

What is in the R script

Background and software

In order to make our analysis as transparent as possible, we decided to use the free software R (<http://www.r-project.org>) for statistical analysis and graphic and to make all data and commands available online.

We also used R studio (<http://www.rstudio.com>): a free user-friendly interface for R.

All necessary commands and data are included in the provided zip file.

If you are using R Studio, you can use the R project to immediately open all scripts.

If you prefer to run R directly, you will need to open all scripts individually.

Content

File name	File type	Opens with	Task
sponge_project.Rproj	+ R project	R studio	Opens all necessary scripts in R studio
Rhistory			
size_mass.R	R script	R, R studio, text editor	Determines the relation between sponge surface and sponge biomass
counts.R	R script	R, R studio, text editor	Counts the sponges in each size class for each transect
transect_stat.R	R script	R, R studio, text editor	Compute statistics on transects length, width and surface
abundances.R	R script	R, R studio, text editor	Test if sponges abundances have changed between 2007 and 2011
size_frequency.R	R script	R, R studio, text editor	Plot size frequency of sponge population in 2007 and 2011
biomass.R	R script	R, R studio, text editor	Compare sponge biomass between 2007 and 2011
asteroids.R	R script	R, R studio, text editor	Test if abundance of asteroids have changed between 2007 and 2011
RV_size_mass.txt	Input data	R, R studio, text editor	Size (H,W) and mass (DM, AFDM) of <i>Rossella cf. villosa</i>
AJ_size_mass.txt	Input data	R, R studio, text editor	Size (H,W) and mass (DM, AFDM) of <i>Anoxycalyx (Scolymastra) joubini</i>
sponge_size.txt	Input data	R, R studio, text editor	Measured size (H,W) of each sponge on every transects
transect.txt	Input data	R, R studio, text editor	Measured transects length and width
sponge_count.txt	Output/Input data	R, R studio, text editor	Number of sponges in each size class on each transect
sponge_mass.txt	Output/Input data	R, R studio, text editor	Sponges DM and AFDM calculated from their size
sponge_mean_size.txt	Output/Input data	R, R studio, text editor	Mean size of sponges on each transect
asteroidss.txt	Input data	R, R studio, text editor	Number of <i>asteroids</i> on each transect

Size_mass.R

Goal

We want to use the sponges size measured in the videos to estimate the sponge biomass over our transects. As such, we need to determine the relation between sponge surface (S) and sponge biomass in terms of dry mass (DM) and ash free dry mass (AFDM) for *Anoxycalyx (Scolymastra) joubini* (AJ) and *Rossella cf. villosa* (RV), the most abundant sponges species on our transects.

Input

Sponges were collected by bottom trawling, their size measured (height-H, maximum width-W), dried and weighted before (DM) and after burning (AFDM). For the biggest sponges only subsamples were burned.

The measurements are listed in two files:

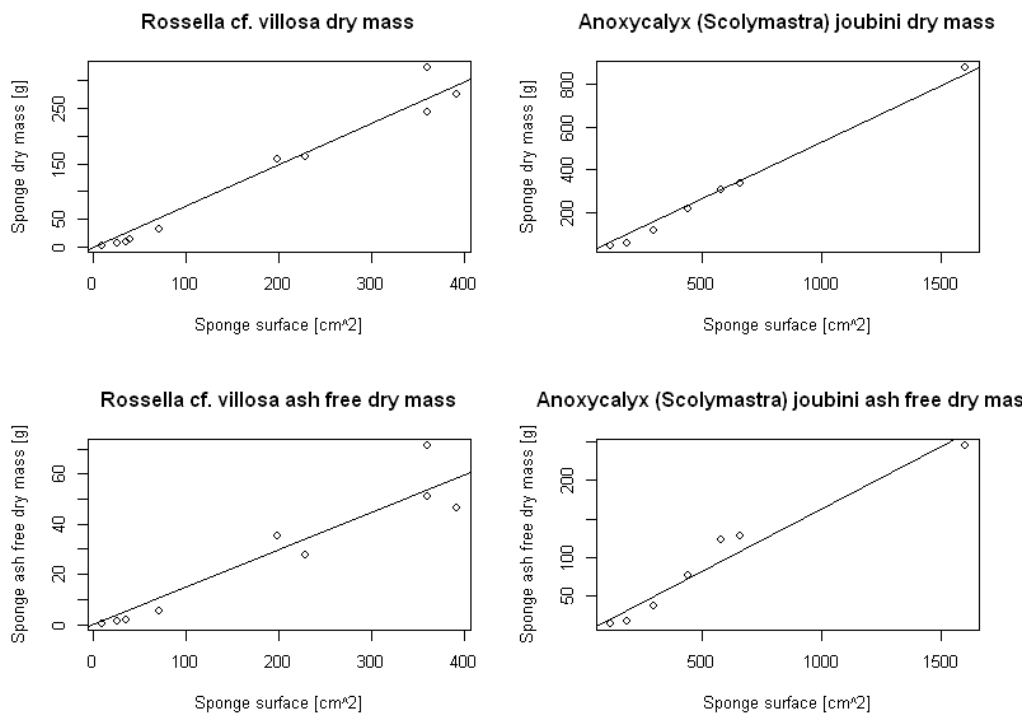
- AJ_size_mass.txt: for *Anoxycalyx (Scolymastra) joubini*
- RV_size_mass.txt: for *Rossella cf. villosa*

Steps

1. Calculate the sponge surface ($S=H*W$)
2. Calculate the sponge ash free dry mass (organic dry mass)($AFDM=DM*organic\ ratio$)
3. Calculate sponges ash dry mass (inorganic dry mass)($ADM=DM*inorganic\ ratio$)
4. Calculate mean inorganic ratio
5. Surface-dry mass regression (linear model)
6. Surface-ash free dry mass regression (linear model)

Output

- **Plot size_mass_regression**



- **Equation** and R-squared

Species	N	Inorganic ratio	Dry mass (DM)	Ash-free dry mass (AFDM)
<i>Rossella cf. villosa</i>	10	0.80 ± 0.02	DM[g] = $0.7491 * S[\text{cm}^2]$ $r^2=0.9798$	AFDM[g] = $0.1489 * S[\text{cm}^2]$ $r^2=0.9471$
<i>Anoxycalyx</i> (<i>Scolymastra</i>) <i>joubini</i>	7	0.66 ± 0.04	DM[g] = $0.5299 * S[\text{cm}^2]$ $r^2=0.9933$	AFDM[g] = $0.1629 * S[\text{cm}^2]$ $r^2=0.9787$

Counts.R

Goal

Calculate the DM and AFDM of each sponge.

Counts the sponges in each size class for each transect and the sponge mean size (surface S) on each transect.

Input

All glass sponges seen in the videos during every transect were measured together with the laser scale and, when possible, the species was identified (RV: *Rossella* cf. *villosa*, AJ: *Anoxycalyx* (*Scolymastra*) *joubini*, RA: *Rossella Antarctica*, H: hexactinellid, N: no sponges on the transect).

The measurements are listed in one file:

- sponge_size.txt

Steps

1. Calculate real size of sponges (sponge real size = sponge measured size*laser real size/laser measured size)
2. Calculate sponges surface ($S=H*W$)
3. Calculate sponges dry mass (equations obtained from size_mass.R script)(if not AJ then considered as RV)
4. Calculate sponges ash free dry mass (AFDM)(equations obtained from size_mass.R script)(if not AJ then considered as RV)
5. Classify sponges in size classes (based on the sponges surface S)
6. Counts sponges in each size class for each transect
7. Calculate the sponge mean size (surface S) for each transect

Output

- **Table** sponges_mass.txt: calculated sponges mass (ready to use)
- **Table** sponges_counts.txt: number of sponges in each size class on each transect (header "Transect_ID" must be added manually on top of the first column)
- **Table** sponges_mean_size.txt: Mean size (surface S) of sponges on each transect (header "Transect_ID" must be added manually on top of the first column)

Transect_stat.R

Goal

Compute statistics on transects length, width and surface.

Input

Transect lengths were measured from 3D models and transect widths from the lasers dots (see method and references for full details).

The measurements are listed in one file:

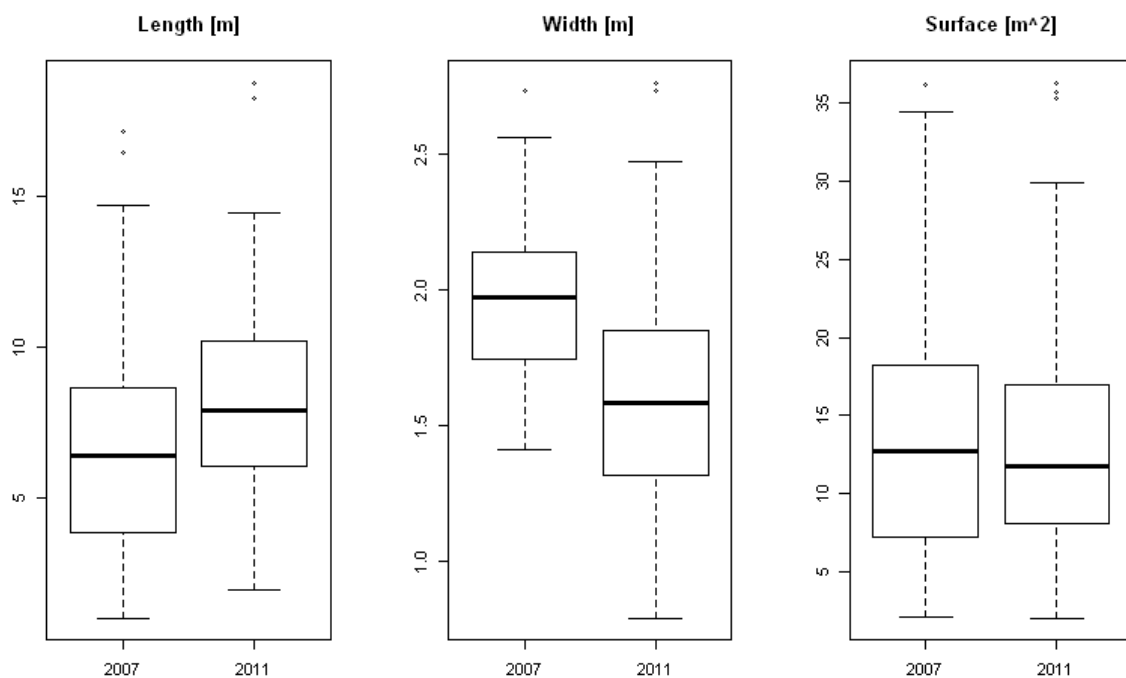
- transects.txt

Steps

1. Calculate transect surface (Surface=Length*Width)
2. Test if length, width and surface are different between 2007 and 2011

Output

- **Plot** mean length, width and surface for each year (boxplot)



- **Statistics:** Wilcoxon test (no normal distribution, non-parametric test)

Parameter	Mean 2007 N=42	Mean 2011 N=71	Alternative	P
Length [m]	6.85 ± 3.88	8.35 ± 3.40	less	0.0058 *
Width [m]	1.94 ± 0.30	1.61 ± 0.42	greater	2.518e-06 *
Surface [m ²]	13.55 ± 8.20	13.91 ± 8.05	two-sided	0.752

Abundances.R

Goal

Test if sponge abundances have changed between 2007 and 2011

Input

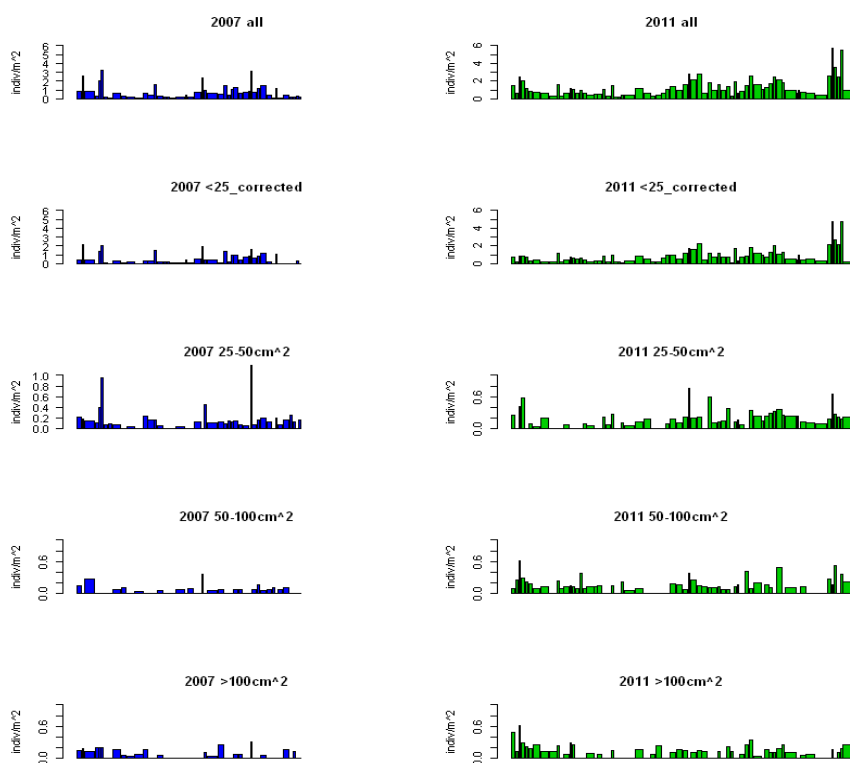
- sponges_count.txt: number of sponges in each size class on each transect (obtained from counts.R script: header "Transect_ID" must be added manually on top of the first column)
- sponges_mean_size.txt: Mean size (surface S) of sponges on each transect (obtained from counts.R script: header "Transect_ID" must be added manually on top of the first column)
- transects.txt (also used in transect_stat.R script)

Steps

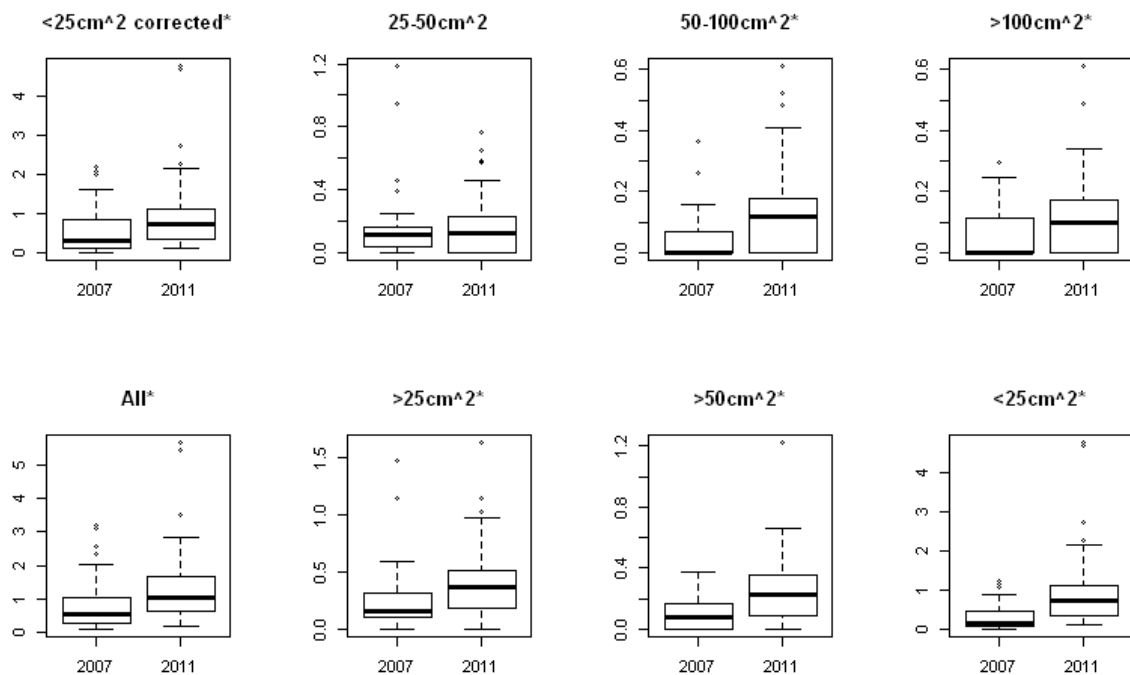
1. Calculate transects surface (Surface=Length*Width)
2. Estimate number of baby sponges ($S < 25\text{cm}^2$) with correction factor for 2007 (45% missed)
3. Sum total number of sponges for each size class in 2007 and 2011
4. Calculate sponges abundances for all size categories for each transect for each year (abundance = number of sponges on transect/surface of transect)
5. Calculate mean abundances for all size categories for each year (all transects merged)
6. Test if abundances changed in each size class between 2007 and 2011 (Wilcoxon test + permutation)

Output

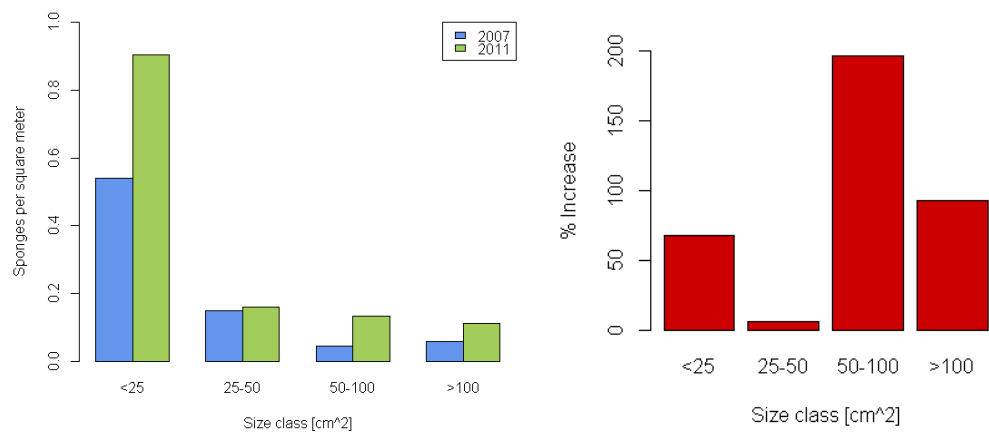
- **Plot** abundance along the transects (barplot)



- **Plot** abundance distribution during both dives (boxplot)



- **Plot** mean abundances per size class per year (barplot) and abundance increase between 2007 and 2011



- **Statistics:** Wilcoxon tests + permutation

Size class [cm ²]	Abundance 2007 Mean ± SE	Abundance 2011 Mean ± SE	Wilcoxon p	Permutation ASL
<25_uncorrected	0.30 ± 0.05	0.91 ± 0.10	5.38E-09 *	NA
<25_corrected	0.54 ± 0.09	0.91 ± 0.10	0.00046 *	0.0496
25-50	0.15 ± 0.03	0.16 ± 0.02	0.407	0.0491
50-100	0.04 ± 0.01	0.13 ± 0.02	4.02E-06*	0.0479
>100	0.06 ± 0.01	0.11 ± 0.01	0.0074 *	0.0492
all	0.79 ± 0.11	1.31 ± 0.12	0.00011*	0.0497

Size_frequency.R

Goal

Plot size frequency of sponge population in 2007 and 2011.

Input

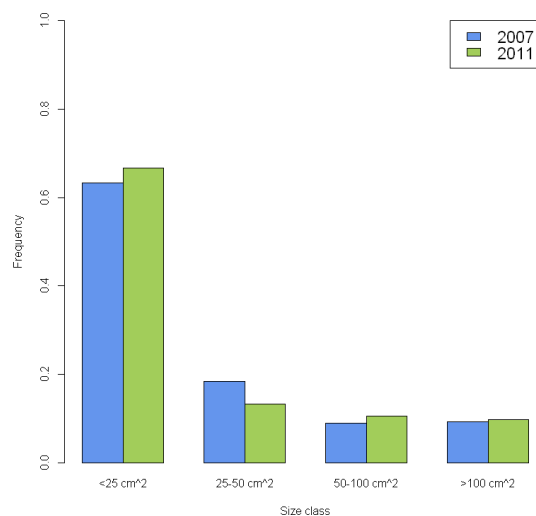
- sponge_size.txt (also used in counts.R script)

Steps

1. Calculate real size of sponges (sponge real size = sponge measured size*laser real size/laser measured size)
2. Calculate sponges surface ($S=H*W$)
3. Classify sponges in size classes
4. Counts sponges in each size class for each transect
5. Estimate number of baby sponges ($S<25\text{cm}^2$) with correction factor for 2007 (45% missed)
6. Calculate the frequency of each size class relative to the total number of sponges on all transects for both years

Output

- **Plot** size frequency distribution in 2007 and 2011



Biomass.R

Goal

Compare sponge biomass (DM and AFDM) between 2007 and 2011.

Input

- sponges_mass.txt: calculated sponges mass (obtained from counts.R script: ready to use)
- transects.txt (also used in transect_stat.R and abundances.R scripts)

Steps

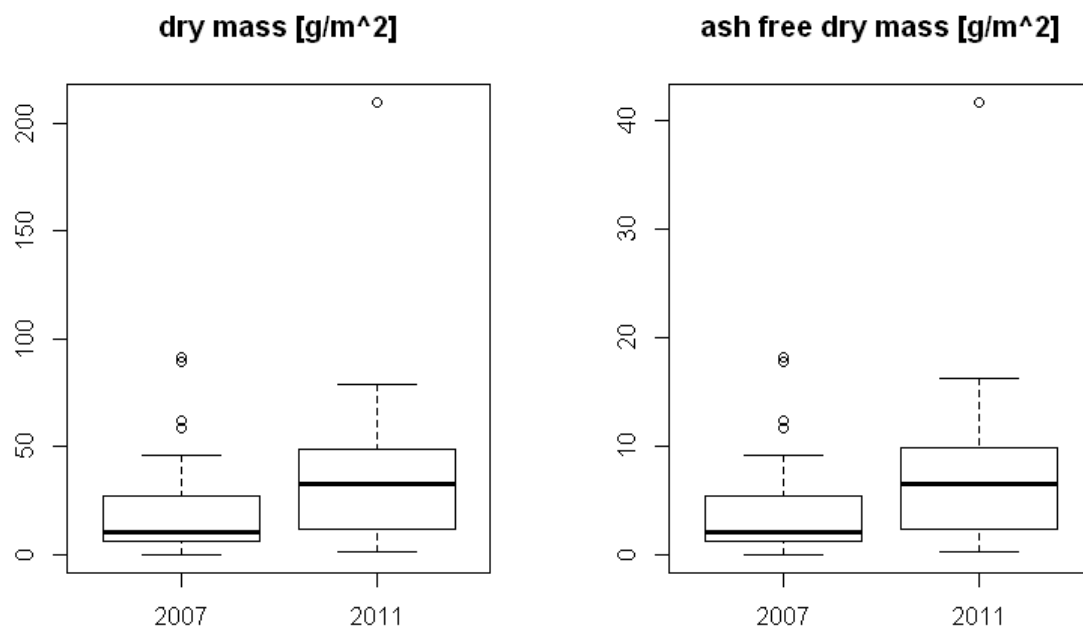
1. Calculate transect surface (Surface=Length*Width)
2. Sum up total biomass (DM and AFDM) for each year
3. Sum up total area surveyed (transect surface) for each year
4. Calculate total biomass/m² for each year (total biomass/area surveyed)
5. Determine the biomass per m² for each transect

Output

- **Table** total biomass per m² for each year

Biomass [g/m ²]	2007	2011
Dry mass	17.5	32.5
AFDM	3.5	6.5

- **Plot** biomass distribution for both years(boxplot)



Asteroids.R

Goal

Test if abundance of asteroids in general and *Odontaster meridionalis* (identified sponges predator) in particular have changed between 2007 and 2011

Input

All asteroids bigger than 5cm and smaller than 5cm seen in the videos were counted during every transect. Whenever possible *Odontaster meridionalis* was identified

Those counts are listed in:

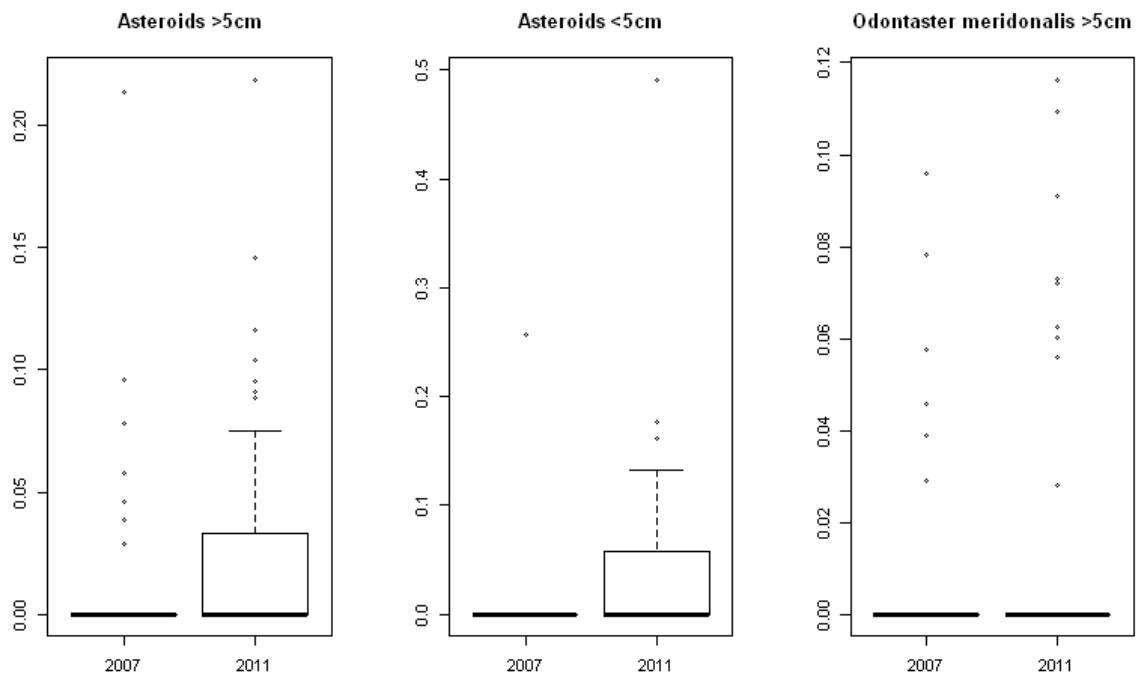
- aseroids.txt: number of asteroids (>5cm, <5cm and *Odontaster meridionalis*)on each transect
- transects.txt (also used in transect_stat.R script)

Steps

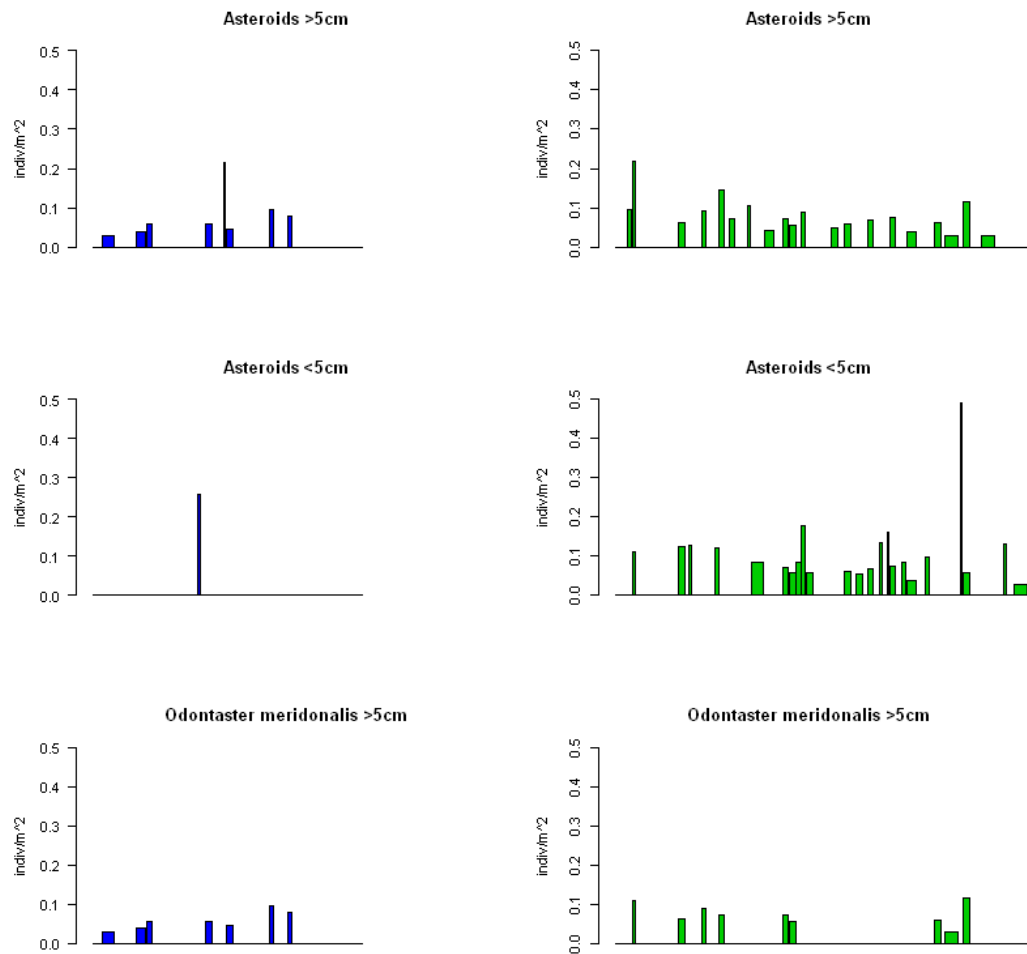
1. Calculate transects surface (Surface=Length*Width)
2. Calculates asteroids abundances for each transect for each year (abundance = number of asteroids on transect/surface of transect)
3. Test if asteroids abundances changed between 2007 and 2011 (Wilcoxon test)
4. Calculate the proportion of small asteroids (possibly juveniles) for each year

Output

- **Plot** abundance distribution during both dives (boxplot)



- **Plot abundance along the transects(barplot)**



- **Statistics: Wilcoxon tests**

	2007 mean	2007 SD	2007 SE	2011 mean	2011 SD	2011 SE	Wilcoxon p
Asteroids >5cm	0.0131	0.0373	0.0054	0.0221	0.0424	0.0050	0.074
Asteroids <5cm	0.0055	0.0375	0.0055	0.0351	0.0726	0.0086	5e-05
O.meridionalis >5cm	0.0086	0.0223	0.0033	0.0094	0.0265	0.0031	0.595

- **Proportion of asteroids <5cm (juveniles?)**

2007: 20% of asteroids <5cm

2011: 54% of asteroids <5cm