Sponges regeneration and reaggregation of cells experiment

Summer course in embryology of marine invertebrates
WSBS, Russia
CONTENT

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  - Regeneration in marine invertebrates
  - Why study regeneration and reaggregation with sponges?

- Methods
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  - Field work sampling
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  - Experiment one: Regeneration
  - Experiment two: Reaggregation of cells

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INTRODUCTION

- Regeneration in marine invertebrates
  - Is the response to an external amputation (Bely, 2006)
  - Is a process by which many animals can replace lost body parts (Bely & Sikes, 2010)
  - Ability to regenerate is widespread in the animal kingdom. To understand the evolutionary history of the diverse regeneration mechanism, the regeneration processes must be studied in early-development metazoans (Borisenko et al., 2015)
INTRODUCTION

Why study regeneration and reaggregation in sponges?

- Sponges - Basal metazoans
- No true tissue grade organisation
- High regenerative capacity
- High plasticity of cells / ability to transdifferentiate
- Continuous cell movements and rearrangement of anatomical structure
METHODS

Selected species

*Sycon sp.*

Calcareous sponge. Small size, and are tube-shaped and often white to cream in colour

*Leucosolenia complicata*

Shallow sublitoral along most rocky coasts of Europe
METHODS

Selected species

Halisarca dujardinii

Common species in littoral habitats along the European coasts

0.8 cm

Clathrina cf. blanca

Comprises anastomosed tubes. Asconoid aquiferous system

0.5 cm
METHOD

Field work sampling

White sea
Intertidal zone low tide 16:30
Collection of *Sycon* sp. and *Leucosolenia complicata*
Underwater, never let the organisms touch the air
Lab Materials

- Medium size Petri dishes
- Tissue culture plates
- Forceps
- Blades
- Pasteur pipette
- Microscope
- Filtered sea water
- Fridge 10C
- Needles
METHOD

Experiment one: Regeneration

Cut the sponge in 5 layers

Sycon sp.

A1
A2
I
B1
B2

A1
A2
I
B1
B2

3 h 6h 12h 24h 48h

10°C
METHOD

S

❖ Experiment one: Regeneration

_Halisarca dujardinii_

Superficial cut to the sponge

10°C

3 h  6h  12h  24h  48h
Experiment one: Regeneration

*Leucosolenia complicata*

Small cut to the sponge

10°C

3 h  6h  12h  24h  48h
METHODS

 Experiment two: aggregation

Halisarca dujardini

Leucosolenia complicata

Cut into pieces of 1 mm

10°C

3 h  6h  12h  24h  48h
# RESULTS AND DISCUSSION

- **Experiment one: Regeneration**

<table>
<thead>
<tr>
<th>Specie</th>
<th>Time period</th>
<th>Membrane formation</th>
<th>Membrane covering</th>
<th>Full regeneration</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Sycon</em> sp.</td>
<td>3 h</td>
<td>NO</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>6h</td>
<td>NO</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>12h</td>
<td>Small membrane formation</td>
<td>Membrane covering 50% of the opening</td>
<td></td>
</tr>
<tr>
<td></td>
<td>24h</td>
<td></td>
<td>Membrane covering 50% of the opening</td>
<td>Full regeneration</td>
</tr>
<tr>
<td></td>
<td>48h</td>
<td></td>
<td>Membrane covering the opening</td>
<td></td>
</tr>
</tbody>
</table>
RESULTS AND DISCUSSION

- Experiment one: Regeneration

*Sycon sp. A2*

12h

24h

48h
RESULTS AND DISCUSSION

- Experiment one: Regeneration
  
  *Sycon* sp.

24h

A1  A2

I   B1

B2
RESULTS AND DISCUSSION

- **Experiment one: Regeneration**

<table>
<thead>
<tr>
<th>Specie</th>
<th>Time period</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Halisarca dujardini</em></td>
<td>3 h 6h 12h 24h 48h</td>
</tr>
<tr>
<td>NO</td>
<td>NO</td>
</tr>
<tr>
<td>Wound surface</td>
<td>Archeocytes accumulate under the wound surface</td>
</tr>
<tr>
<td>recovered</td>
<td>New differentiating exopinacoderm</td>
</tr>
</tbody>
</table>

Borisenko *et al.*, 2015
RESULTS AND DISCUSSION

- Experiment one: Regeneration

<table>
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<th>Time period</th>
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<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Leucosolenia complicata</em></td>
<td>3h</td>
<td>6h</td>
<td>12h</td>
<td>24h</td>
<td>48h</td>
</tr>
<tr>
<td>NO</td>
<td>NO</td>
<td>Body wall contraction</td>
<td>Membrane covering 50% of the opening</td>
<td>Full regeneration membrane covering the opening</td>
<td></td>
</tr>
</tbody>
</table>
RESULTS AND DISCUSSION

- Experiment one: Regeneration

*Leucosolenia complicata*
RESULTS AND DISCUSSION

- Experiment two: Reaggregation

*Halisarca dujardini*
RESULTS AND DISCUSSION

- Experiment two: Reaggregation

*Leucosolenia complicata*
CONCLUSIONS

- Regeneration and aggregation experiments in sponges seems to be a good model to assess that capability due to can be measured in short time and also comprises many processes that can be followed in laboratory.

- *H. dujardini* experiment was which showed that reaggregation of cells have the capability to end in regeneration of a functional sponge.

- There are many direct and indirect factors that contribute to regeneration and reaggregation in sponges and they should be taken into account in further studies.