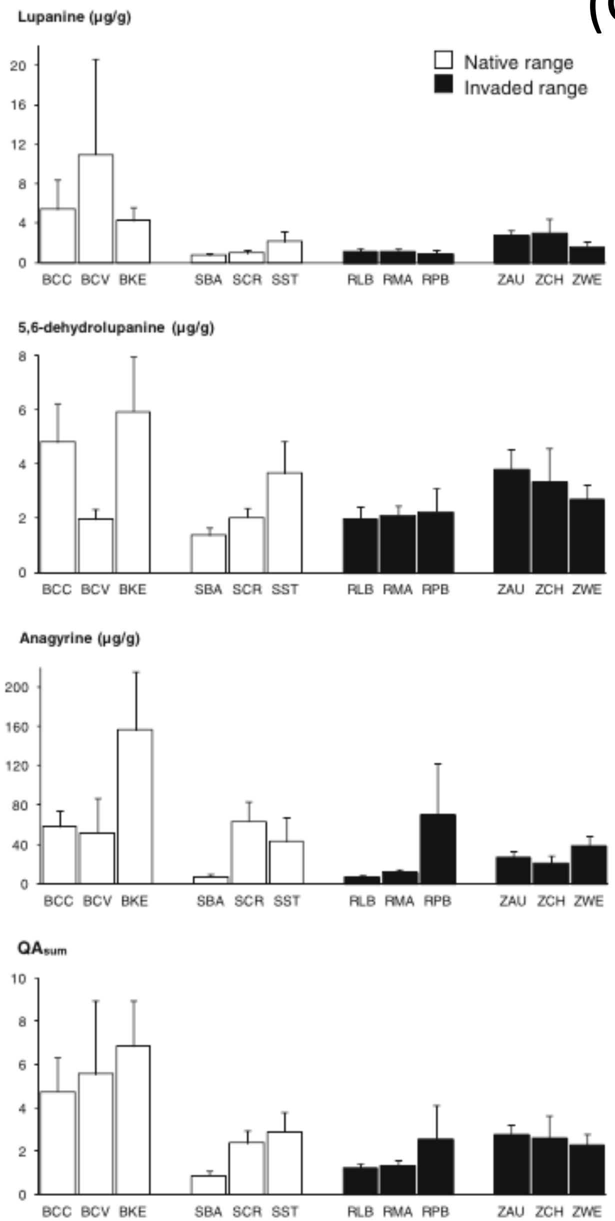
Castilleja miniata
scarlet paintbrush, common red paintbrush
Scrophulariaceae
[root parasite, accumulates lupanine from Lupinus]

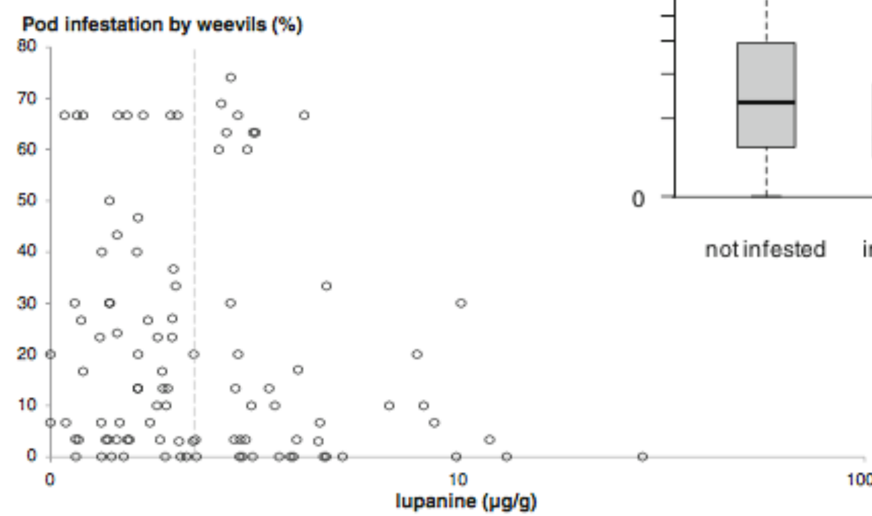
Quinolizidine Alkaloid-Containing Plants

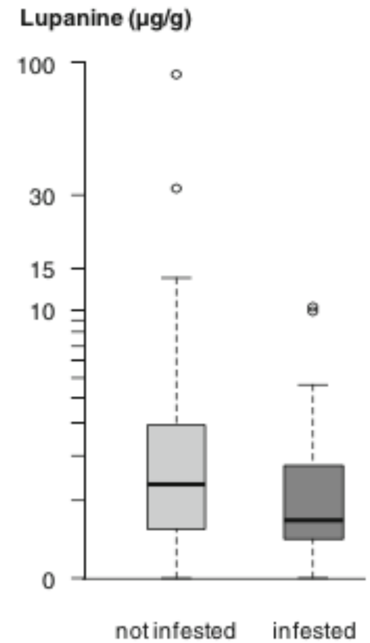
Laburnum anagyroides Medic
Golden chain tree
Fabaceae
(non-native, ornamental)
[Cytisin]

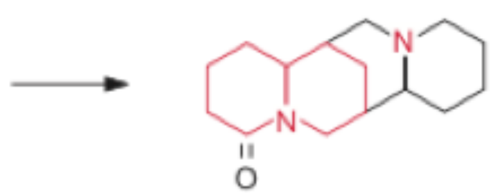
Cytisus scoparius
Scotch broom
Fabaceae
(non-native, invasive)
[Spartein]

Ecology of Lupanine (Quinolizidine Alkaloid)
(Gorse – invasive legume)









Lupanine
(Chinolizidine)

Hornoy et al., Naturwissentsch. 99:8 83 (2012)

4. Solanine and Potato Steroid Alkaloids (Human chemical ecology)

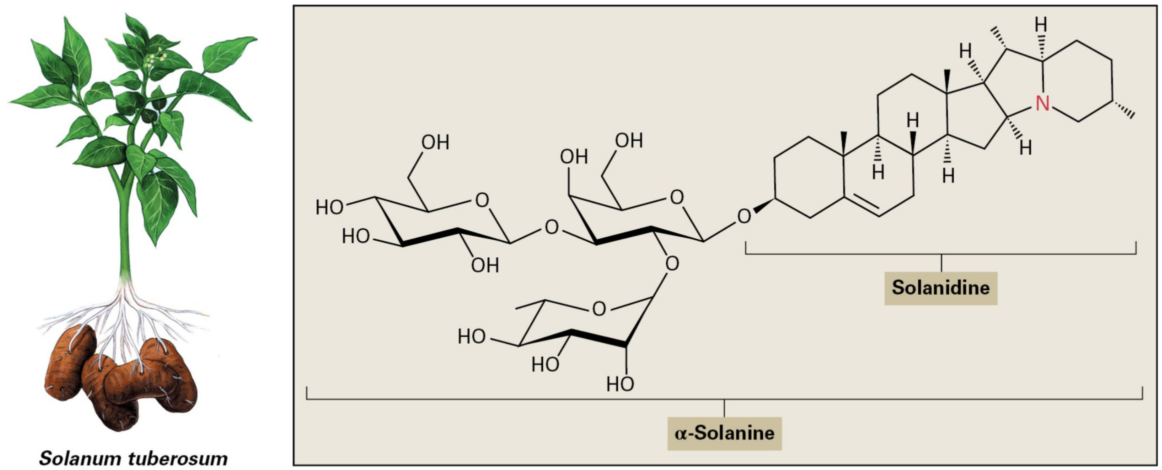


FIGURE 24.34 *Structure of the steroid alkaloid glycoside α-solanine from* Solanum tuberosum *(potato). The aglycone solanidine is derived from cholesterol.*

Buchanan, 2015

solanidine (tripterpene alkaloid of potato)

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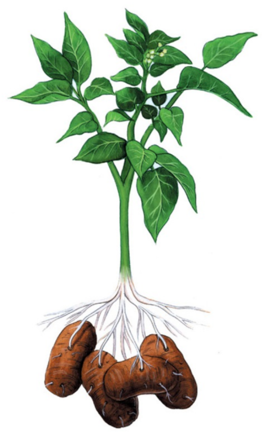
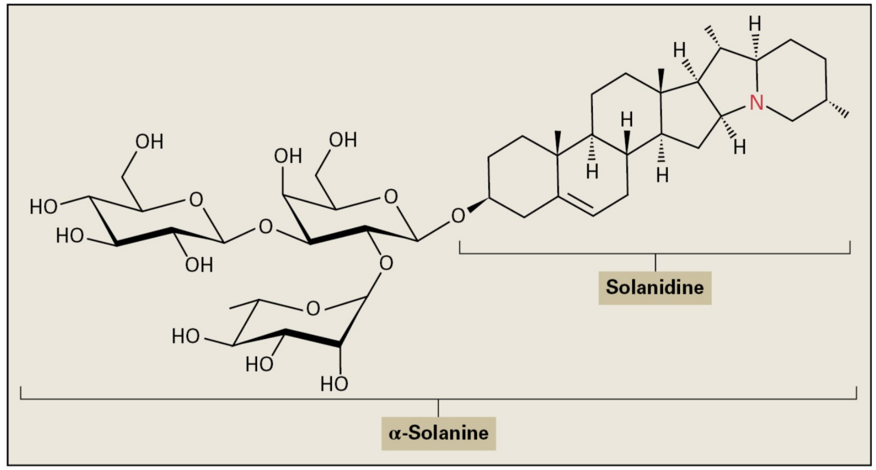


Solanum tuberosum



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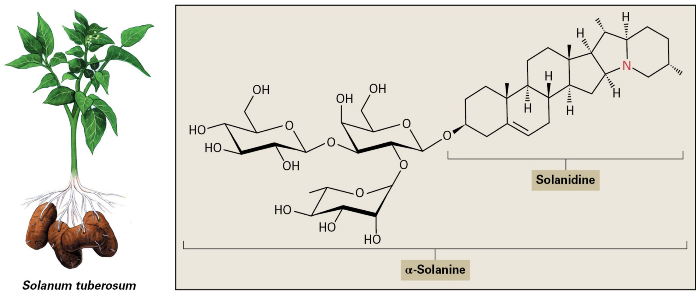
Biological effects:

- can lead to respiratory failure in humans (mammalian defense?)
- antifungal agent in potato and tomato, also toxic to Colorado potato beetle (anti-herbivory)
- low levels in commercial potatoes, often found in leaves, flowers, fruit

4. Solanine and Potato Steroid Alkaloids (Human chemical ecology)



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International Potato Center photo

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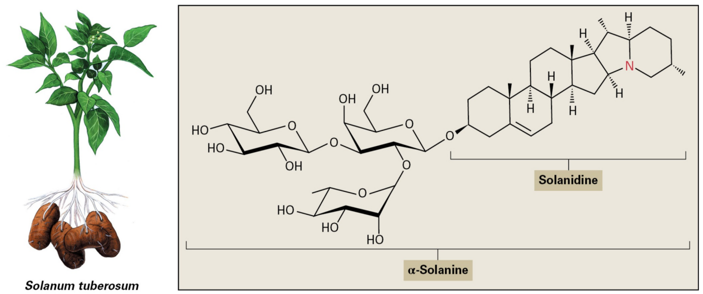

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International Potato Center photo

Journal of Chemical Ecology, Vol. 12, No. 3, 1986

DETOXIFICATION FUNCTION OF GEOPHAGY AND DOMESTICATION OF THE POTATO

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Abstract—Detoxification as the adaptive function of geophagy is demonstrated from field and historical data associating clay consumption with the domestication of potentially toxic potatoes. In vitro analyses showed that the glycoalkaloid, tomatine, was effectively adsorbed by four classes of edible clays over a range of simulated gastrointestinal conditions. These results, in conjunction with reports of geophagy by nonhuman primates, suggest geophagy as a solution to the impasse chemical deterrents pose to the process of domestication and to chemical constraints on plant exploitation by non-fire-using hominids. The inorganic component of the chemical environment deserves increased attention from chemical ecologists.

Key Words—Geophagy, hominid, primate, detoxification, glycoalkaloids, tomatine, domestication, potatoes, clay–organic interactions.

Implications for human diet:

- solanidin is found in edible potatoes at very levels, so it doesn't have discernable effects.
- synthesis induced by greening (triggered by light)
- wild potato species (Peru, Bolivia) contain high levels of these compounds. Commercial varieties were selected for lower levels (at the risk of greater disease or herbivory)

Geophagy (eating of soil or clay) = binding of alkaloids by special clays

- detoxification and removal of alkaloids.
- effectiveness can be shown in lab experiments (binding to clay)

Concluding thoughts on alkaloids

- there are many very toxic plant chemicals (ie alkaloids) in common plant species
- toxic chemicals are present in many food plants, and in nature
- plant toxins are effective against generalist insects or herbivores, but specialists have evolved counter-adaptations
- alkaloids are part of evolved chemical warfare systems between plants and insects (of different trophic levels)

Study Question

Describe the evidence (using specific examples and experiments) that demonstrates the importance of alkaloids in plant-insect interactions or their effectiveness in defense

[Think of different types of arguments you could use, in terms of different adaptations, that demonstrate the efffectiveness of alkaloids as protective agents]