Pathogen emergence: Evolution of parasitism and host adaptation
Pathogen emergence

QUESTION: Evolution of signalling pathways
QUESTION: Evolution of signalling pathways

→ The dauer signalling pathways
QUESTION: Evolution of signalling pathways

The dauer signalling pathways

*C. elegans* life cycle
QUESTION: Evolution of signalling pathways

The dauer signalling pathways

*C. elegans* life cycle

Daure larva:
- Arrested developmental stage
- Non-feeding
- Morphological characteristics
- ↓ metabolism
- Resistant to environmental stresses
QUESTION: Evolution of signalling pathways

The dauer signalling pathways

Parasitic nematodes’ life cycle

- Infective larva (iL)  
  Specialised stage for infestation

- adult
- egg
- L4
- L3
- L2
- L1
QUESTION: Evolution of signalling pathways

The dauer signalling pathways

Parasitic nematodes’ life cycle

- Egg
- L2
- L3
- L4
- Adult
- Infective larva (iL)
- Specialised stage for infestation

Trichinella spiralis
QUESTION: Evolution of signalling pathways

The dauer signalling pathways

Parasitic nematodes’ life cycle

Infective larva (iL)
Specialised stage for infestation

Meloidogyne hapla
QUESTION: Evolution of signalling pathways

The dauer signalling pathways

Parasitic nematodes’ life cycle

Infective larva (iL)  Specialised stage for infestation

Haemonchus contortus
QUESTION: Evolution of signalling pathways

The dauer signalling pathways

Parasitic nematodes’ life cycle

Infective larva:

- Arrested developmental stage
- Non-feeding
- Morphological characteristics
- \( \downarrow \) metabolism
- Resistant to environmental stresses
Dauer hypothesis

Pre-adaptation to parasitism

⇒ Multiple independent transitions

Blaxter & Koutsovoulos (2015) Parasitology, 142 (S1): S26-S39
Dauer hypothesis

Pre-adaptation to parasitism

Existence of common genetic pathways that would likely control the dauer transition across the phylum?

Gene candidate approach

Multiple independent transitions

Blaxter & Koutsovoulos (2015) Parasitology, 142 (S1): S26-S39
Bioinformatic search of orthologues
- 47 genes involved in the dauer transition
- 24 species

Evolution of parasitism in nematodes
- Free-living
- Animal parasites
- Plant parasites
Bioinformatic search of orthologues
- 47 genes involved in the dauer transition
- 24 species

Free-living Animal parasites Plant parasites
Bioinformatic search of orthologues

- 47 genes involved in the dauer transition
- 24 species

**Proteomes**

- orthoMCL
- Reciprocal BLASTP
- MEME suite

Gene models

- Gene-Sp.1
- Gene-Sp.2
- Gene1-Sp.1
- Gene2-Sp.1
- Gene1-Sp.2
- Gene2-Sp.2

Free-living Animal parasites Plant parasites
Bioinformatic search of orthologues
- 47 genes involved in the dauer transition
- 24 species
Bioinformatic search of orthologues
- 47 genes involved in the dauer transition
- 24 species

Genomes

- Reciprocal TBLASTN
- DNA reads-to-protein alignments
- Protein-to-genome alignments
- ARN-to-ADN alignments
- RAxML
- spaln
- FigMOP

Orthologues

Free-living
Animal parasites
Plant parasites
Exons
CDS
Bioinformatic search of orthologues
- 47 genes involved in the dauer transition
- 24 species

Other search strategies
- Not found
- Reciprocal BLAST
- Minor changes
- Major changes

EVOLUTION OF PARASITISM IN NEMATODES
EVOLUTION OF PARASITISM IN NEMATODES

- Not found
- Reciprocal BLAST
- Minor changes
- Major changes

Other search strategies

- Insuline receptor DAF-2
- PDK-1
- AKT-1
- AKT-2
- FTT-2
- PAR-5
- DAF-16
EVOLUTION OF PARASITISM IN NEMATODES

Non-Caenorhabditis

Insuline receptor DAF-2

PDK-1

AKT-1

AKT-2

FTT-2

DAF-16

DAF-16

Other search strategies

Not found  Reciprocal BLAST  Minor changes  Major changes
EVOLUTION OF PARASITISM IN NEMATODES

DAF-7

TGF-β receptors

DAF-1/DAF-4

DAF-8

DAF-4

DAF-3

DAF-5

Phenotypes:
Dauer development
Egg-laying

Other search strategies

Not found  Reciprocal BLAST  Minor changes  Major changes
EVOLUTION OF PARASITISM IN NEMATODES

Phenotypes:
- Dauer development
- Egg-laying
- Dauer development

Other search strategies:
- Not found
- Reciprocal BLAST
- Minor changes
- Major changes
Co-option of the DAF-7–TGF-β pathway into dauer control
- Fundamental ancestral role
- Dauer control in clade V nematodes

Phenotypes:
- Dauer development
- Egg-laying

Other search strategies
- Not found
- Reciprocal BLAST
- Minor changes
- Major changes
EVOLUTION OF PARASITISM IN NEMATODES

⇒ Gene duplications, gene losses & pathway co-option

Other search strategies

- Not found
- Reciprocal BLAST
- Minor changes
- Major changes
HOST ADAPTATION IN *Plasmodium* species
QUESTION: Human pathogens emergence

Plasmodium falciparum & Plasmodium vivax, malaria causing pathogens
HOST ADAPTATION IN *Plasmodium* species

- *P. reichenowi*
- *P. falciparum*
- *P. praefalciparum 1*
- *P. praefalciparum 2*
- *P. billcollinsi*
- *P. blacklocki*
- *P. gaboni*
- *P. billbrayi*
- *P. adleri*
- *P. cynomolgi*
- *P. vivax-like*
- *P. vivax*

HOST ADAPTATION IN *Plasmodium* SPECIES

- **Origins**
- **Host adaptation**
- **Comparative genomics**

*Plasmodium* species:
- *P. reichenowi*
- *P. falciparum*
- *P. praefalciparum 1*
- *P. praefalciparum 2*
- *P. billcollinsi*
- *P. blacklocki*
- *P. gaboni*
- *P. billbrayi*
- *P. adleri*
- *P. cynomolgii*
- *P. vivax-like*
- *P. vivax*

Sources:
- Human
- Chimpanzee
- Gorilla
- Rodents
- Asian monkeys
Comparative genomics & bioinformatics

- **Gene transfer, convergent evolution**
- **Selection**
- **Multi-genic families**

Diversity & genetic differentiation (SNPs)

**PLASMODIUM SPECIES**

- **P. reichenowi**
- **P. falciparum**
  - **P. praefalciparum 1**
  - **P. praefalciparum 2**
- **P. billcollinsi**
- **P. blacklocki**
- **P. gabin**i
- **P. billbrayi**
- **P. adleri**

Plasmodium:
- **P. cynomolgi**
- **P. vivax-like**
- **P. vivax**

**HOST ADAPTATION**

- Human
- Chimpanzee
- Gorilla
- Rodents
- Asian monkeys

SNP: Single Nucleotide Polymorphism
**Laverania genomics**

Technical challenges:
- Low parasitemia
- Co-infections
- Host contamination
- AT rich
**Laverania genomics**

Technical challenges:
- Low parasitemia
- Co-infections
- Host contamination
- AT rich

⇒ CF11 cellulose columns, whole genome amplification (WGA) & PacBio

long reads
(mean = 10kb)
Sample | Sample preparation
--- | ---
P. reichenowi | Fresh blood | CF11 / WGA
P. falciparum | Fresh blood | CF11 / WGA
P. praefalciparum 1 | Fresh blood | CF11 / WGA
P. praefalciparum 2 | Fresh blood | CF11 / WGA
P. billcollinsi | Fresh blood | CF11 / WGA
P. blacklocki | Frozen blood | CF11 / WGA + sWGA
P. gaboni | Fresh blood | CF11 / WGA
P. billbrayi | Fresh blood | CF11 / WGA
P. adleri | Fresh blood | CF11 / WGA
## Host Adaptation in *Plasmodium* Species

<table>
<thead>
<tr>
<th>Sample</th>
<th>Sample preparation</th>
<th># genes</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>P. reichenowi</em></td>
<td>Fresh blood</td>
<td>5941</td>
</tr>
<tr>
<td><em>P. falciparum</em></td>
<td>CF11 / WGA</td>
<td>5548</td>
</tr>
<tr>
<td><em>P. praefalciparum 1</em></td>
<td>Fresh blood</td>
<td>6476</td>
</tr>
<tr>
<td><em>P. praefalciparum 2</em></td>
<td>CF11 / WGA</td>
<td></td>
</tr>
<tr>
<td><em>P. billcollinsi</em></td>
<td>Fresh blood</td>
<td>5637</td>
</tr>
<tr>
<td><em>P. blacklocki</em></td>
<td>Frozen blood</td>
<td>5346</td>
</tr>
<tr>
<td><em>P. gaboni</em></td>
<td>Fresh blood</td>
<td>5421</td>
</tr>
<tr>
<td><em>P. billbrayi</em></td>
<td>CF11 / WGA</td>
<td></td>
</tr>
<tr>
<td><em>P. adleri</em></td>
<td>Fresh blood</td>
<td>5515</td>
</tr>
</tbody>
</table>

### 4269 one-to-one orthologues
HOST ADAPTATION IN *Plasmodium* species

Species tree

- **P. adleri**
- **P. gaboni**
- **P. blacklocki**
- **P. billcollinsi**
- **P. reichenowi**
- **P. praefalciparum**
- **P. falciparum**

Reference genome assemblies

Chromosome 4

# scaffolds
Gene transfer & convergent evolution

Species tree topology

Pgab
Padl
Pbko
Pbcl
Prei
Ppra
Pfal

4269 CDS
Gene transfer & convergent evolution

Species tree topology

- Pgab
- Padl
- Pbko
- Pbcl
- Prei
- Ppra
- Pfal

4269 CDS $\Rightarrow$ 4251 CDS
Gene transfer & convergent evolution

Species tree topology

Host-mediated convergent evolution topology

4269 CDS  $\Rightarrow$  4251 CDS
Gene transfer & convergent evolution

Species tree topology

- Pgab
- Padl
- Pbko
- Pbcl
- Prei
- Ppra
- Pfal

Host-mediated convergent evolution topology

- Pgab
- Pbcl
- Prei
- Padl
- Pbko
- Ppra
- Pfal

4269 CDS  ⇒  4251 CDS

⇒ 0 CDS
Gene transfer & convergent evolution

Species tree topology
- Pgab
- Padl
- Pbko
- Pbcl
- Prei
- Ppra
- Pfal

Host-mediated convergent evolution topology
- Pgab
- Pbcl
- Prei
- Padl
- Pbko
- Ppra
- Pfal

4269 CDS ⇒ 4251 CDS

⇒ 0 CDS

⇒ 4 CDS
Gene transfer & convergent evolution
Gene transfer & convergent evolution

CDS & intergenic regions

Species tree topology

Pgab
Padl
Pbko
Pbcl
Prei
Ppra
Pfal

Species tree topology

Pgab
Padl
Ppra
Pfal
Pbko
Pbcl
Prei

Species tree topology

Pgab
Padl
Pbko
Pbcl
Prei
Ppra
Pfal
Gene transfer & convergent evolution

CDS & intergenic regions

Species tree topology

=> Gene transfer
Convergent evolution

Fixed differences between host species

- *P. adleri*
  - LFIPS YNTYD NITYND
- *P. gaboni*
  - LFIPS YNSYD NIANN
- *P. blacklocki*
  - LFIPS NNTYD NVOND
- *P. billcollinsi*
  - LFIPS NNLYD NMAAND
- *P. reichenowi*
  - LFIPS NNLYD NVAND
- *P. praefalciparum*
  - LFIPS NKTYD NVOND
- *P. falciparum*
Convergent evolution

Fixed differences between host species
Convergent evolution

Fixed differences between host species

- P. adleri
- P. gaboni
- P. blacklocki
- P. billcollinsi
- P. reichenowi
- P. praefalciparum
- P. falciparum

Human Chimpanzee Gorilla

- P230
- doc-2
HOST ADAPTATION IN *PLASMODIUM* SPECIES

- **Convergent evolution**
  - Fixed differences between host species

- **Gene ontology enriched:**
  - Erythrocyte invasion

**Figure:**
- *P. adleri*, *P. gaboni*, *P. blacklocki*, *P. billcollinsi*, *P. reichenowi*, *P. praefalciparum*, *P. falciparum*
- Graphs showing distribution of fixed differences in genes among these species.
- **X-axis:** Position (x 10^7 nucleotide)
- **Y-axis:** Number of fixed differences in genes

**Legend:**
- Human
- Chimpanzee
- Gorilla

**Data:**
- PF3D7_0424000
- EBA-165
- RH5
- RH4
- CyRPA
- PF3D7_0423900

**Scale:**
- 0, 1, 2, 3, 4, 5
- 1,080,000, 1,080,000
HOST ADAPTATION IN *Plasmodium* species

⇒ Gene transfer & convergent evolution

**P. falciparum** Chr. 4

cysteine-rich protective antigen

reticulocyte binding protein homologue 5

○ CyRPA/Ripr/RH5 complex
  ⇒ Interaction with the human receptor basigin
  ⇒ Erythrocyte invasion & host tropism

6-cysteine protein P230
(pre-zygotic reproductive barrier)

DOC2
(Microneme secretion)

RH3, RH5 + CyRPA, EBA-175

Thank you!

James Wasmuth
Dave Curran
Brian McDonald
Keyu Li
Jeff Wintersinger
Ivan Kryukov

John Gillearad

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Franck Prugnolle
François Renaud
Virginie Rougeron

Matthew Berriman
Ulrike Böhme
Thomas D Otto
Samuel Oyola
Mandy Sanders

Chris Newbold