pleiotropic effects in grey poplar caused by UV radiation in wt & isoprene non-emitting plants
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• BVOCS contribute to ozone-formation & global warming

• BVOS are carbon bases molecules

• chances in carbon-fluxes might alter BVOC emissions

→ UV as a trigger to alter “C”-sequestration within the cell
- in our work-group we study isoprene-emission

- mainly on grey poplar (*Populus x canescens*) in wt & transgenic plants

- considerable amounts of fixed C are released from the plant as isoprene

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crosstalk between isoprene formation & other biochemical pathways

quantification of variable „C“ pools

regulation of MEP pathway (feedback control; post-translational modifications)

export of IPP import of DMAPP contribution to different isoprenoid pathways

metabolic competition: C5/C10 versus C40 isoprenoids

introduction idea set-up results conclusions out-look
what is UV radiation anyway?

- high-frequent light
- low wavelength (280-380nm)
- not visible to human eyes
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UV effects - direct:

- oxidative damaging of macro-molecules such as DNA, aromatic amino-acids & membrane-lipids
- generation of reactive oxygen species (ROS)

UV effects - indirect:

- induction of synthesis of protective compounds such as phenolic compounds & carotinoids...
- changes in gene-expression at different levels:
  - transcription
  - translation
  - post-translational modification
- induction of anti-oxidative enzymes

e.g. Brown et. al., 2005; Jordan, 2002
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what is the idea of the experiment, then?

- UV has in impact on the cells metabolism
- so it might change the use of carbon & its fluxes

→ therefore UV possibly also affects isoprenoid metabolism & isoprene emission?

→ the data should become integrated in a cellular model of BVOC emission
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Fatty acid biosynthesis

Acetaldehyde emission

Phenylpropanoid/Flavonoid pathway

Shikimate pathway aromatic aa

ABA

Carotenoids

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DTDP

DOXP

MEP

GPP

IPP + DMAPP

IDI

NADPH

NADP+

DXS

TP

Pyr

PEP

PEP

data by Katja Behnke

20 40 60 80 100 120 140 160

DMAPP [nmol mg dw⁻¹]
SO:

- exposure of poplar plants to UV radiation in comparison to un-radiated plants
- also we compare wt vs. transgenic lines

→ 4 treatments

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how did we set up the experiment:

- sun-simulators at the Helmholtz Centre, Munich
- these allow a good approximation to the solar spectrum
- under controlled conditions
- 2 repeats

on-line monitoring:

- PTR-MS (Ionicon, Austria)
- GFS-3000 (Walz, Germany)
- Mini-PAM (Walz, Germany)
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schematic outline of the sun simulators

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Metal halide lamps
Quartz halogène lamps
Blue fluorescent tubes
UV fluorescent tubes

picture by Dr. Andreas Albert
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timing:

- in total 2 weeks of monitoring
- on-line measurements at days 0,1,5,9,13
- harvest of leaves at days 0,1,2,3,5,9,13
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**in vitro measurements include:**

- analysis of:

  - PS-Pigments via HPLC
  - Metabolomics via ICR-FT/MS
  - Phenolic compounds via HPLC-MS
  - Gene-expression via RT-PCR
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Morphology

- Wild type
- \(Pc\text{ISPS}\text{-RNAi}\) line

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wild type
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No Differences

- photosynthetic parameters
- carotinoid-content
- α-tocopherol-level
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**DMAPP & Isoprene**

**introduction**

**idea**

**set-up**

**results**

**conclusions**

**out-look**
Anthocyanins & other Phenolic compounds

...more than 20 different compounds were found & quantified...
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Anthocyanins & other Phenolic compounds

phenolic compound 8:
- no line-effect
- no treatment-effect
- no time-effect

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- **K - UV**
- **K + UV**
- **RB - UV**
- **RB + UV**
Anthocyanins & other Phenolic compounds

Anthocyanins:
- treatment-effect
- time-effect
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Anthocyanins & other Phenolic compounds

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Camphorol-derivate:

- line-effect
- treatment-effect
- time-effect

R-lines show less flavonoids!
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**Results**

- Up and down regulation of pathways

- Transcription/translation: downregulated (71), upregulated (138)
- Others: downregulated (36), upregulated (87)
- Signal transduction: downregulated (3), upregulated (12)
- Photosystems: downregulated (1), upregulated (2)
- Antioxidative system: downregulated (3), upregulated (11)
- Stress metabolism: downregulated (3), upregulated (8)
- Isoprenoid metabolism: downregulated (2), upregulated (6)
- Phenylpropanoid/flavonoid metabolism: downregulated (1), upregulated (2)
- Amino acid (aa) metabolism: downregulated (1), upregulated (1)
- Sugar metabolism: downregulated (1), upregulated (1)
- Unknown: downregulated (1), upregulated (2)

Data by Katja Behnke
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review:

- by eye: clear effect of treatments
  - wild type
- effects in anthocyanins & flavonoids
- isoprene metabolism emission shows clear trends

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PcISPS-RNAi
line
interpretation:

- isoprene emission is probably linked to UV-response in poplar

- UV-radiation can be used to alter "C"-fluxes in plant systems

- isoprene is not a compound protecting the plant from UV effects, but rather acting competitive for "C"
interpretation:

- interesting however, is the observed decrease in flavonoid content in the ISPS-repressed plants
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this study:

- promising to analyse flavonoid gene expression
  - i.e. find target genes responsible for expression-pattern
  - quantify there expression levels

- structure analysis of phenolic compounds

- METABOLOMIC analysis should show compound composition & distribution in more detail
future prospects:

• METABOLOMICS provide a powerful tool for easy data-access

• transgenic lines will help to better understand cellular processes

• cooperation with modelers will help to better:
  - predict
  - understand

future scenarios
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- Peter Kary
- Werner Rupprecht

my supervisor:
Prof. Dr. Joerg-Peter Schnitzler
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...and...of course:

you, for your attention...