



This site provides online material for the [www.cosmoss.org](http://www.cosmoss.org) 3rd Physcomitrella Genome Workshop that will be held

on

**September 13th-15th 2010**

at

University of Freiburg, Germany



## Contents

- 1 Sponsors
- 2 Information
  - ◆ 2.1 Cosmoss accounts
- 3 Registration
- 4 Program
  - ◆ 4.1 Covered topics
  - ◆ 4.2 Schedule
  - ◆ 4.3 Poster session
- 5 Venue
  - ◆ 5.1 Biology I
  - ◆ 5.2 Biology II
- 6 Speakers
- 7 Workshop Content
  - ◆ 7.1 First Day
    - ◇ 7.1.1 Morning Session
      - 7.1.1.1 The Physcomitrella Genome
      - 7.1.1.2 cosmoss.org: resources, databases and versions
    - ◇ 7.1.2 Afternoon Session
      - 7.1.2.1 Lecture: Homology searching / BLAST
      - 7.1.2.2 Hands-on: BLAST

## Cosmoss\_workshop

- [7.1.2.3 Lecture: gene families](#)
- [7.1.2.4 Hands-on: gene family tools](#)
- [7.1.2.5 Lecture: sequence retrieval and genome browser](#)
- [7.1.2.6 Hands-on: sequence retrieval/keyword search](#)
  - [7.1.2.6.1 Sequence Retrieval](#)
  - [7.1.2.6.2 Keyword search](#)
- ◆ [7.2 Second Day](#)
  - ◇ [7.2.1 Morning Session](#)
    - [7.2.1.1 Lecture: Genome Browser Basics](#)
    - [7.2.1.2 Hands-on: Genome Browser Basics](#)
    - [7.2.1.3 Lecture: Genome Browser hidden treasures](#)
      - [7.2.1.3.1 Hands-on experience](#)
      - [7.2.1.3.2 BLAST gbrowse integration](#)
      - [7.2.1.3.3 Advanced navigation and zooming](#)
      - [7.2.1.3.4 Exporting sequence annotations and publication quality images](#)
      - [7.2.1.3.5 Highlighting](#)
      - [7.2.1.3.6 Displaying custom annotation](#)
    - [7.2.1.4 Lecture: functional annotation](#)
    - [7.2.1.5 Live presentation: Upcoming structural annotation at cosmoss.org using gbrowse and Apollo](#)
- ◆ [7.3 Third Day](#)
  - ◇ [7.3.1 Morning Session](#)
    - [7.3.1.1 Invited talk: Annotating MADS-box Genes and KNOX Genes in the Physcomitrella Genome](#)
    - [7.3.1.2 Invited talk: Gene Annotation in Transcriptomic Analysis](#)
    - [7.3.1.3 Lecture: Annotation Guidelines, Student Annotation Project and Moss Ontology](#)
    - [7.3.1.4 Lecture: Annotation examples](#)

## Sponsors



**FRIAS**  
FREIBURG INSTITUTE FOR ADVANCED STUDIES  
ALBERT-LUDWIGS-UNIVERSITÄT FREIBURG



Organization is in cooperation with Kongress & Kommunikation GmbH,  
<http://www.kongress-und-kommunikation.de>

## Information

- [Have a look at previous workshops](#)
- Spread the news: [Workshop Announcement Poster](#)

## Cosmoss accounts

You have to be a registered cosmoss user in order to access some of linked material.

For the workshop participants, accounts have been created (unless you already had one) with an initial password handed out at the registration desk. Please change it as soon you [log in](#).

Everyone else is also invited to register! Please contact [helpdesk-cosmoss@uhura.biologie.uni-freiburg.de](mailto:helpdesk-cosmoss@uhura.biologie.uni-freiburg.de) for an account!

## Registration

- **Deadline for registration:** August 29, 2010.
- **Registration fee:** 200?

## Program

Learn how to work with cosmoss.org and the *Physcomitrella patens* genome. Find and annotate your gene(s) of interest. Get in touch with other people working with *Physcomitrella* and other bryophytes.

## Covered topics

Tutorial sessions for the computational tools and databases available on cosmoss.org. E.g.:

- BLAST
- Sequence Retrieval
- Genome Browser
- Functional Annotation Interface *genonaut*
- Structural Annotation Interface [Apollo](#)

Annotation jamboree:

- Invited Speakers on gene annotation (see [below](#))

- Functional and structural annotation *Howto*
- Hands-on annotation session (annotate your genes of interest)

## Schedule

### Monday September 13

**09:00 13:00** Registration

**09:30 13:15** **Morning session including lunch**

**09:30** Welcome

**09:30 10:00** The *Physcomitrella* Genome

**10:00 10:30** cosmoss.org: resources, databases and versions

**10:30 13:15** Welcome reception, industry exhibits and lab tours

**13:30 17:30** **Afternoon session including coffee break**

**13:30 14:00** Lecture: Homology searching / BLAST

**14:00 14:50** Hands-on: BLAST

**14:50 15:15** Lecture: gene families

**15:15 15:30** Coffee break

**15:30 16:00** Hands-on: gene family tools

**16:00 16:40** Lecture: sequence retrieval

**16:40 17:15** Hands-on: sequence retrieval/keyword search

**17:30** ? Social event: Bus trip to a typical south German locality (Strauße) including evening meal

---

### Tuesday September 14

**09:00 12:00** **Morning session including coffee break**

**09:00 10:00** Lecture: Genome browser and Hands-on: genome browser basics

**10:00 10:15** Lecture: Genome Browser Tracks

**10:15 10:30** Coffee break

**10:30 11:00** Lecture: Genome Browser hidden treasures

**11:00 12:00** Hands-on: Advanced gbrowsing & Questions

**12:00 13:30** Poster session & Lunch

**13:30 17:30** **Afternoon session including coffee break**

**13:30 14:00** Lecture: functional annotation

**14:00 15:00** Hands-on: genonaut

**15:15 15:30** coffee break

**15:30 16:00** Lecture: structural annotation (Gbrowse)/Apollo annotation editor

**16:00 17:00** Hands-on: structural annotation

**17:30** ? Social event: Conference dinner and industry night with talks on *Physcomitrella*/genome-specific applications

**18:00 18:20** BMG

**18:20 18:40** Agrisera

**18:40 19:00** Genxpro

19:00 19:20 High-throughput transcriptomics in *Physcomitrella*

19:20 ? Dinner

---

### Wednesday September 15

09:00 12:00 Morning session including coffee break

09:00 09:30 Annotating MADS-box Genes and KNOX Genes in the *Physcomitrella* Genome

09:30 10:00 Gene Annotation in Transcriptomic Analysis

10:15 10:30 Coffee break

10:30 12:00 Annotation guidelines, Evidence Codes, examples (Student Annotation Project) and discussion

12:00 13:30 Poster session & Lunch

13:30 17:00 Afternoon session (Annotation Jamboree) including coffee break

13:30 15:15 Gene family assignment (publications) / Jamboree

15:15 15:30 Coffee break

15:15 16:30 Jamboree

16:30 17:00 Concluding remarks / discussion

## Poster session

Carolin Aldinger

Examination of the annotated tRNA<sup>Arg</sup> GCG in *Physcomitrella patens*

Alexandra Castro

Differentially expressed genes in response to pathogens in *Physcomitrella patens* cancelled

Mahmoud Madgy

The genetic structure of the *Funaria hygrometrica* populations in Sierra Nevada (Spain) using the sequence-related amplified polymorphism (SRAP) technique

Bernadeta Kukuczka

Characterization of iron-deficiency responses and targeted knockout of the pgr11 gene in *Physcomitrella patens*

James Llyod

Can moss read nonsense? Exploring gene regulation by nonsense-mediated decay in *Physcomitrella*

Stefanie Müller

Mitochondrial proteomics in the moss *Physcomitrella patens*: Are there two subpopulations of mitochondria?

Sergio Pisa

Genetic diversity of *Bryum argenteum* Hedw. along an altitudinal gradient in Sierra Nevada, Spain

Cecilia Ruibal

Search for genes involved in abiotic stress tolerance in *Physcomitrella patens* cancelled

## Venue

University of Freiburg, Germany

## Biology I

- Campus, Hauptstr. 1, Computer Pool & Lounge
- main workshop location

[Google map of the location](#)

## Biology II

- Campus, Schänzlestr. 1, Main Entrance, Small Lecture Hall & Lecture Hall Foyer
- morning session on the first day
- the registration desk is located in the Lecture Hall Foyer

[Google map of the location](#)

## Speakers

- Elizabeth Barker (Invited talk: [Annotating MADS-box genes and KNOX genes in the \*Physcomitrella\* genome](#))
- Andrew C. Cuming (Invited talk: [Gene annotation in transcriptomic analysis](#))
- [Stefan Rensing](#)
- Andreas Zimmer
- [Daniel Lang](#)

## Workshop Content

### First Day

#### Morning Session

location

[Biology II Small Lecture Hall](#)

#### The *Physcomitrella* Genome

speaker

Stefan

slides

[Genome.pptx](#)

**cosmoss.org: resources, databases and versions**

speaker

Daniel

slides

[cosmoss\\_resources\\_databases\\_versions.pdf](#)

---

**Afternoon Session**

location

[Biology I Computer Lab](#)

**Lecture: Homology searching / BLAST**

speaker

Stefan

slides

[BLAST.pptx](#)

**Hands-on: BLAST**

speaker

Stefan

slides

[BLAST\\_hands-on.pptx](#)

**Lecture: gene families**

speaker

Stefan

slides

[GeneFamilies.pptx](#)

**Hands-on: gene family tools**

speaker

Stefan

slides

[GeneFamiliesHands-On.pptx](#)

**Lecture: sequence retrieval and genome browser**

speaker

Andreas

slides

[Sequence Retrieval 2010.pptx](#)

## **Hands-on: sequence retrieval/keyword search**

speaker

Andreas

### **Sequence Retrieval**

documentation

[Sequence Retrieval](#)

Familiarize yourself with the Cosmoss Retrieval system:

Tasks

### **Try to retrieve the following sequences:**

Pp1s1\_770V6.1  
Pp1s1\_128V6.2  
Phypa\_430365

You can access the sequence retrieval via the transcriptome and genome menu.

Hint: These accession numbers above are from the annotated V1.6 P.patens transcripts database. You can retrieve multiple sequences by providing their accession numbers as a comma- or space-separated

Have a look at the receiving FASTA files.

What are the first five bases of Phypa\_430365 transcript sequence? \*mark text from here --> ACTA  
What is the description of Pp1s1\_128V6.2 ? \*mark text from here --> patatin t5<-- to here for the  
What is the CGI (cosmoss gene id) of Phypa\_430365 ? \*mark text from here --> Pp1s33\_278V6.1<-- t

As you may have already noticed, you could retrieve your P.patens sequence (V1.1, V1.2, V1.6) of interest using the Phypa\_id or CGI. e.g. Phypa\_430390 or Pp1s33\_43V6.1

### **Batch retrieval**

For a vast number of sequences you could upload your request in a text file.

File format: text  
Provide accession number per line.

**Select the following accession numbers and copy them into a new text file (e.g. notepad) and save it.**

Pp1s459\_1V2.1



## Cosmoss\_workshop

Pp1s7\_181V2.1  
Pp1s204\_88V2.1  
Pp1s204\_91V2.1  
Pp1s352\_13V2.1  
Pp1s312\_46V2.1  
Pp1s56\_169V2.1  
Phypa\_125839  
Pp1s56\_166V2.1  
Phypa\_125903  
Pp1s188\_40V2.1  
Pp1s545\_4V2.2  
Pp1s109\_143V2.1  
Pp1s352\_57V2.1  
Pp1s66\_46V2.1  
Pp1s12\_223V2.1  
Pp1s46\_41V2.1  
Phypa\_146969  
Phypa\_87740  
Phypa\_87752  
Phypa\_151552  
Phypa\_8310  
estExt\_fgenesht1\_pg.C\_330027  
EDQ76486  
XM\_001758564

**Goto the cosmoss.org sequence retrieval:**

**Select database: P.patens\_all\_models\_transcripts**

This all models database contains all P.patens gene predictions (V1.0 V1.1 V1.2 V1.6) proteins.

The *Physcomitrella patens* genome accession numbers work for both transcripts and proteins databases. Just change the database to change to your favored sequence type.

**Browse for the previously created file and submit your request**

**Save the sequences in a new file in FASTA format**

**Select only a subset of the sequences and save it to a new file**

### Keyword search

documentation

Keyword search

How many *geranyl geranyl pyrophosphate synth(et)ases* (GGPS) are in the virtual transcriptome?

Try to find the corresponding pp0304 virtual transcripts by keyword search!

Tasks

1. Read the documentation
2. Use the simple search menu to find the GGPS's in pp0304
3. Play around with the advanced search option.

## Cosmoss\_workshop

4. How many geranyl geranyl pyrophosphate synthase hits to you find with the keyword search in the V1.6 transcripts?

Hits in pp0304

\*mark text from here --> 3 Hits in the pp0304 transcripts <-- to here for a possible query\*

Hits in V1.6 transcripts

\*mark text from here --> 5 Hits on 4 loci<-- to here for a possible query\*

This is the advanced query that works:

\*mark text from here --> "geranyl geranyl"[DESC] AND "phosphate"[DESC] AND "synth"[DESC] <-- to h

Finally, here are the four loci in the genome:

- [Pp1s167\\_155V6.1](#)
- [Pp1s3\\_343V6.1](#)
- [Pp1s55\\_266V6.1](#)
- [Pp1s8\\_131V6.1 Pp1s8\\_131V6.2](#)

## Second Day

### Morning Session

#### Lecture: Genome Browser Basics

speaker

Andreas

slides

[Zimmer\\_GenomeBrowser\\_10.pptx](#)

#### Hands-on: Genome Browser Basics

speaker

Andreas

slides

[Hands\\_on\\_gbrowse\\_basics.pptx](#)

## Lecture: Genome Browser hidden treasures

speaker

Daniel

slides

[cosmoss\\_gbrowse\\_treasures.pdf](#)

### **Hands-on experience**

Genome browser: customization and special features

prepared by

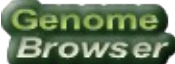
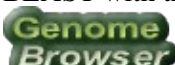
Daniel

### **BLAST gbrowse integration**

documentation

[BLAST\\_gbrowse\\_integration](#)

Tasks

1. BLAST with an arbitrary transcript/EST vs the scaffolds and follow the  link.
2. Compare results when BLASTing w/o low complexity filtering!
3. Compare the BLAST to the spliced-alignment results. Are there lonely exons?
4. BLAST with the *Arabidopsis* protein AT5G13930.1 vs the v1.2 gene models and follow the  link.
5. Compare one of the hit *Physcomitrella* loci vs the *Arabidopsis* locus [AT5G13930](#)

### **Advanced navigation and zooming**

Tasks

1. Find out the definition of a *gene* in [SQ](#) by using the *Ontology\_term* cross-link in a gene feature's mouse-over window
2. Zoom into a CDS exon and back again to the region of its *mommy* mRNA or gene
3. Play around with the zoom function to inspect EST and cDNA spliced alignments!

### **Exporting sequence annotations and publication quality images**

Tasks

1. Export the upstream 10kbp your most favorite region to [FASTA](#) format and reverse it when necessary to reflect the gene's orientation.
2. *OPTIONAL*: Get another upstream region and try to find shared putative promoter elements using e.g. [AlignACE](#)
3. Save your most favorite region as a [png](#) image.

## Cosmoss\_workshop

4. Check out the exemplary PDF created with Inkscape from the SVG of a region:  
[www.cosmoss.org/inkscape\\_example.pdf](http://www.cosmoss.org/inkscape_example.pdf)

### Highlighting

example locus

[scaffold\\_29:573541..579040](#)

documentation

[available colors](#) (scroll down to *Colors*)

Tasks

1. Let gbrowse highlight a feature and zoom out again, in order to see whether it overlaps with another feature in another track (*highlight and zoom to the region of this feature* link in the mouse-over)
2. Highlight your favorite gene model for the locus using the *Highlight feature* box in the *Display panel*.
3. Visualize an PCR experiment on the genomic locus! You've just amplified and sequenced a genomic PCR product for the locus using the primer coordinates below. Highlight the genomic region using the *Highlight regions* box in the *Display panel*.

forward primer mapping

[scaffold\\_29:575397..575417](#)

reverse primer mapping

[scaffold\\_29:577964..577950](#)

The screenshot shows the gbrowse web interface. At the top, there are several tracks with checkboxes: 'MIPS ANGELA long\_terminal\_repeat', 'Helitrons Bennetzen lab', and 'TE related PFAM domains'. Below these tracks are buttons for 'Configure tracks...' and 'Update'. A section titled 'Display Settings' is expanded, showing three input fields: 'Highlight feature(s) (feature1 feature2...)' with the value 'all\_Phypa\_205678@yellow', 'Highlight regions (region1:start..end region2:start..end)' with the value 'scaffold\_29:577964-577950', and 'Region Size (bp)' with a dropdown menu set to '10 kbp'. There is an 'Update' button at the bottom right of this section. Below the 'Display Settings' section is another section titled 'Add your own tracks'.

### Displaying custom annotation

You can draw your own features and have your own custom track in the browser!

documentation

[gbrowse custom annotation help](#)

Exporting sequence annotations and publication quality images

example locus

[scaffold\\_29:573541..579040](#)

Tasks

1. Go through the documentation
2. Visualize an RT-PCR experiment on the genomic locus! You've just amplified and sequenced an RT-PCR product for the locus below. Display it as custom annotation using the coordinates:

```
RT-exon1 scaffold_29:575397..575570
RT-exon1 scaffold_29:576849..576875
RT-exon1 scaffold_29:577042..577335
RT-exon1 scaffold_29:577629..577692
```

See the [example file](#) which combines everything. Adjust it, if you like!

## Lecture: functional annotation

speaker

Daniel

slides

[cosmoss\\_genonaut.pdf](#)

## Live presentation: Upcoming structural annotation at cosmoss.org using gbrowse and Apollo

speaker

Daniel

## Third Day

### Morning Session

#### Invited talk: Annotating MADS-box Genes and KNOX Genes in the *Physcomitrella* Genome

speaker

Elizabeth Barker

abstract

[Talk.Elizabeth\\_Barker.abstract.pdf](#)

slides

[Barker\\_Cosmoss\\_Workshop\\_2010.ppt](#)

#### Invited talk: Gene Annotation in Transcriptomic Analysis

speaker

Andrew Cuming

Displaying custom annotation

abstract

[Talk.Andrew.Cuming.abstract.pdf](#)

slides

[Cuming\\_Freiburg\\_2010.ppt](#)

### **Lecture: Annotation Guidelines, Student Annotation Project and Moss Ontology**

speaker

Daniel

slides

[Annotation.pdf](#)

### **Lecture: Annotation examples**

speaker

Stefan

slides

[AnnotationExamples.pptx](#)