

I. Intro. to Plant Secondary Compounds

A. Examples of secondary metabolites (based on Table 10.3)

| Compound | Example Source | Human Use |
|----------------------|----------------------|--|
| ALKALOIDS | | |
| Codeine | Opium poppy | Narcotic pain relief; cough suppressant |
| Nicotine | Tobacco | Narcotic; stimulant |
| Quinine | Quinine tree | Used to treat malaria; tonic |
| Cocaine | Coca | Narcotic, tea, anesthetic, stimulant |
| PHENOLICS | | |
| Lignin | Woody plants | Hardwood furniture & baseball bats |
| Tannin | Leaves, bark, acorns | Leather tanning, astringents |
| Salicin | Willows | Aspirin precursor |
| Tetrahydrocannabinol | Cannabis | Treatment for glaucoma & nausea |
| TERPENOIDS | | |
| Camphor | Camphor tree | Component of medicinal oils, disinfectants |
| Menthol | Mints & eucalyptus | Strong aroma; cough medicines |

B. Ecological role of secondary metabolites.

Open access, freely available online PLOS BIOLOGY

Nicotine's Defensive Function in Nature

Anke Steppuhn, Klaus Gase, Bernd Krock, Rayko Halitschke, Ian T. Baldwin*

Department of Molecular Ecology, Max Planck Institute for Chemical Ecology, Jena, Germany

Plants produce metabolites that directly decrease herbivore performance, and as a consequence, herbivores are selected for resistance to these metabolites. To determine whether these metabolites actually function as defenses requires measuring the performance of plants that are altered only in the production of a certain metabolite. To date, the defensive value of most plant resistance traits has not been demonstrated in nature. We transformed native tobacco (*Nicotiana attenuata*) with a consensus fragment of its two *putrescine N-methyl transferase (pmt)* genes in either antisense or inverted-repeat (IR*pmt*) orientations. Only the latter reduced (by greater than 95%) constitutive and inducible nicotine. With D₄-nicotinic acid (NA), we demonstrate that silencing *pmt* inhibits nicotine production, while the excess NA dimerizes to form anatabine. Larvae of the nicotine-adapted herbivore *Manduca sexta* (tobacco hornworm) grew faster and, like the beetle *Diabrotica undecimpunctata*, preferred IR*pmt* plants in choice tests. When planted in their native habitat, IR*pmt* plants were attacked more frequently and, compared to wild-type plants, lost 3-fold more leaf area from a variety of native herbivores, of which the beet armyworm, *Spodoptera exigua*, and *Trimerotropis* spp. grasshoppers caused the most damage. These results provide strong evidence that nicotine functions as an efficient defense in nature and highlights the value of transgenic techniques for ecological research.

Citation: Steppuhn A, Gase K, Krock B, Halitschke R, Baldwin IT (2004) Nicotine's defensive function in nature. PLoS Biol 2(8): e217.

Introduction

Plants produce many secondary metabolites, of which some

putative defense trait should be determined in plants differing only in a single gene that controls the expression of a resistance trait and are otherwise identical (Bergelson and

Steppuhn et al. 2004. *PLoS Biology* 2: 1074-1080.

B. Ecological role of secondary metabolites.

Nicotine negatively affects function of herbivores.

Update on *Nicotiana attenuata*

An Ecologically Motivated Analysis of Plant-Herbivore Interactions in Native Tobacco¹

Ian T. Baldwin*

Department of Molecular Ecology, Max Planck Institute for Chemical Ecology, Carl Zeiss Promenade 10, D-07745 Jena, Ger *Plant Physiology*, December 2001, Vol. 127, pp. 1449-1458,

You can't always get what you want, but if you try some time, you just might find, you get what you need...

Mick Jagger

Unfortunately, a comprehensive understanding of internal processes is not sufficient to test the cost-benefit paradigm, because Darwinian fitness can also be influenced by processes external to the plant (Fig.

B. Ecological role of secondary metabolites.

Plant Compounds are Diuretics to Desert Herbivores

by Denise Dearing, Antonio Mangione and William Karasov

Many plant compounds are recognized deterrents and toxins to a variety of herbivores. The effect of such compounds on water balance of herbivores is virtually unexplored, yet many plant compounds cause diuresis by elevating urine production and decreasing urine concentration. Caffeine from coffee and black tea is probably the most familiar diuretic agent from plants. However, caffeine is not exceptional.

Plant products that cause diuresis in humans.

Diuretic Plant Extracts



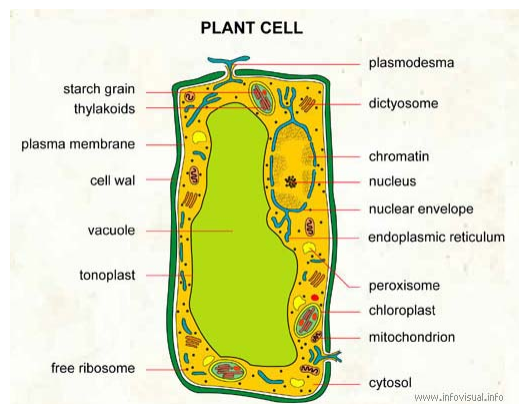
B. Ecological role of secondary metabolites.



Jasminum

I. Intro. to Plant Secondary Compounds

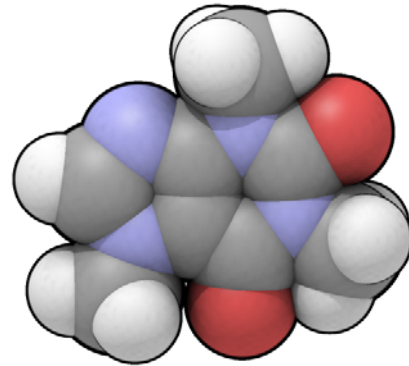
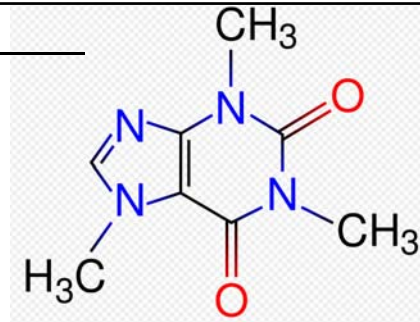
C. Storage of secondary metabolites



II. Caffeine case study

Caffeine

- Coffea*, *Theobroma*, *Camelia*, *Kola*, etc.
- Psychoactive stimulant, diuretic
- Alkaloid



II. Caffeine case study

Caffeine

- syn. w/ guaranine

Paullinia cupana (guarana' vine)
Sapindaceae. Native of S. America



II. Caffeine case study

Caffeine

-syn. w/ theine

Camelia sinensis (tea bush)
Theaceae. Native of S. Asia



II. Caffeine case study

Caffeine

-syn. w/ mateine

Ilex
paraguariensis
(yerba mate)
Aquifoliaceae.
Native of S.
America.



II. Caffeine case study

Caffeine

-known as caffeine (orig. *kaffein*, from *kaffee*)

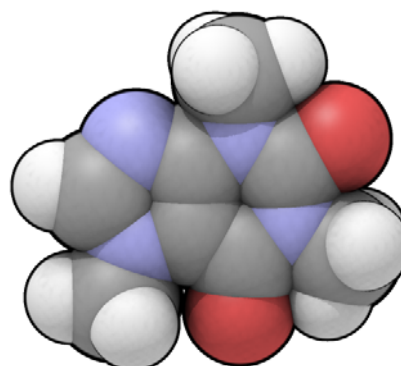
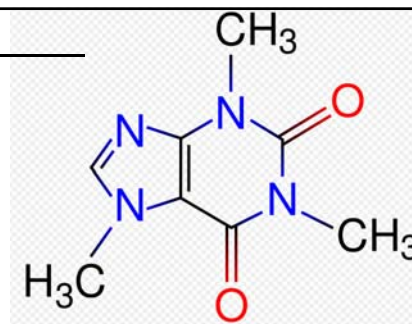


Coffea arabica
(arabica coffee)
Rubiaceae.
Native of NE
Africa.

II. Caffeine case study

Alkaloids in general

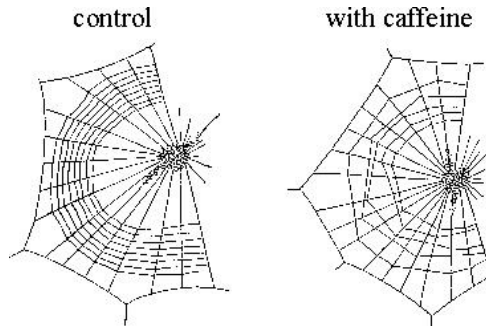
- Secondary metabolites
- Nitrogenous cmpds.
- Psychoactive (act on CNS)



II. Caffeine case study

Ecological role of alkaloids

Spider web manufacture when influenced by caffeine.



II. Caffeine case study

Ecological role of alkaloids

Caffeine's natural role noticed by Monsanto.



Information Systems for Biotechnology

By Janaki Krishna
Friday, 6th April, 2007

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Caffeine Producing Transgenic Tobacco: A Novel Pest Control Strategy

Caffeine (1,3,7-trimethylxanthine) is an alkaloid compound that acts as a central nervous system stimulant in humans and is likely the world's most popular psychoactive substance. Caffeine is generally found in the beans, leaves, and fruits of over 60 plants, where it acts as a natural pesticide, paralyzing and killing certain phytophagous insects and repelling slugs and snails. In the last fifteen years, considerable advances have been made in the genetic transformation of coffee plants. Researchers have been able to transform coffee plants with genes for insect resistance and herbicide tolerance, engineer decaffeinated coffee, and control coffee fruit maturation.

Herbivory accounts for approximately 37% loss in world agriculture production. Due to its natural antiherbivory function, caffeine production within food crops may provide one useful means for protecting important crops. Research indicates the reproductive potential (ovary length and egg number) of lepidopterans is significantly reduced in insects fed leaves treated with caffeine and the related compound theophylline, found in tea.

The Nara Institute of Science and Technology in Japan recently reported research on the development of caffeine-producing transgenic tobacco plants tolerant to tobacco cutworms (*Spodoptera litura*). Previously, the researchers isolated genes encoding three distinct N-methyltransferases and demonstrated in vitro production of the recombinant enzymes responsible for caffeine yield. They also published a review of the metabolic engineering of the caffeine biosynthetic pathway utilizing both gene silencing and over-expression approaches. The application of this research supported further efforts to employ transgenic caffeine-expressing plants as insect repellents.

www.monsanto.co.uk

II. Caffeine case study

Ecological role of alkaloids

Nitrogen availability is a major limiting factor in plant growth.



Nitrogen Limitation Restricts CO₂ Absorption by Trees

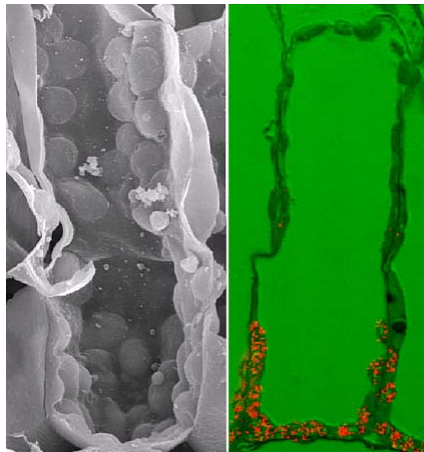
New research suggests that trees may not be able to limit climate change by storing rising atmospheric CO₂ as was previously believed. Limited availability of nitrogen in the soil, which will become more common as atmospheric CO₂ levels rise, may inhibit plant growth, which in turn would affect plants' accumulation of atmospheric CO₂. Atmospheric CO₂ levels may therefore rise even faster than anticipated.

II. Caffeine case study

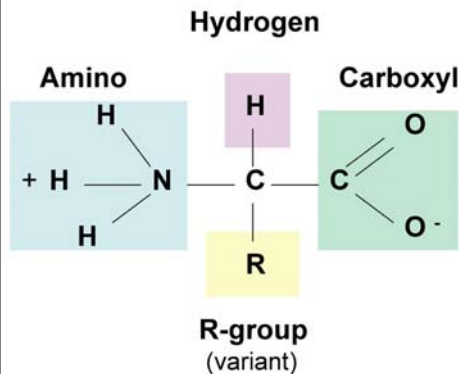
Ecological role of alkaloids

Proteins (which drive biological reactions) require large amounts of N.

*Proteins drive life processes (e.g., RuBisCO) and are important structural elements.



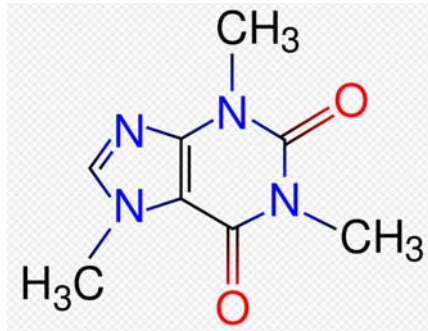
*Amino acid structure



II. Caffeine case study

Ecological role of alkaloids

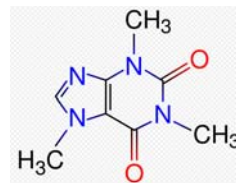
*Classic alkaloid composition exemplified by caffeine



Caffeine's effects on CNS

- Caffeine from coffee in blood w/in 5 min
- Stimulates heart
- Increases stomach acidity
- Increases urine output
- 10% rise in metabolic rate

- Mimics feelings assoc. w/ adrenaline



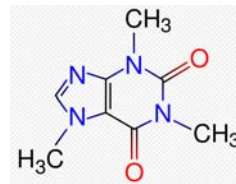
caffeine

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•Mimics feelings assoc. w/ adrenaline

•Excess (1 g; 10 cups) can cause anxiety, headache, dizziness, insomnia, heart palpitations, delirium, 4% lower birth weights.



caffeine

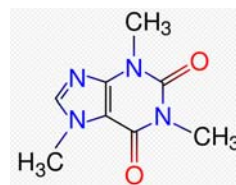
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•Ranks as most widely used psychoactive drug worldwide (coffee, tea, additives to soft drinks)



caffeine

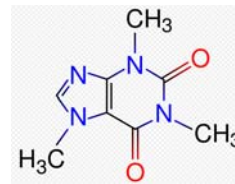
Caffeine's effects on CNS

How?

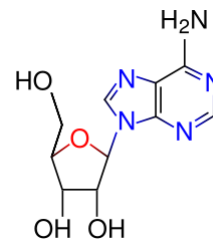
Antagonist of adenosine.

Adenosine:

- Attaches to brain cell receptors.
- Neurotransmitter inhibitor.
- Promotes sleep
(accumulates in brain each waking hour).
- Suppresses arousal.



caffeine



adenosine

Caffeine and Parkinson's prevention?

What is Parkinson's Disease?

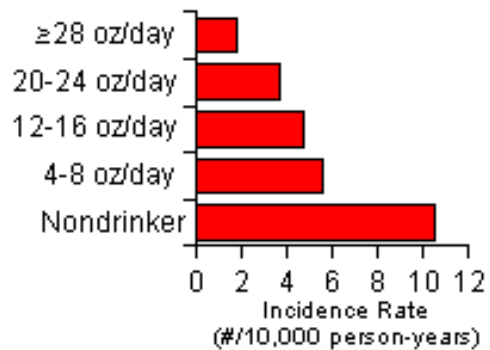
Journal of the American Medical Association, March 24, 2000

- afflicts ca. 1-1.5 million people in the U.S., mostly 60 years +
- no known cause and no cure, just treatments
- symptoms of trembling arms and legs, trouble speaking, and difficulty coordinating movement

- neuron degeneration in spec. part of brain
- many of these neurons contained the neurotransmitter dopamine
- dopamine levels fall, and the balance between dopamine and other neurotransmitters disrupted, affecting muscular control

Caffeine and Parkinson's prevention?

Honolulu Heart Program study of 8,000+ men over 30?

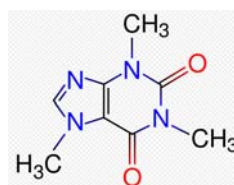


Mechanism: When adenosine receptors are blocked, dopamine levels increase.

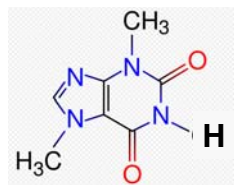
Caffeine and Theobromine are similar in structure and action

Table 1. Stimulant alkaloids in world's major stimulating beverages (Simpson 1986). Given in % weight. Amt. in particular beverage depends on how it is made.

| Plant, part | Caffeine | Theobromine |
|--------------------------------|----------|-------------|
| Coffee, unroasted, dried seeds | 1-1.5 | -- |
| tea, dried lvs. | 2.5-4.5 | -- |
| Cacao, dried or fresh seeds | 0.6-0.8 | 1.7-2.4 |
| Kola, fresh seeds | 2.0 | -- |
| Guarana, dried fruit | 3.0-4.5 | -- |



caffeine

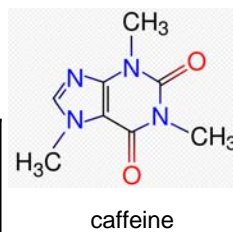


theobromine

Caffeine in some beverages

Table 2. Caffeine quantities in select beverages.

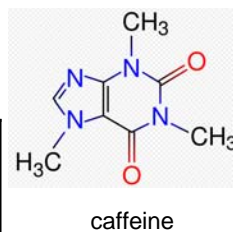
| Drink | Caffeine (mg) |
|---------------------------------------|-----------------|
| Coffee | |
| 5 oz Drip, percolator, instant, decaf | 146, 110, 53, 2 |
| 1 oz espresso | ? |



Caffeine in some beverages

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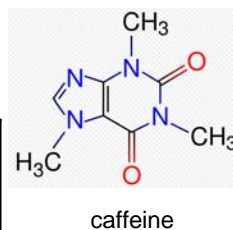
| Drink | Caffeine (mg) |
|---------------------------------------|-----------------|
| Coffee | |
| 5 oz Drip, percolator, instant, decaf | 146, 110, 53, 2 |
| 1 oz espresso | 50 |



Caffeine in some beverages

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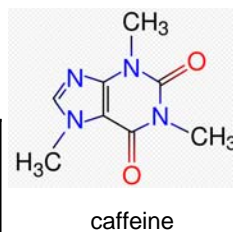
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| Coffee | |
| 5 oz Drip, percolator, instant, decaf | 146, 110, 53, 2 |
| 1 oz espresso | 50 |
| Tea (5 oz) | |
| Brewed 1 min, 3-5; 12 oz can | 9-33, 20-50, 22-36 |
| Cocoa and chocolate | |
| 6 oz, from powder | 10 |
| 1 oz milk choc | 6 |
| 1 oz dark choc | 20 |
| 1 oz baking choc | 35 |
| Soda (12 oz) | |
| Mt. Dew | 52 |
| Dr. Pepper | 37-38 |
| Pepsi | 37 |
| Coca cola | 34 |



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99.93% caffeine-free
(drip coffee is 99.90%)

III. Coffea

Coffee

Coffee is world's second most traded commodity.



Coffea arabica
(arabica coffee)

Table 3. Production of top 3 stimulant beverages.

| Top 3 continents | Total (MT) |
|------------------|------------|
| Coffee | 5,919 |
| 1. S Amer | |
| 2. Africa | |
| 3. N & C Amer | |
| Tea | 2,473 |
| 1. Asia | |
| 2. Africa | |
| 3. S Amer | |
| Cocoa | 2,329 |
| 1. Africa | |
| 2. S Amer | |
| 3. Asia | |

III. Coffea

Coffee breeding

Coffee Biotech Group (Campinas, Brazil)



III. Coffea

Coffea berries and flowers



III. Coffea

Coffea berries and flowers



III. Coffea

Coffea seeds



III. Coffea

Coffea seeds



III. Coffea

Coffee origins

Coffea arabica, old world tropical/subtropical crop grown at mid to upper-middle elevations

- Relatively few Africans drink it.
- Traditionally chewed leaves & fruits.
- Relief of fatigue, hunger on hunts.



Farmer in Ethiopia

Coffee (the drink) developed in Yemen

Arrival in Yemen 13-14 century, where it was first brewed (hence, *Coffea arabica*).



ca. 1900

11059—A Coffee-house in Palestine.

Coffee timeline

1. First brewed in Yemen 13-14 century (hence, *Coffea arabica*).
2. Arabia to Egypt by 1510.
3. To Italy & Europe by 1616.
4. Vienna priests threatened by “coffee culture”, but Pope Clement VIII would not ban coffee.
5. To England by 1650 and coffee houses became important socio-politico institutions.
6. Europe looked to break Arabian monopoly on production.
(*Arabians killed embryos in seeds before export*).

Spread of Coffee production

- Dutch obtained live seeds from Mocha (Red Sea Coast, Yemen, 1706)
- Throughout Dutch colonies in Indonesia (e.g., Java) and to S America by 1717.
- Today, Brazil is world's leading producer.

