

# Colonization of maritime heritage sites:

## Frame grab analysis exercise

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### Frame grab analysis

One of the challenges in marine biology is collecting standardized data. For example, both sessile and mobile organisms live on shipwrecks, so we must come up with ways to quantify their relative abundances. My solution for analyzing shipwreck communities is to convert ROV video to frame grabs and count the animals within each frame grab. You could count individuals, but this is difficult for some animals like sponges, which grow flat to the surface and can cover large areas. There is an additional difficulty in that not every frame grab is the same size. The ROV is carried around by the current underwater, so its distance from the wreck varies. Some frame grabs show large areas of the wreck while other show small areas, depending on how far away the ROV is. I get around this by estimating the percent cover of organisms in each frame grab. I overlay the images with randomly-placed points and count the number of points that “hit” each organism in the image.

### Steps for frame grab analysis:

1. Examine Frame Grab 1. Decide how much of the image you can reasonably analyze, excluding any areas that are too dark, out of focus, or off the wreck. It may be helpful to print the frame grab or project it on a white board and draw a border around the area you want to analyze.
2. Count the number of red points within the analyzable area. This is your total number of points.
3. Identify the most common species and count the number of red points that “hit” this species in the image.
4. Repeat step 3 for each species in the image, even species that you cannot identify.
5. For each species, divide the number of points “hitting” that species by the total number of points. This is your estimated percent cover.
6. Repeat for Frame Grabs 2 – 7.

## Checking the quality of the data

The first step in any data analysis is to determine if your sampling is sufficient. For biological datasets, we use what are called “species-accumulation curves” to tell if we have analyzed enough samples to find all species present in the community. Begin building your species-accumulation curve by counting the number of species you found in Frame Grab 1. Then count any new species you found in Frame Grab 2 for a *total* number of species observed. Repeat for each frame grab until you fill the following table:

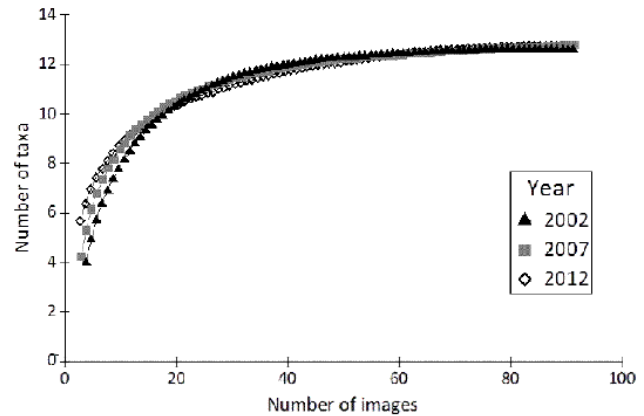
| Frame grab analyzed | Total number of species observed |
|---------------------|----------------------------------|
| Frame Grab 1        |                                  |
| Frame Grab 2        |                                  |
| Frame Grab 3        |                                  |
| Frame Grab 4        |                                  |
| Frame Grab 5        |                                  |
| Frame Grab 6        |                                  |
| Frame Grab 7        |                                  |

Now graph your species-accumulation data as a line plot with “Frame grabs analyzed” on the X axis and “Total number of species observed” on the Y axis. Does the line flatten out and reach an asymptote? In other words, were any new species observed in Frame Grabs 6 and 7, or had you already found all species in the community by then? If you are still finding new species in your last few samples (Frame Grabs 6 and 7), then your number of samples was insufficient to characterize the community, and additional sampling is required.

A more robust way to conduct this analysis is to put the frame grabs in random order, count the number of new species in each, repeat this procedure multiple times, and then calculate the average number of new species for each number of frame grabs analyzed. Your table will be:

| No. frame grabs | No. species | No. species | No. species | No. species | Average |
|-----------------|-------------|-------------|-------------|-------------|---------|
| 1               |             |             |             |             |         |
| 2               |             |             |             |             |         |
| 3               |             |             |             |             |         |
| 4               |             |             |             |             |         |
| 5               |             |             |             |             |         |
| 6               |             |             |             |             |         |
| 7               |             |             |             |             |         |

You can graph the *average* number of new species found for each number of frame grabs analyzed. The curve for this analysis should be smoother. If it reaches an asymptote (becomes flat), then you know that 7 frame grabs are sufficient to find all the species in the community. If, however, the curve does not become flat, then it means more species are out there waiting to be found, and new samples would reveal more species. You should analyze more samples to completely characterize the community.



Species-accumulation curves that reach an asymptote (notice flat portion of curves for > 30 images). After Meyer et al. (2013).

## Interpreting the data

In order to truly understand the biological community, it is important to compare samples from different parts of the wreck. The frame grabs come from the following areas:

- Frame Grab 1: Ship's bow, starboard side, near the top
- Frame Grab 2: Debris on the seafloor next to the wreck
- Frame Grab 3: Ship's hull, starboard side, near the top
- Frame Grab 4: Top deck of the ship, near the bow
- Frame Grab 5: Top deck of the ship, middle of the ship
- Frame Grab 6: Top deck of the ship, middle of the ship
- Frame Grab 7: Top deck of the ship, middle of the ship

**What do you notice about the biological community?** Are the organisms evenly distributed? Are some areas more densely populated than others? Are there differences between the top of the ship and areas closer to the seafloor?

**Why do you think the animals live on different parts of the wreck?** Are there advantages to living in some places over others?

## References cited

1. Meyer, K. S., Bergmann, M. & Soltwedel, T. Interannual variation in the epibenthic megafauna at the shallowest station of the HAUSGARTEN observatory (79 N, 6 E). *Biogeosciences* **10**, 3479–3492 (2013).

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## Frame grab analysis

I see the following species in the frame grabs:

Frame Grab 1: Plumose anemones along the edge, one white vase sponge in the middle

Frame Grab 2: Finger sponge, yellow encrusting sponge

Frame Grab 3: All plumose anemones (they can pull their tentacles in, so the brown, pink, and white lumps are just anemones with their tentacles retracted)

Frame Grab 4: Plumose anemones, yellow encrusting sponge, muddy ascidian

Frame Grab 5: One finger sponge

Frame Grab 6: Finger sponge, plumose anemone, yellow encrusting sponge, stalked ascidian

Frame Grab 7: Finger sponge, yellow encrusting sponge, stalked ascidian

## Checking the quality of the data

An additional check you could do for this section is to have two different individuals or groups analyze the same frame grab and compare their results. If their percent cover estimates are 1 – 5 points different, that's normal/acceptable variation, but if the numbers are very different (> 10 percentage points), it's important to talk about what the groups did differently. Scientists actually run into this problem a lot, and there are studies showing that different people analyzing the same set of images will get different results. It's important to standardize analysis methods as much as possible.

## Interpreting the data

There are multiple correct answers for this exercise. Some of them include:

- The organisms have patchy distributions (dense populations some places, zero individuals other places)
- The top of the ship has lower population density than the side of the ship. This is because upward-facing (horizontal) surfaces tend to accumulate sediment, which can be difficult for organisms to handle.
- Areas closer to the seafloor tend to have lower population density because the current is slower closer to the sediment. Most sessile invertebrates are suspension feeders, meaning they filter particles out of the water for food, so they want to be exposed to a fast current.
- The anemones tend to live along the edges. I think this is because they want to be exposed to the current for suspension feeding.