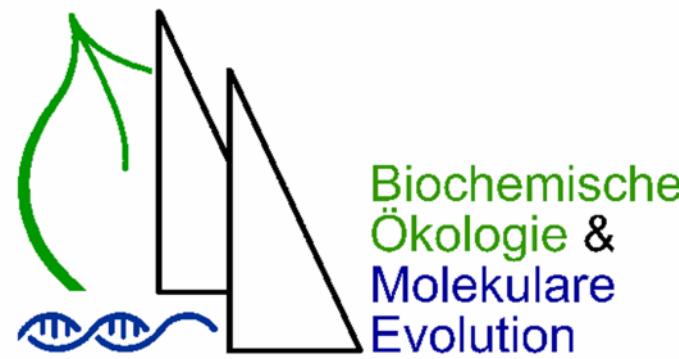




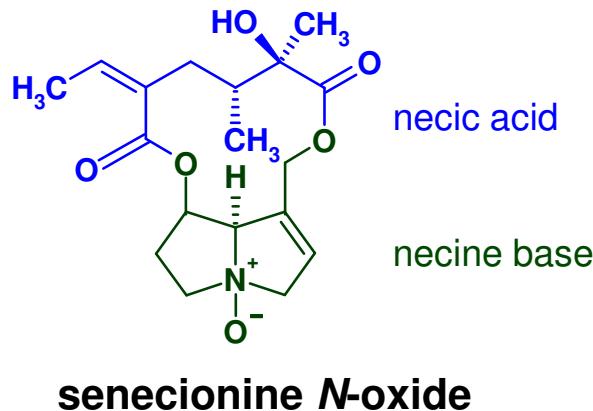
Vorkommen und Verbreitung von Pyrrolizidinalkaloiden in Pflanzen



Dietrich Ober

Botanisches Institut und Botanischer Garten
Universität Kiel

The Model System: Pyrrolizidine Alkaloids (PAs)



- **ester alkaloids**, constitutively produced
- in most cases **toxic** (to vertebrates and insects)
- part of fascinating **interactions** between plants and specialized insects

Our motivation:

- understanding the function and evolution of
 - PA biosynthesis in **plants**
 - of specific adaptations to PAs by **insects**



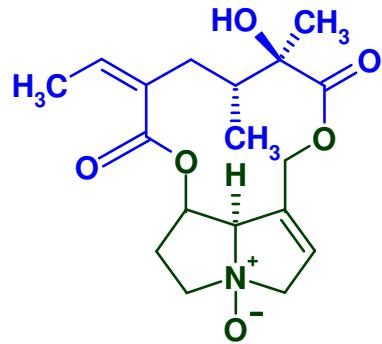
Tyria jacobaeae at the Kiel fjord

© Dietrich Ober



© Dietrich Ober

Insects with specific adaptations to PA-containing plants



Alkaloid accumulation in the insect

Warning coloration of the insect

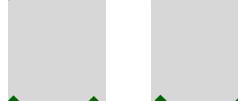
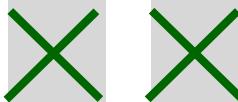
Alkaloid-based pheromones

PA-dependent morphology

PA-dep. behaviour

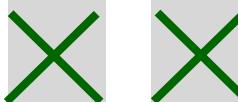
Beetles (Coleoptera)

larvae of certain leaf beetles



Grasshoppers (Orthoptera)

e.g. West-african *Zonocerus*



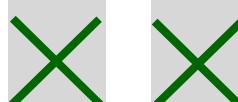
Aphids

certain species



Lepidoptera

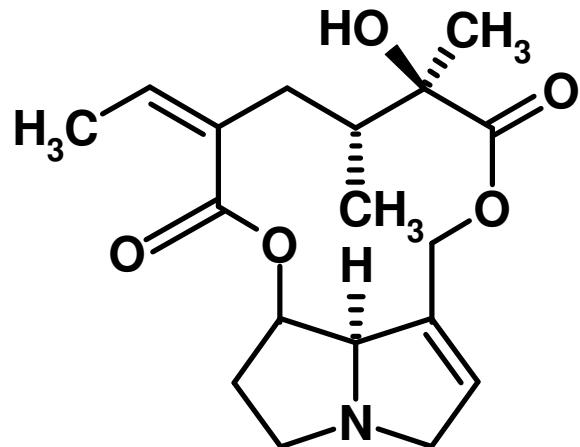
various moths and butterflies



Toxic principle of pyrrolizidine alkaloids

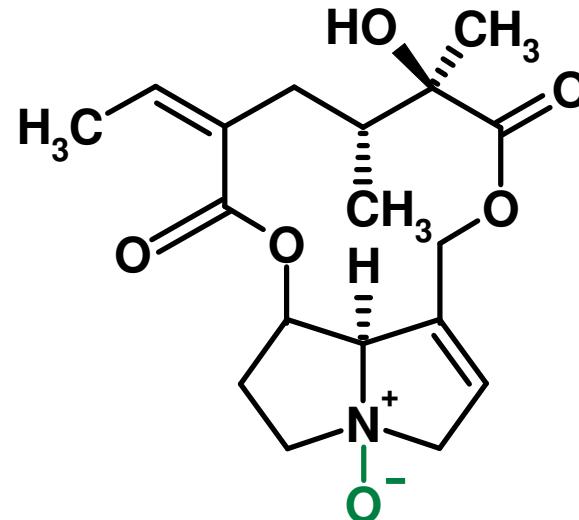


The two faces of pyrrolizidine alkaloids



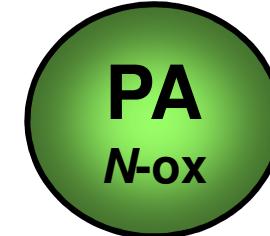
tertiary PA

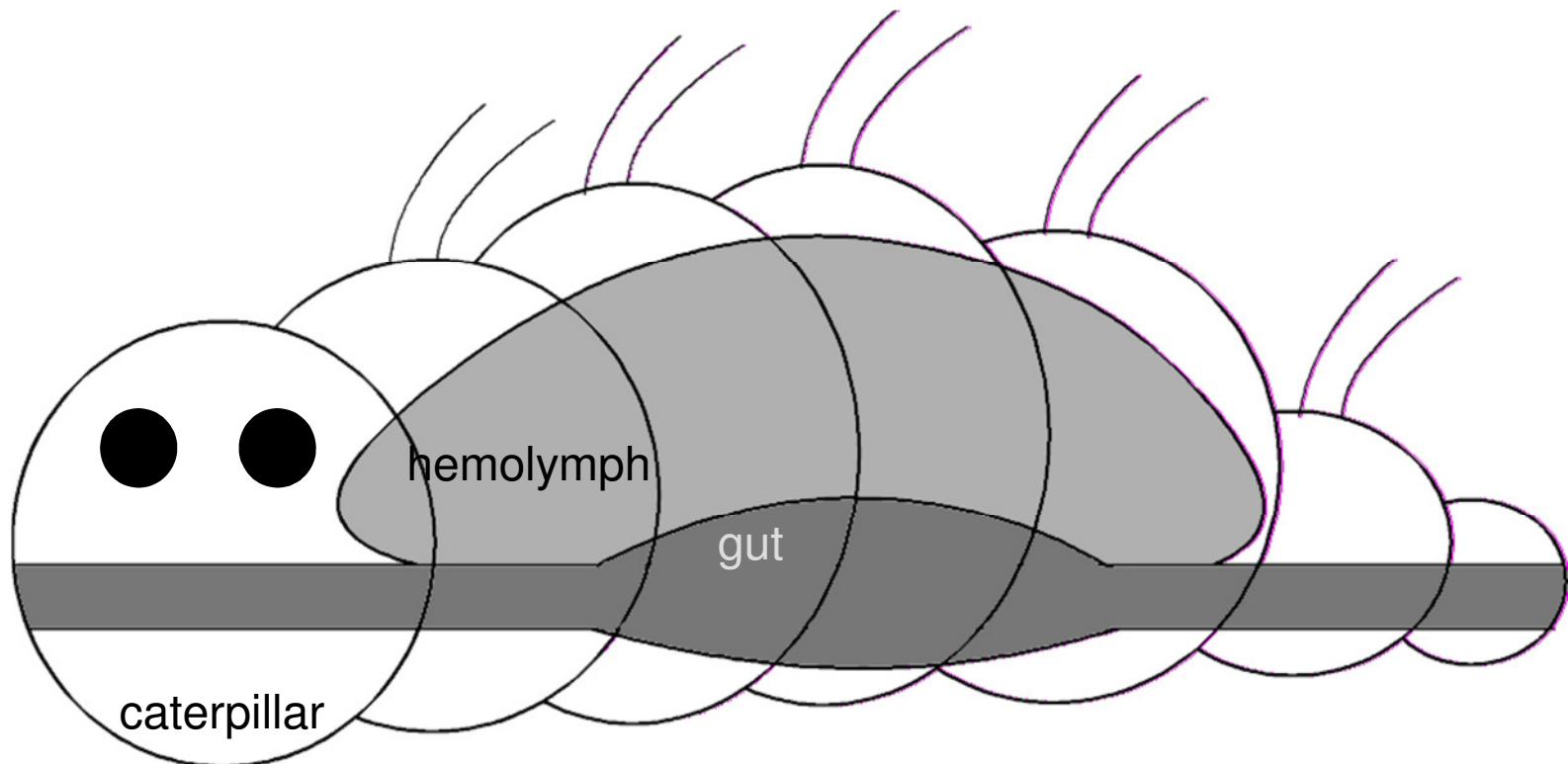
- lipophilic
- toxic after bioactivation



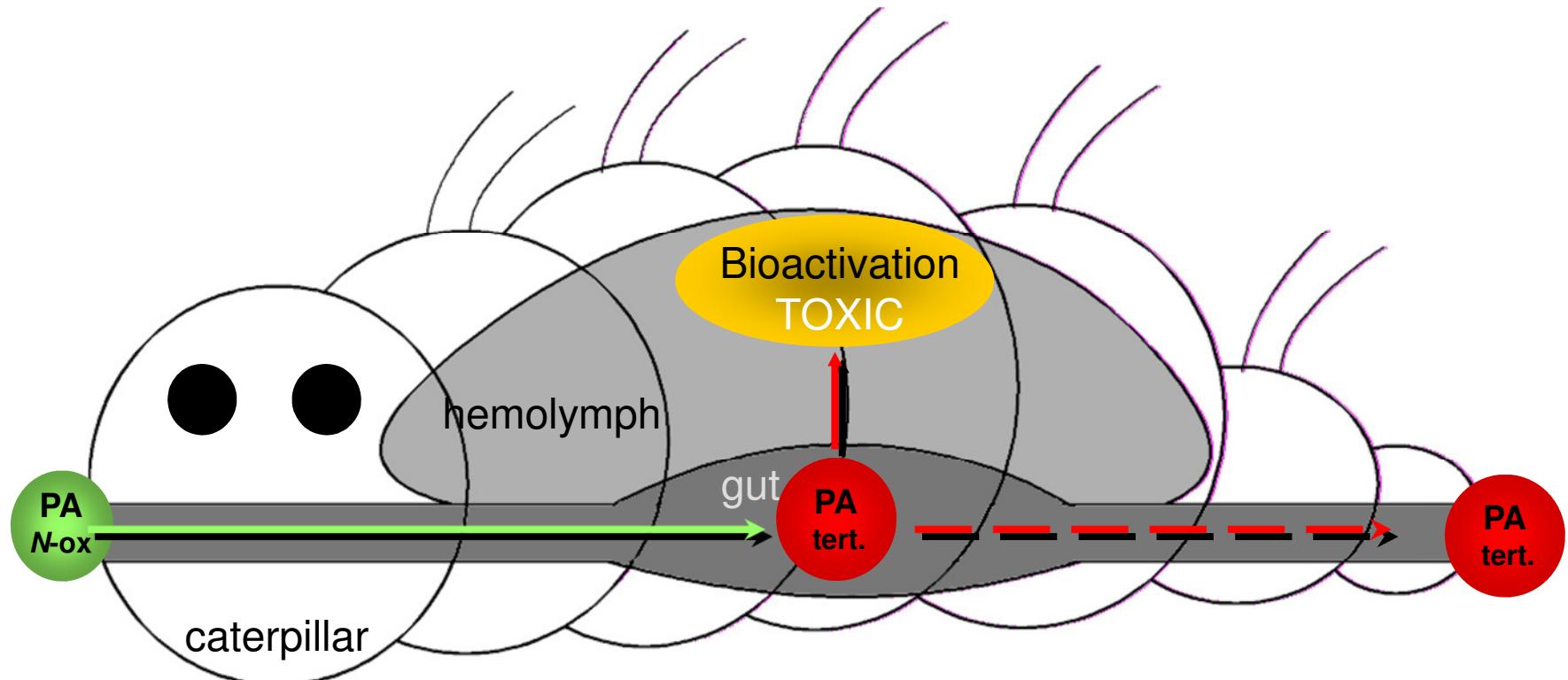
PA N-oxide

- hydrophilic
- nontoxic





Toxic principle of pyrrolizidine alkaloids in a nonadapted herbivore



- unspecific reduction
 - partial excretion
 - passive resorption
- bioactivation by cyt.P450s in hemolymph



Sequestration of toxic pyrrolizidine alkaloids



© Linzhu Wang

Orthoptera

(order of grasshoppers, locusts, and crickets)

genus: *Zonocerus*

e. g. *Zonocerus variegatus*



© Dietrich Ober

Lepidoptera

(order of butterflies and moths)

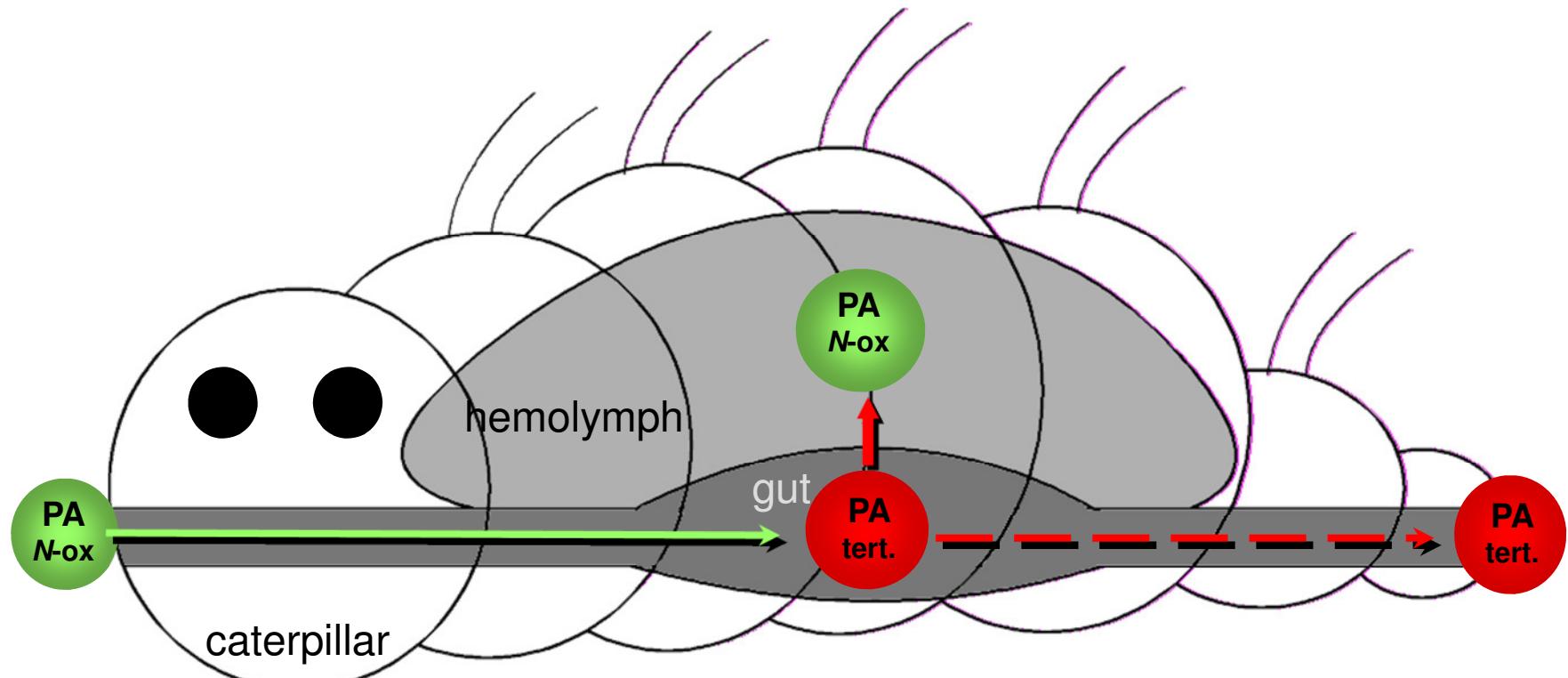
family: Arctiidae (certain moths)

e. g. *Tyria jacobaeae*

family: Nymphalidae (butterflies)

© Dietrich Ober

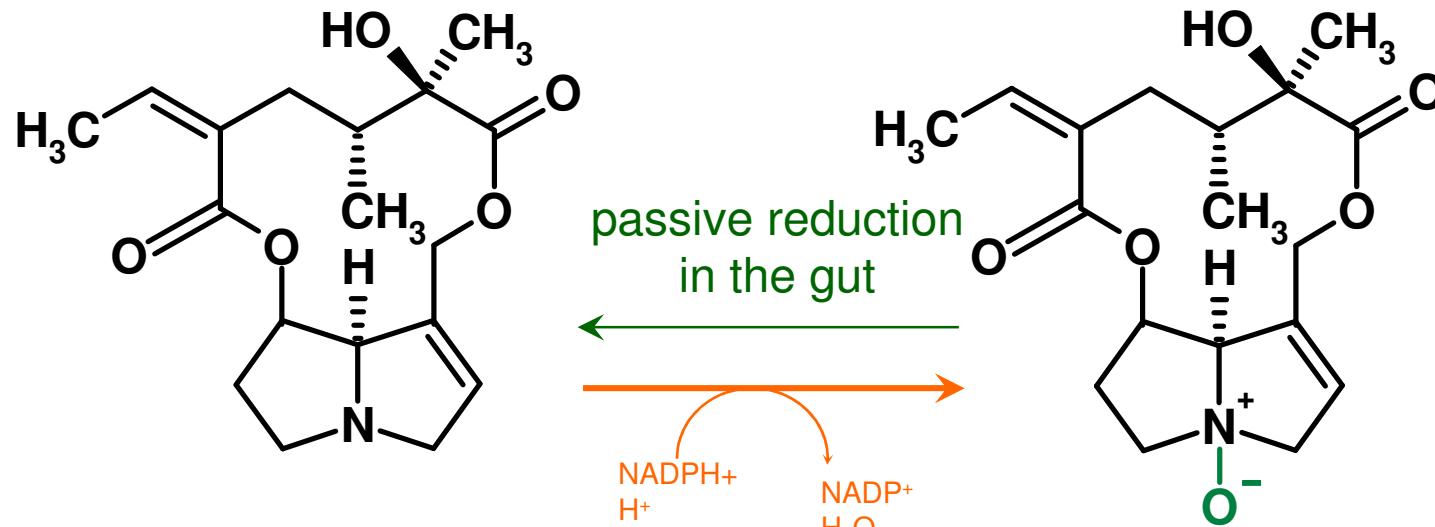
Sequestration of toxic pyrrolizidine alkaloids in an adapted herbivore



Sequestration of toxic pyrrolizidine alkaloids in an adapted herbivore



The two faces of pyrrolizidine alkaloids



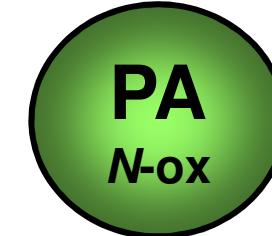
tertiary PA

- lipophilic
- **toxic** after bioactivation



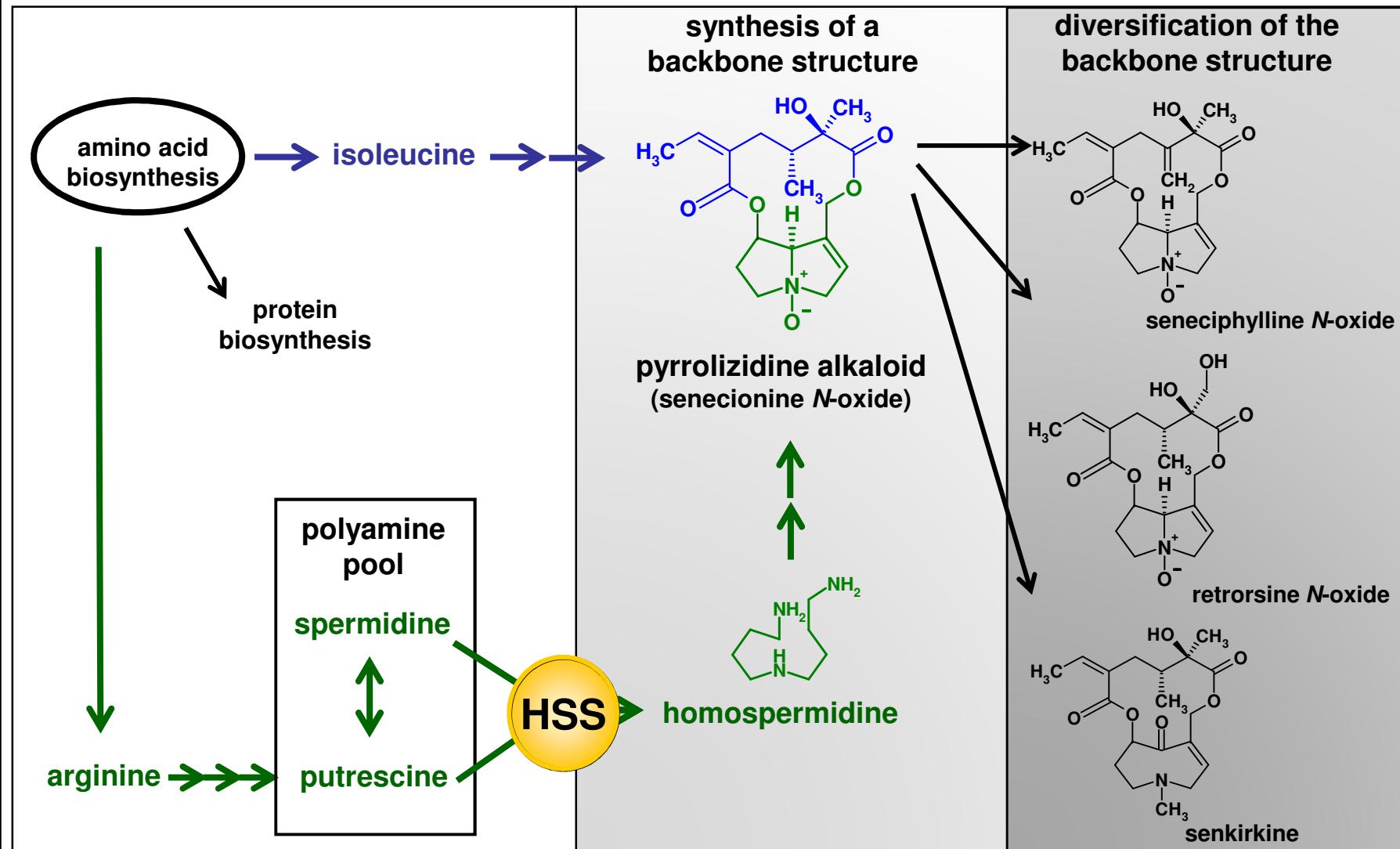
PA N-oxide

- hydrophilic
- **nontoxic**

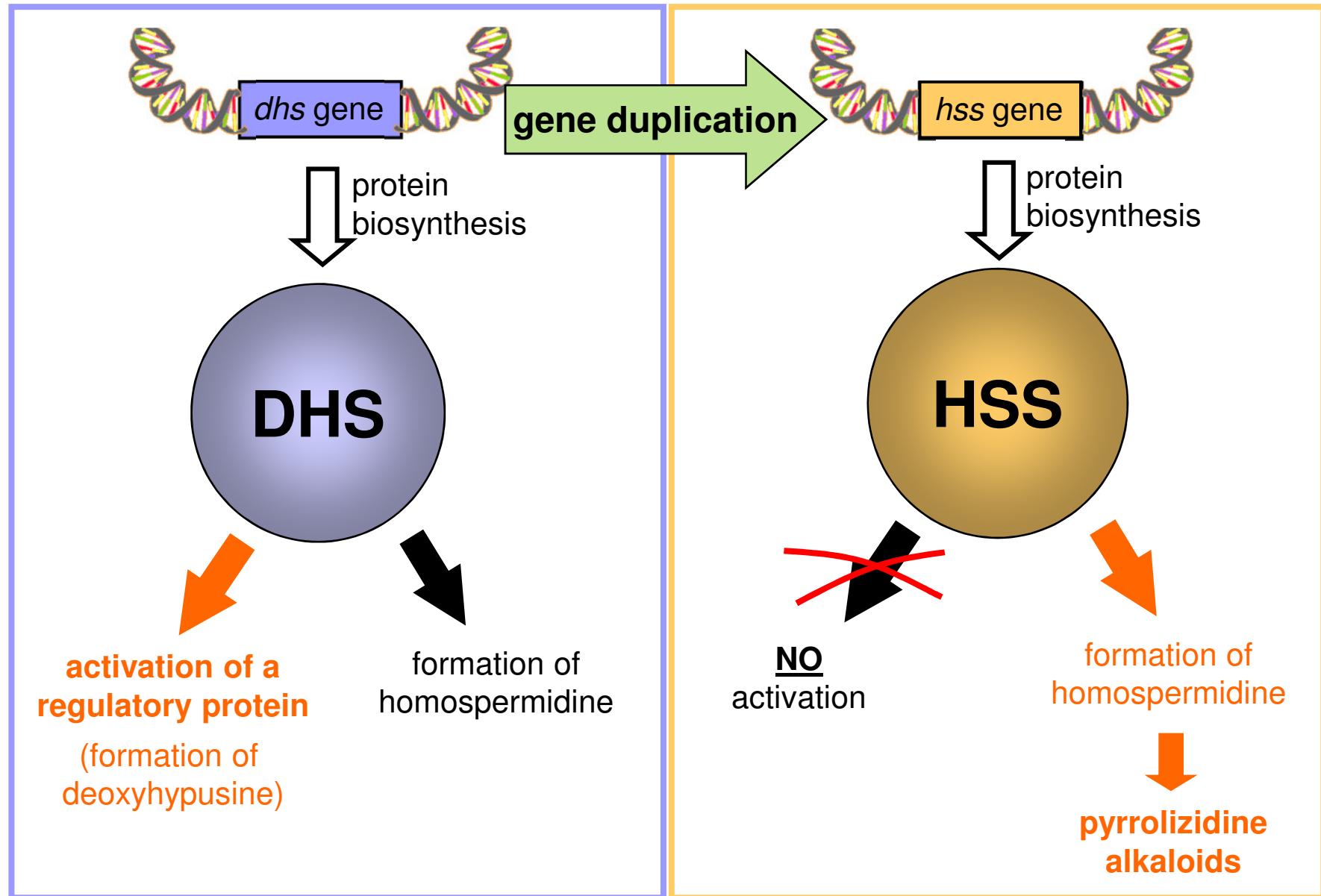


primary metabolism

secondary metabolism

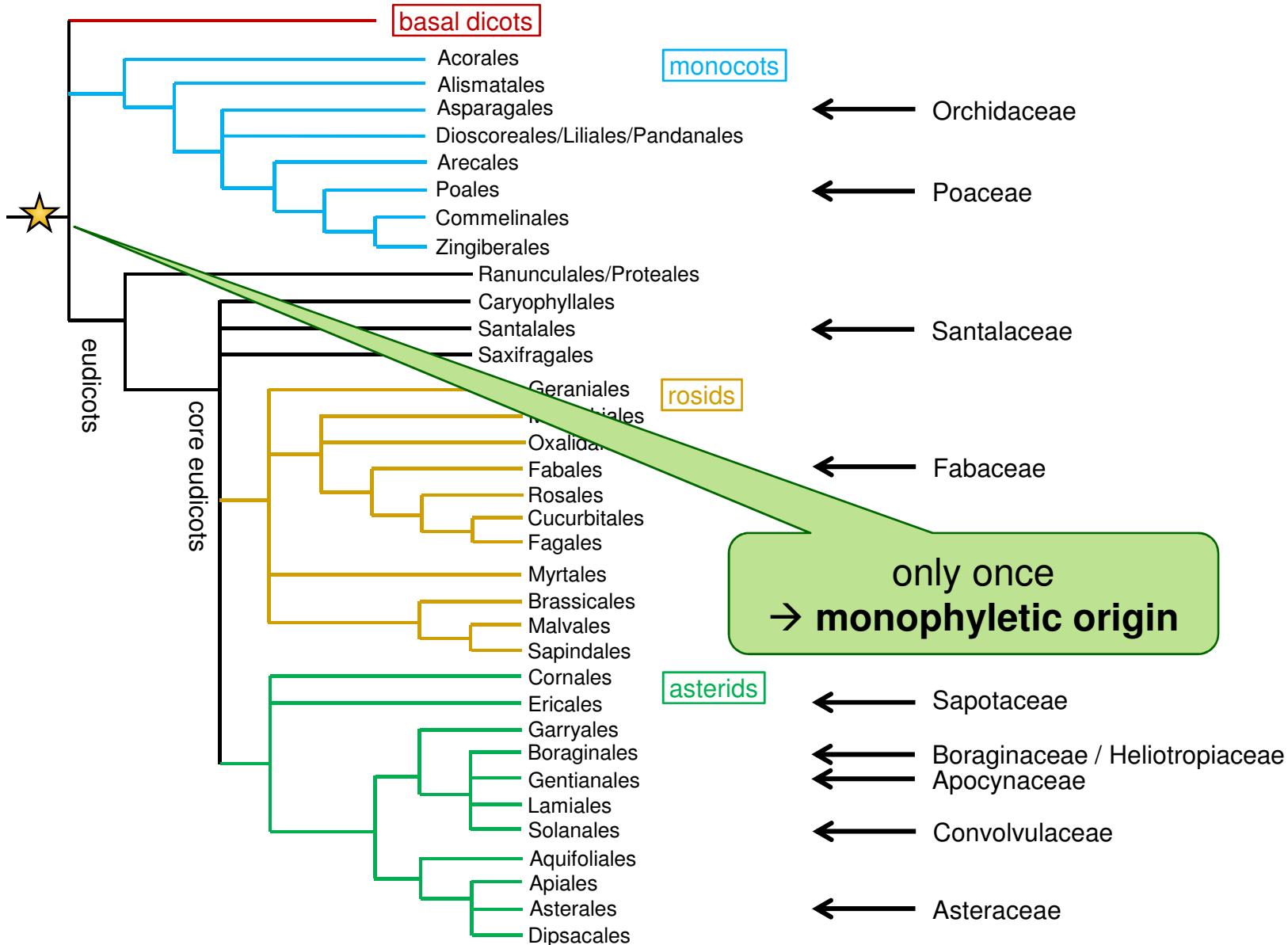


Evolution of homospermidine synthase by gene duplication





Occurrence of pyrrolizidine alkaloids

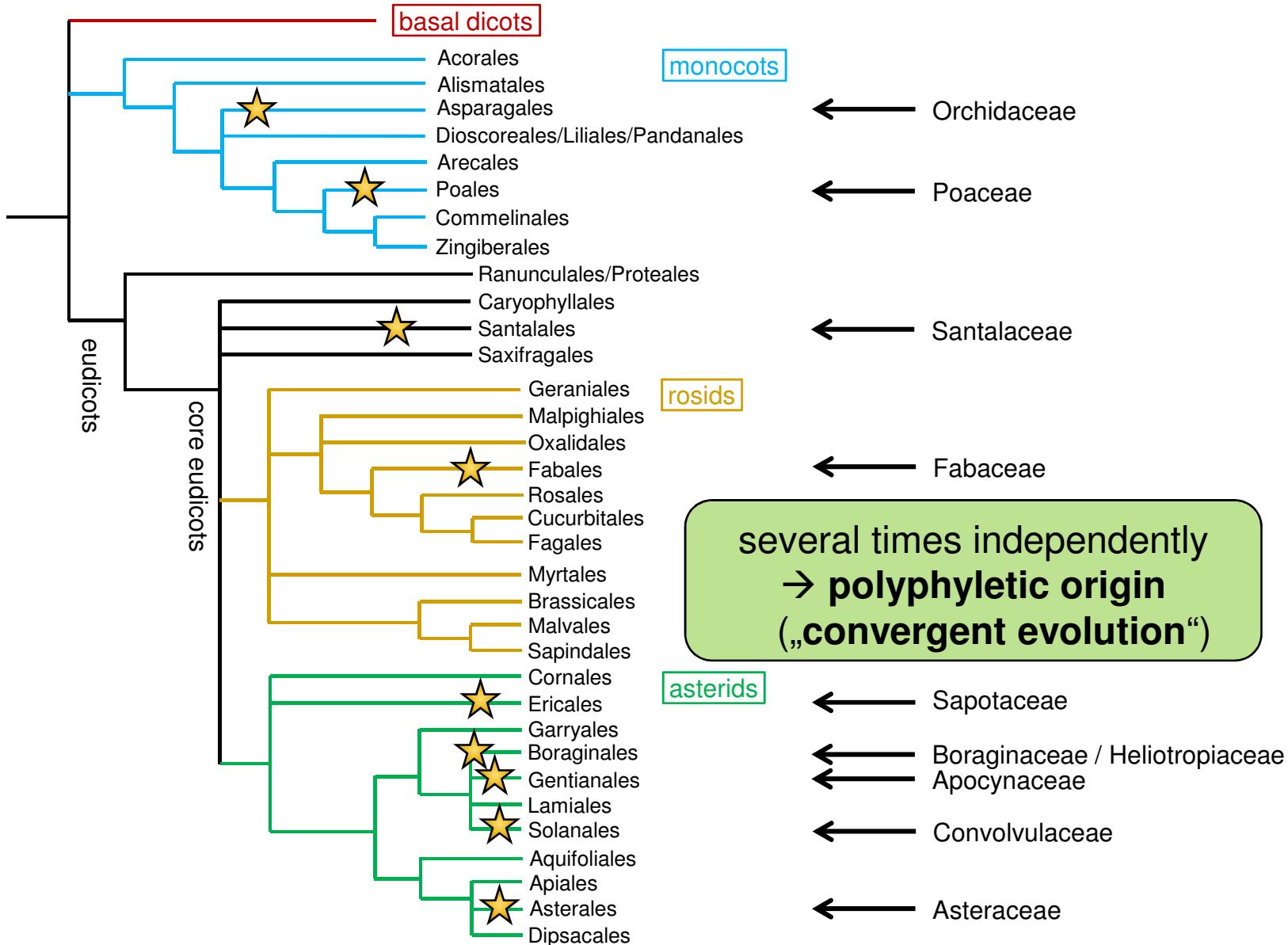


modified from Angiosperm Phylogeny Group III (2009)

© Dietrich Ober



Occurrence of pyrrolizidine alkaloids



modified from Angiosperm Phylogeny Group III (2009)

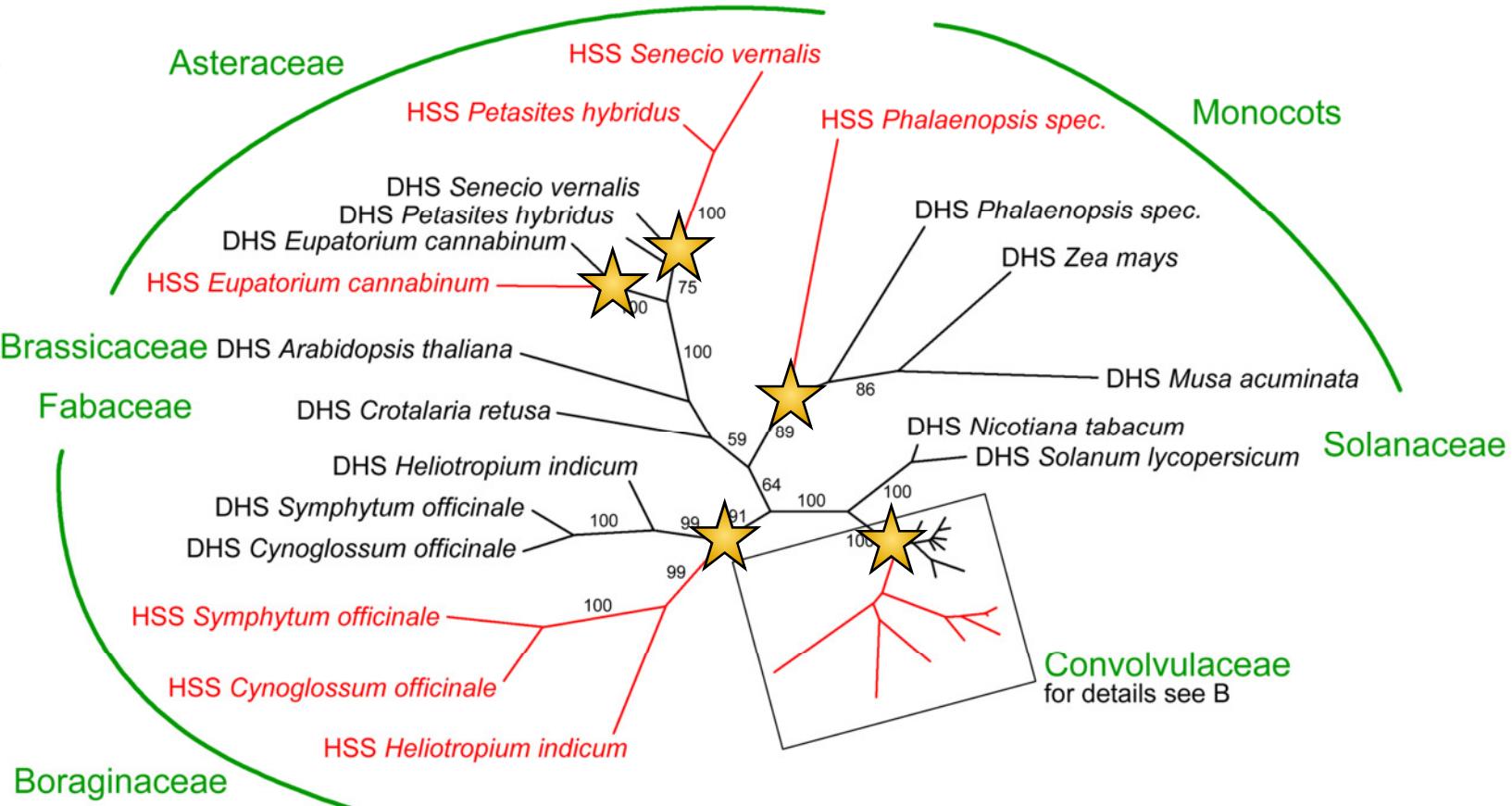
© Dietrich Ober

Phylogenetic tree of DHS- and HSS-coding cDNA sequences



HSS coding cDNA
DHS coding cDNA

Maximum Likelihood Tree
Bootstrap proportions: 1000 replicates

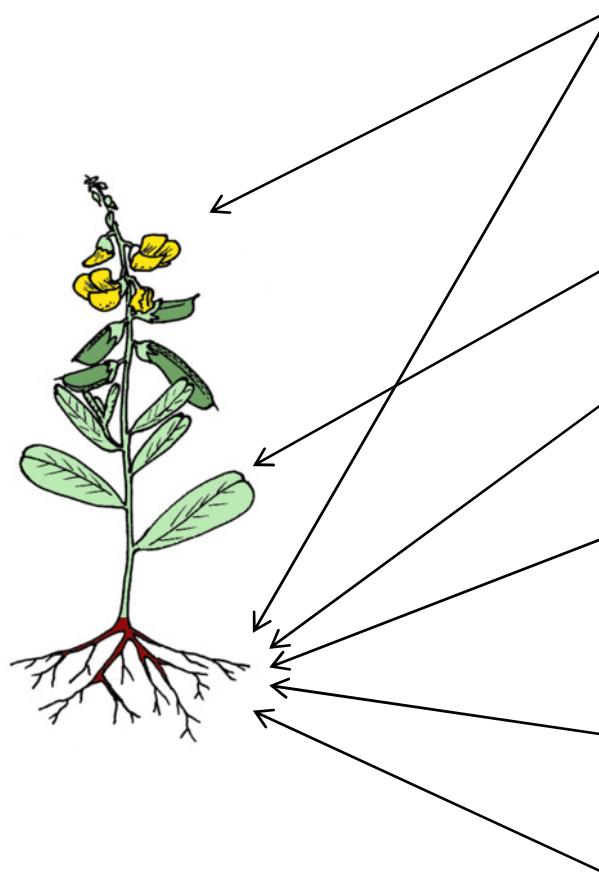


0.1 subst. per site

Kaltenegger et al. (2013) Plant Cell

© Dietrich Ober

Tissue-specific expression of HSS



Phalaenopsis (orchid)

- flower buds
- root tips

Heliotropium (Heliotropiaceae, Boraginales)

- leaf epidermis

Sympytum (Boraginaceae, Boraginales)

- root: only endodermis

Cynoglossum (Boraginaceae, Boraginales)

- root: endodermis and pericycle

Eupatorium (Asteraceae)

- cortex cells in the root

Senecio (Asteraceae)

- group of cells in the root

→ HSS expression is highly specific

→ HSS highly variable between different plant lineages

Tissue-specific expression of HSS



leaf cross section of *Heliotropium indicum*





Analysis of HSS-Evolution within the Convolvulaceae



Morning Glory Family

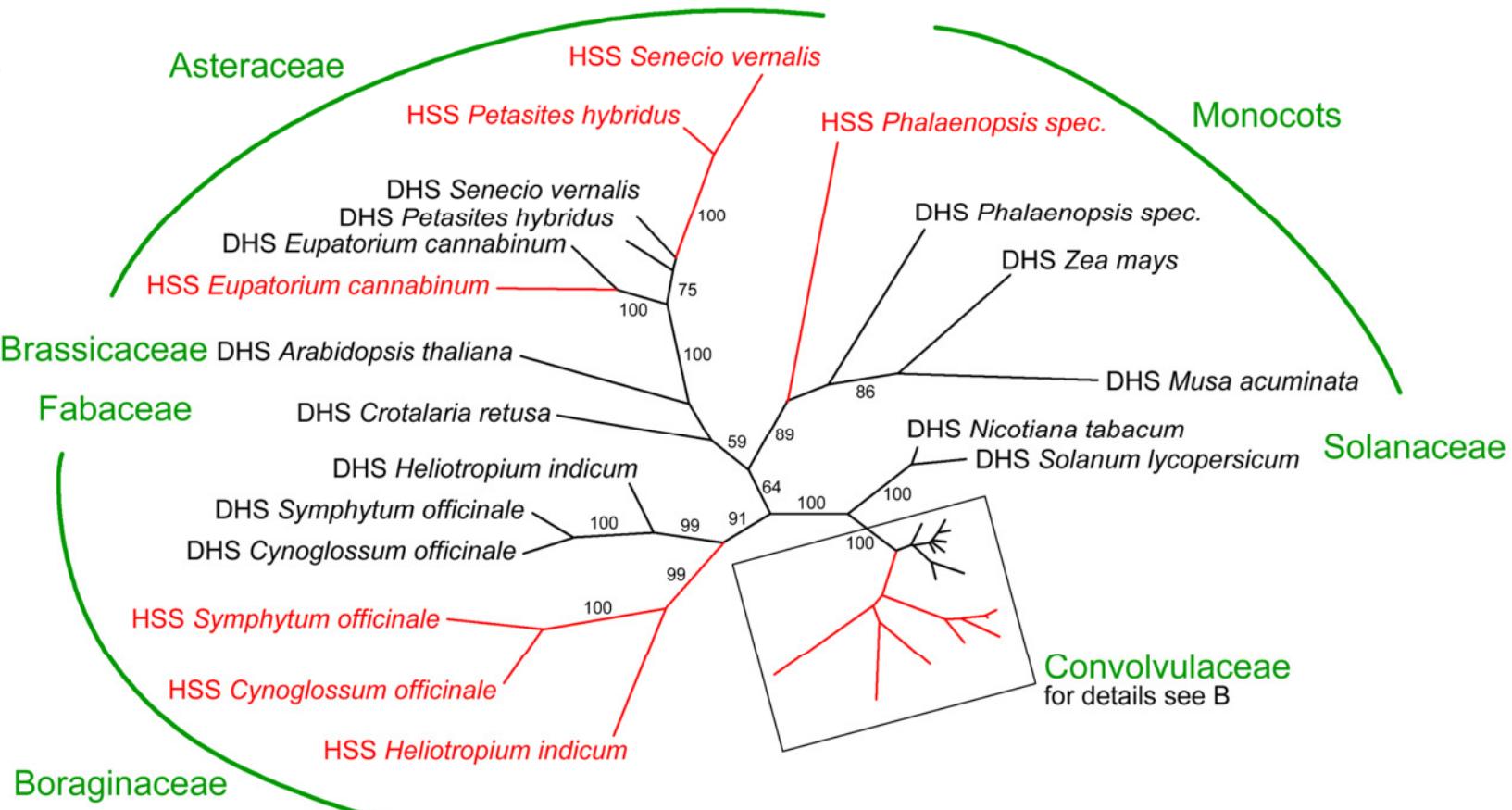
- belongs to the order Solanales
- approximately 1600-1700 species
- PAs have been detected
 - only in individual unrelated species
 - of quite diverse chemical structures

Phylogenetic tree of DHS- and HSS-coding cDNA sequences



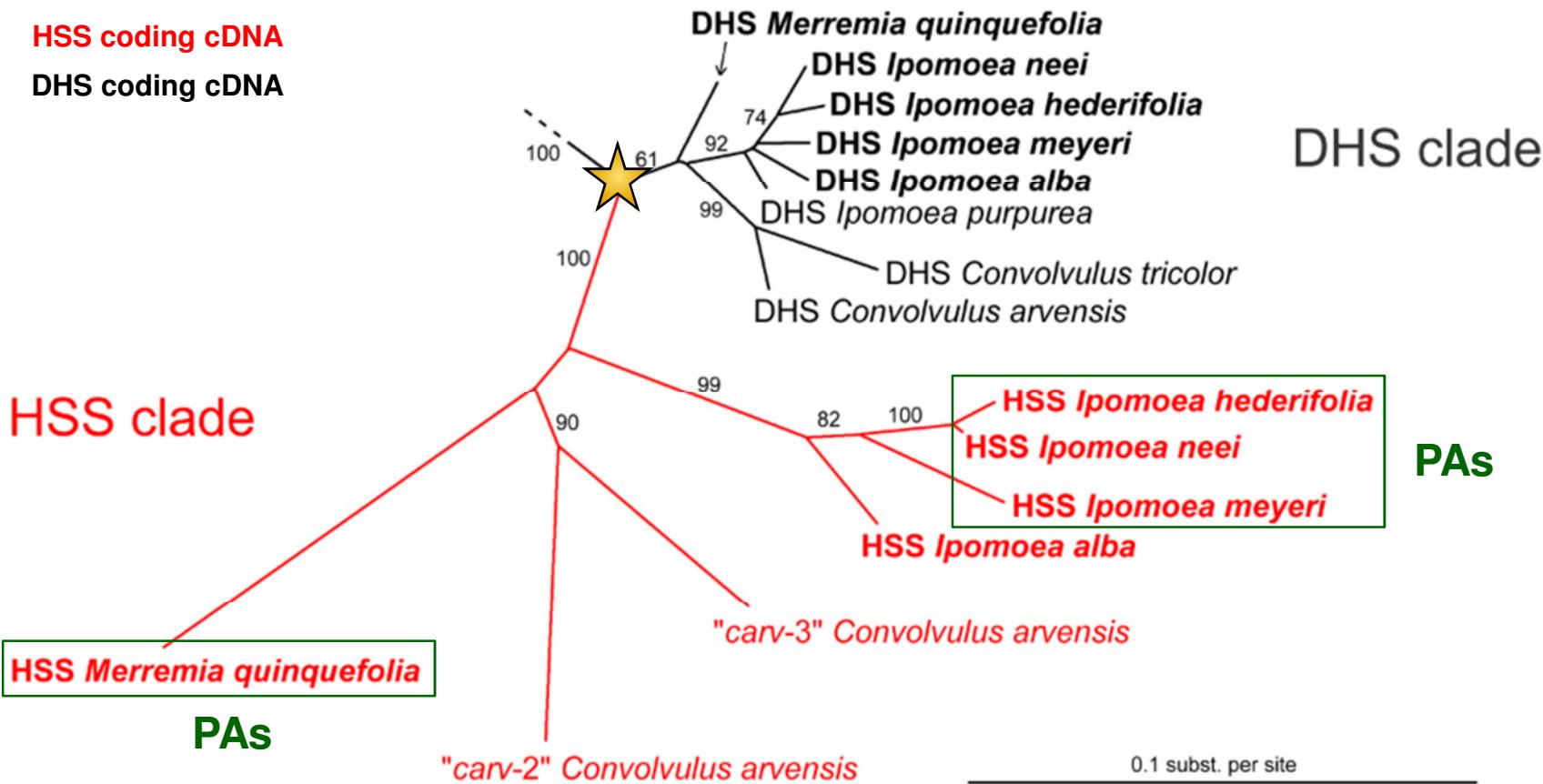
HSS coding cDNA
DHS coding cDNA

Maximum Likelihood Tree
Bootstrap proportions: 1000 replicates



0.1 subst. per site

Analysis of HSS-Evolution within the Convolvulaceae



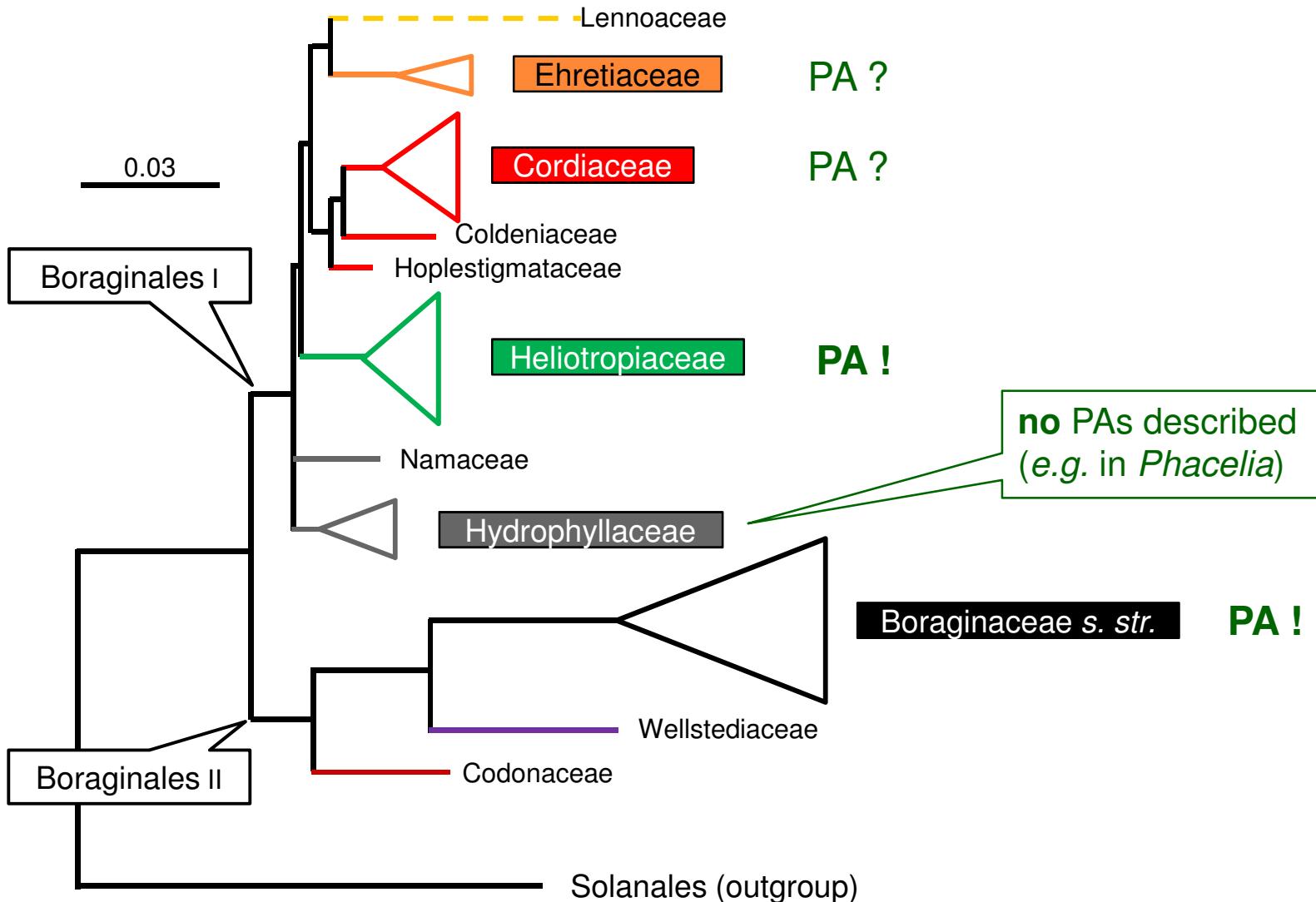
- only **one gene duplication** event within the Convolvulaceae lineage
- **pseudogenization** and **gene loss** of the HSS-related gene copy in several PA-free species

Phylogeny of the Boraginales



Phylogeny based on four chloroplast sequences

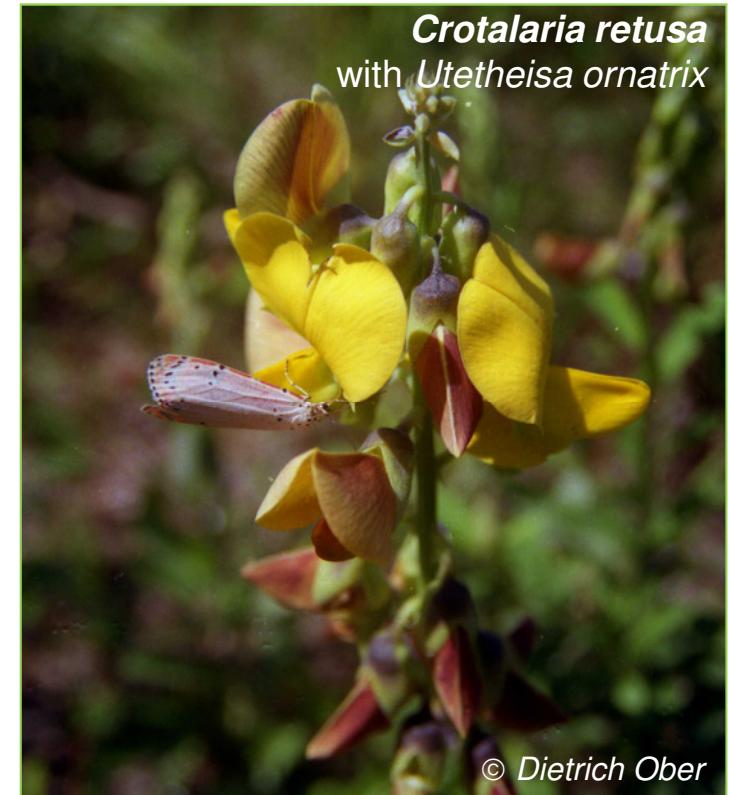
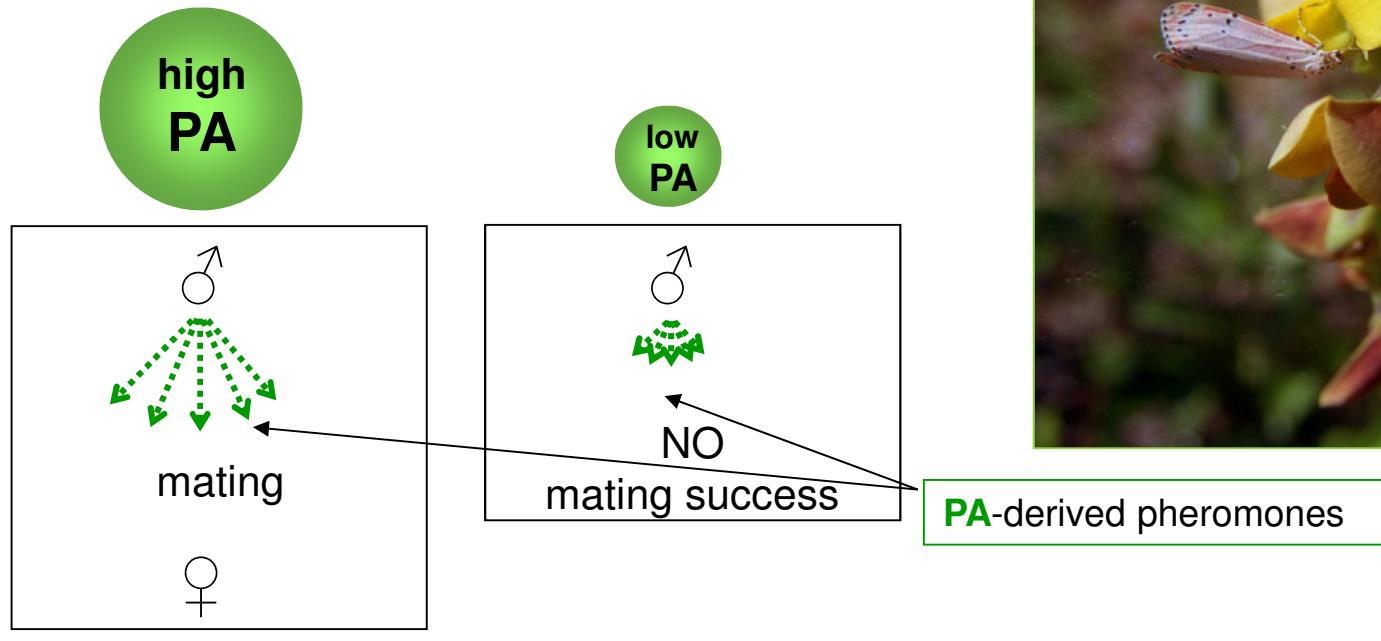
modified from Weigend et al. (2013) Cladistics

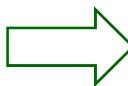


PA biosynthesis within Fabaceae



- PA occurrence is described for the two closely related genera of *Crotalaria* and *Lotononis*
 - PAs are used by specialized insects like *Utetheisa ornatrix*
- search for a PA-specific HSS was not successfull



 **PAs are transferred** → from the male to the female
→ to the next generation

fitness advantage by
plant-derived toxins

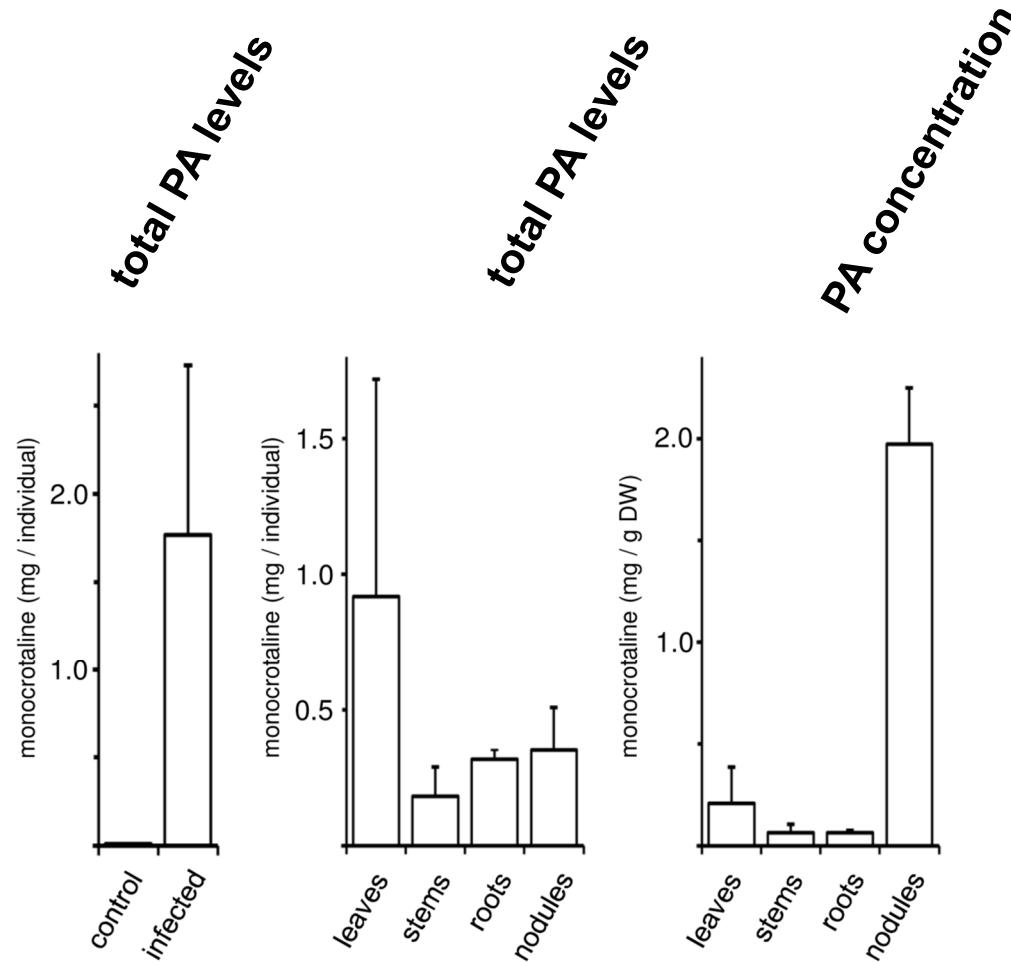


PA biosynthesis within Fabaceae



Comparison of nodulated and non-nodulated *C. spectabilis*

→ PA content



C. spectabilis
40 days after infection

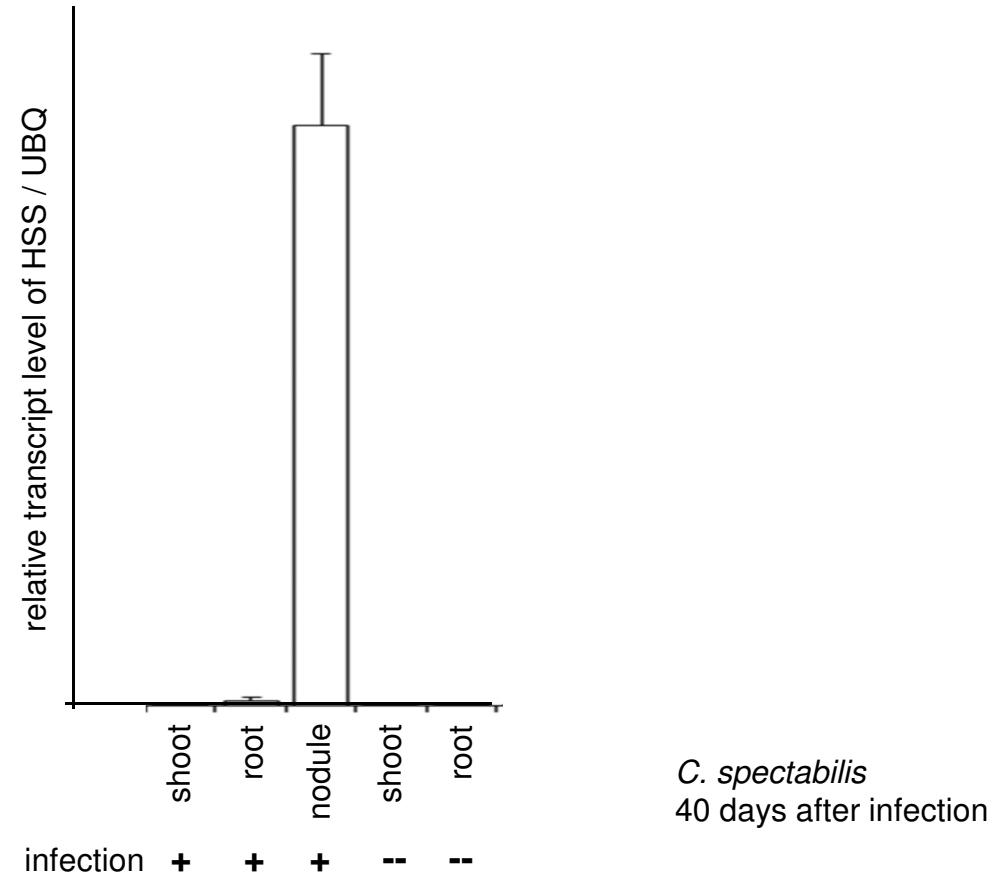
→ data suggest the nodules to be the site of PA biosynthesis

PA biosynthesis within Fabaceae



Comparison of nodulated and non-nodulated *C. spectabilis*

→ HSS expression



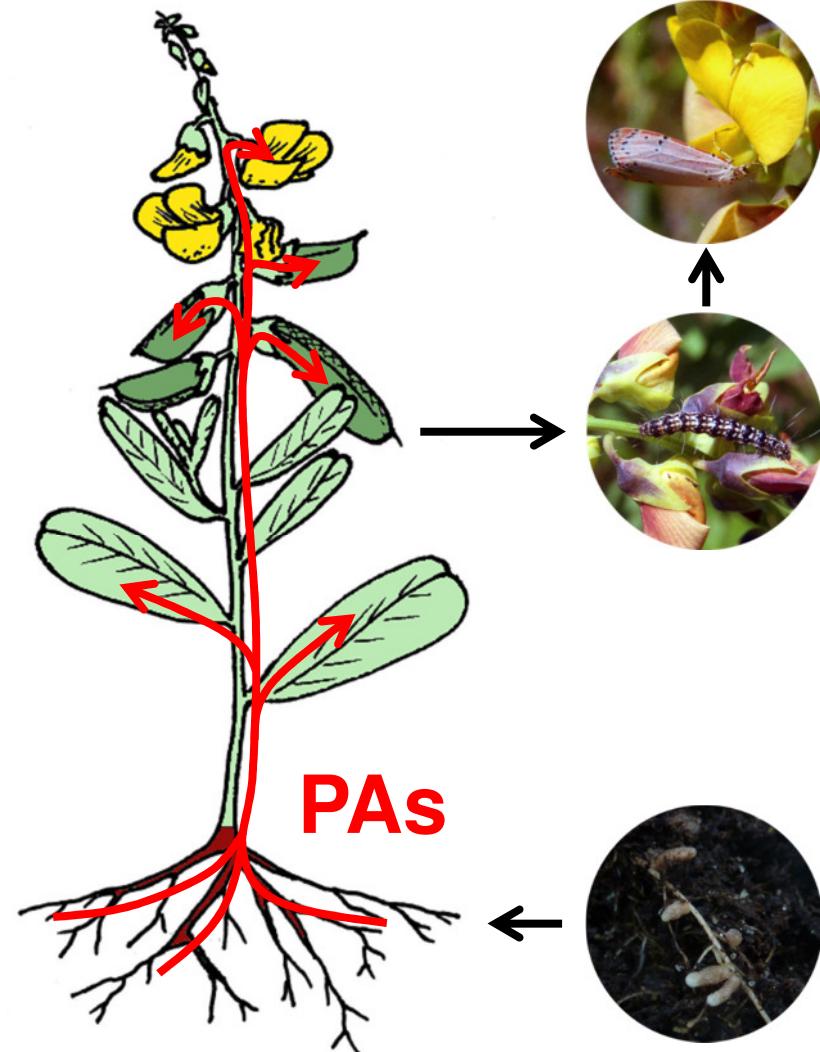
→ HSS is expressed exclusively in the nodules

PA biosynthesis within Fabaceae



in *Crotalaria*

- PAs are a link between
- symbiotic bacteria
 - plant
 - specialized insect





C | A | U KIEL Biochemical Ecology and Molecular Evolution

DFG
DAAD

Molecular Evolution of Chemical Diversity



Thank you !



zeller

