High potential imaging technology for analysis of *in situ* patchiness

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Abstract:

Modern ecological theory is largely based off mean field assumptions that can overlook the critically important "patchiness", areas of high and low resource distributions, that define ecosystems. Also, studying the fine scale distributions of microorganisms in these patches has traditionally involved very expensive, resource intensive methods. This has greatly hindered the ability to conduct research in this area. This paper present an alternative, high tech, low cost method that can be used to study these patch dynamics across a range of scales, from that of 10's of centimeters to that of kilometers. This method involves the use of a waterproof enclosure that houses a microcomputer and imaging capture device that, with accompanying software, can analyze large batches of video for the presence, or lack thereof, of organisms of a desired size class. This paper also suggests a method, namely fractal analysis, which can be used to compare the self-similarity, or lack thereof, of various organism distributions over a range of scales. Preliminary results seem to indicate a rough correlation between organisms of a small size and those of a larger size, though further data and analysis is necessary for confirmation. Further fine-tuning and utilization of this imagine system promises a high potential yet low cost way in which researchers in both terrestrial and marine landscapes can simultaneously analyze a greater volume of data while reducing their workload in terms of manual analysis.

Introduction:

The way an organisms interacts with its environment is critical to its ability to survive and thrive. This is true from the smallest of bacteria to the largest of mammals, and yet many theoretical ecological models do not accurately portray this reality. Most theoretical models of consumer-resource dynamics are based on a 'mean field' approach in which the average distributions of consumers and resources are taken over a large area (Grunbaum 2012). A mean distribution assumes that a predator has an equal chance of encountering a desired resource at anywhere in it's environment; this simply not the case in nature. In the natural world organisms tend to exist in "patches" rather than in equal distributions. This "patchiness" can be defined as "the spatial heterogeneity typical of organism distributions" and "is the rule, rather than the exception, in most terrestrial and marine ecosystems" (Greene et al. 1994).

This paper deals with the development of an affordable, in situ method that can be used to compare the patchy distribution of smaller planktonic organisms to those of their larger predators. These distributions will be studied from the microscale (cm's) to the mesoscales (100 of m's), using a video imaging device. Unlike plankton tows, which destroy fine scale distribution by concentrating organisms over some distance, the use of an imager will allow for high resolution *in situ* observations of patches across scales. Also unlike other *in situ methods*, such as acoustic backscatter arrays or particles counters, which are very expensive, this imager will offer an effective, low cost approach to *in situ* analysis. This analysis over multiple scales will allow for the comparison of the patchy distribution of small

planktonic organisms, whose patch formations are almost exclusively determined by physical factors, to those of larger planktonic predators, whose morphology potentially allows swimming to play a much larger role in their patch formation, especially at smaller scales. If a consumers swimming ability is able to overcome oceanographic forces at smaller scales then these consumers distributions should closely match those of their phytoplankton prey, whereas at larger scales, where physical oceanographic forces dominate, the distributions should show less correlation.

Patchiness has been shown to play a significant role in the survivability of larvae (Lasker 1975) and "how effectively predators navigate within heterogeneous prey environments dictates whether they experience significantly higher (within patches) or lower (between patches) prey concentrations"(Menden-Deur 2004). Furthermore, it has also been shown that the spatial configuration of prey has crucial consequences for the dynamics of nutrient and energy fluxes in ecosystems (Wiens et al. 1985; Schneider et al. 1987). Studies such as these have highlighted the importance of patches in the ecological landscape and have inspired mathematical models that oppose the standard mean distribution methods.

There have been a number of ways in which researchers have attempted to model the consequences and formation of these patchy distributions, and this paper develops the materials and data collection methods that will be necessary in order to successfully apply fractal analysis and patch dynamics to the study of fine scale organism distributions. Multifractal analysis, which looks at the self-similarity of patch distribution between organisms and scales, has been successfully used to

model planktonic patchiness across various spatial scales (Pascual et al. 1995, Seuront et al. 1996). This study seeks to test whether the use of multifractals can be expanded to not only analyze plankton distributions, but to also model the distribution of small planktonic resource organisms in relation to their larger consumers. Fractal analysis can also allow us to see when there is a "break" in the fractal dimension between scales. This break shows a difference in organism correlation between scales and can signify the scales at which different forcing factors are dominant. This unity of theory with practice is an especially critical task given the current academic landscape in which there is a break between empirical and theoretical ecologists. This is troubling because both aspects are vital to ecological study; theory cannot be tested without evidence, and evidence cannot be strengthened without theory (Codling et al. 2012).

The patchy make up of the marine environment and how successfully organisms can exploit these patches can have huge impact on the ecological makeup of the ocean. Organisms such as zooplankton, whom are of critical importance to the marine ecosystem, are no exception to this. A better understanding of the way in which these creatures interact and form patches will lay the foundation for greater comprehension as to how the ocean landscape is constituted, as well as to how future changes will affect these all important patches.

Materials and Methods:

To assess the horizontal distribution of plankton at the centimeter to kilometer scale an improvised submersible imaging system will be utilized. The base of this imaging system consists of a BeagleBoard microcomputer (shown in Figure 1) with a Linux operating system that has a Leopard Imaging camera attached to it. This computer was then mounted into a waterproof Otterbox enclosure into which a Lexan lens has been mounted and a waterproof Ethernet cable has been connected to. This Ethernet cable allows the unit to transmit video to an above the surface computer. It also allows the imager to receive power from above surface batteries through the utilization of power-over-Ethernet connectors. The unit was connected to a laptop computer via an Ethernet cable, and the computer utilized the Linux program FOSICA to capture video. This unit was then attached to a larger mount that consisted of a refrigerator grate, L strut, 30 lbs of lead weights, and an aluminum fin as depicted in Figure 2 below. Under water LED light enclosures constructed of PVC sea table ends and Plexiglas were then attached to the refrigerator grate below the imager for illumination as shown in Figure 3. The light fixture used most often, due to it's mixture of battery life longevity and small size to minimize perturbation of flow was powered by the homemade battery pack created through the soldering together of 8AAA batteries in series, and is shown in Figure 4. This larger system was used to capture video by either towing it behind a boat or by tying it off the dock at Friday Harbor Labs, and the system was used to capture video at depths ranging from 5m-10m.

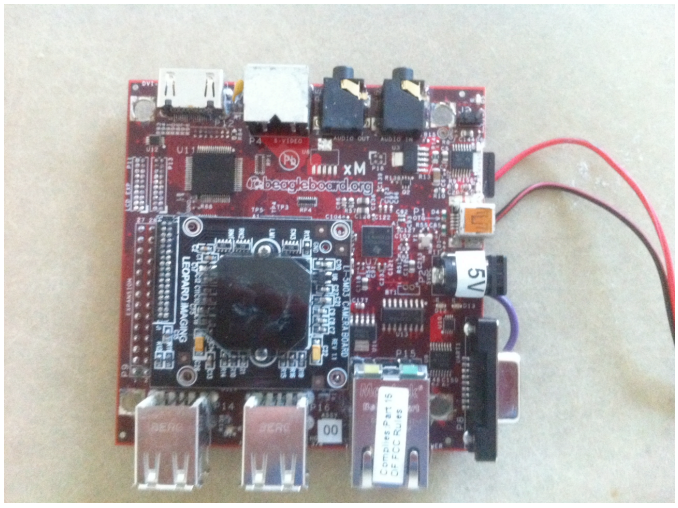

Figure 1: BeagleBoard microcomputer with camera

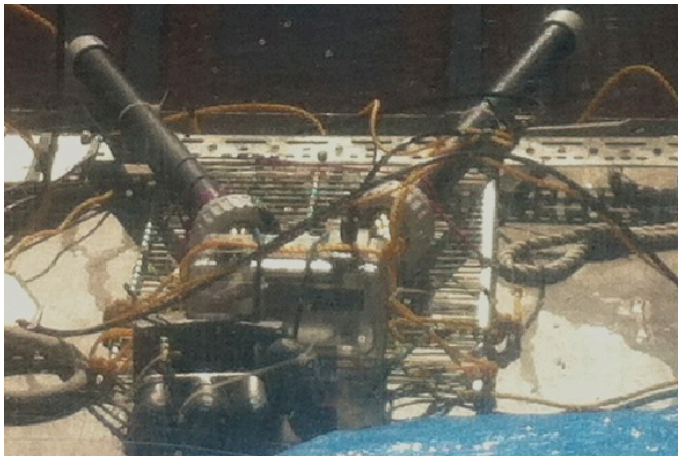

Figure 2: Underwater Imaging Structure (lighting, Otterbox enclosure, L-Strut, refridgerator grate, and lead weight)

Figure 3: PVC light enclosures. Unit utilized most frequently is second from the left.

Figure 4: Power supply for most utilized light enclosure composed of 8AAA batteries soldered in series.

The imager captured video at a rate of 50fps, with an image volume of approximately 9 cubic centimeters. This allowed for particles comparisons at a fine scale of tens of centimeters under most flow conditions. It also allowed for comparisons across any scale larger than this, with the only being limitation being the capacity of the hard drive to store large amounts of video. The captured video was then analyzed by the program FOSICA, which allows for differentiation between low biomass, non-patch areas, and high biomass patches. This is accomplished by the utilization of a variety of filters. In the order in which they are applied the video is first analyzed using a contrast filter, which creates greater contrast between a narrow range of pixel values, allowing the user to distinguish particles from background noise. The video is then subjected to a convolution filter, which depending on the size of the filter can normalize pixels in a given area. This is both useful in eliminating interference from the light source, and in blurring out small particles to look for large particles of interest. This is followed by a threshold filter, which creates a binary file in which pixels that fall between a certain range are considered "in range" and turn white, whereas those that do not are considered "out-of-range" and turn black. Since organisms tend to refract light, they are brighter than most background particles and this threshold filters can be used to differentiate them from the background noise. The final step is the utilization of a "Find Particles" filter that detects particles based on certain pixel area thresholds. Varying these area thresholds can allow for discrimination between small and large particles. Examples of raw video versus that seen after filtering can be seen in Figures 6 & 7. Processing the video in this way results in a .txt file, which can then

be used to create graphs that show particle counts either second by second, or averaged over an entire video clip.

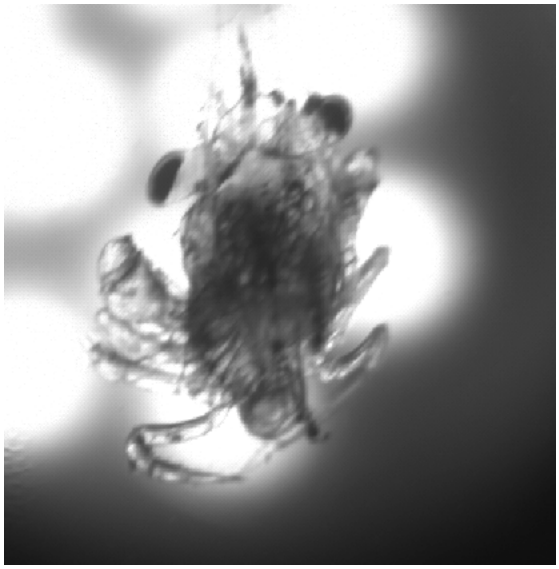

Figure 5: Crab megalope, demonstrating possible image resolution *in situ*

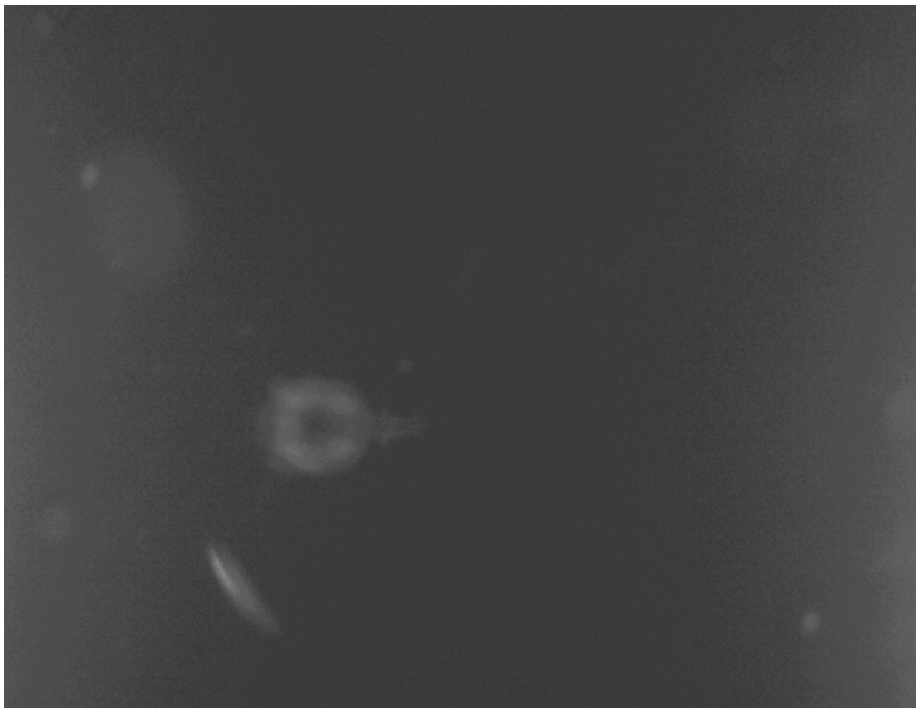

Figure 6: Example of raw video captured in low light conditions. Note multitude of various particles and pixel brightness's.

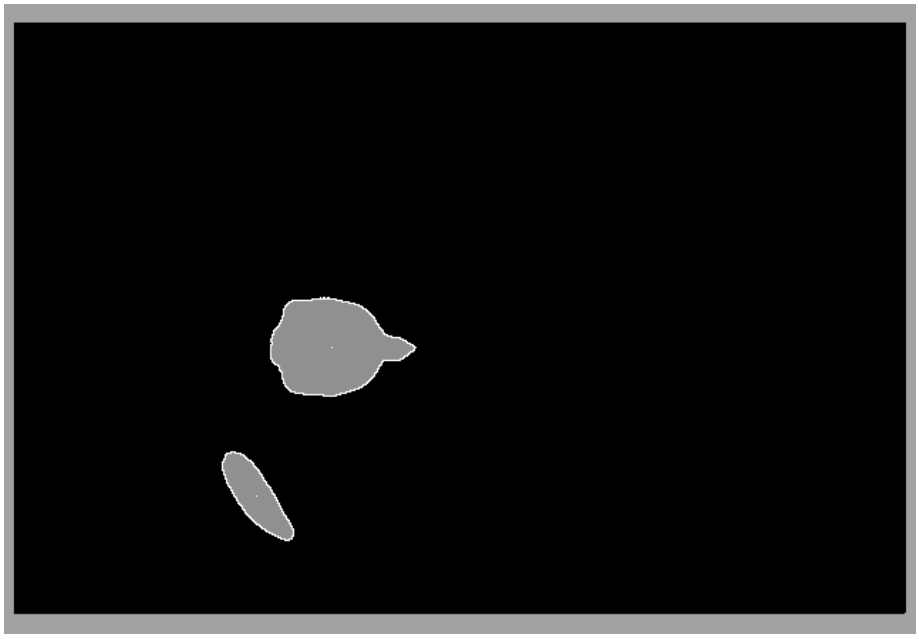

Figure 7: After processing above frame through various filters specifying particle brightness and size, a count of only large organisms can be obtained.

Data & Analysis:

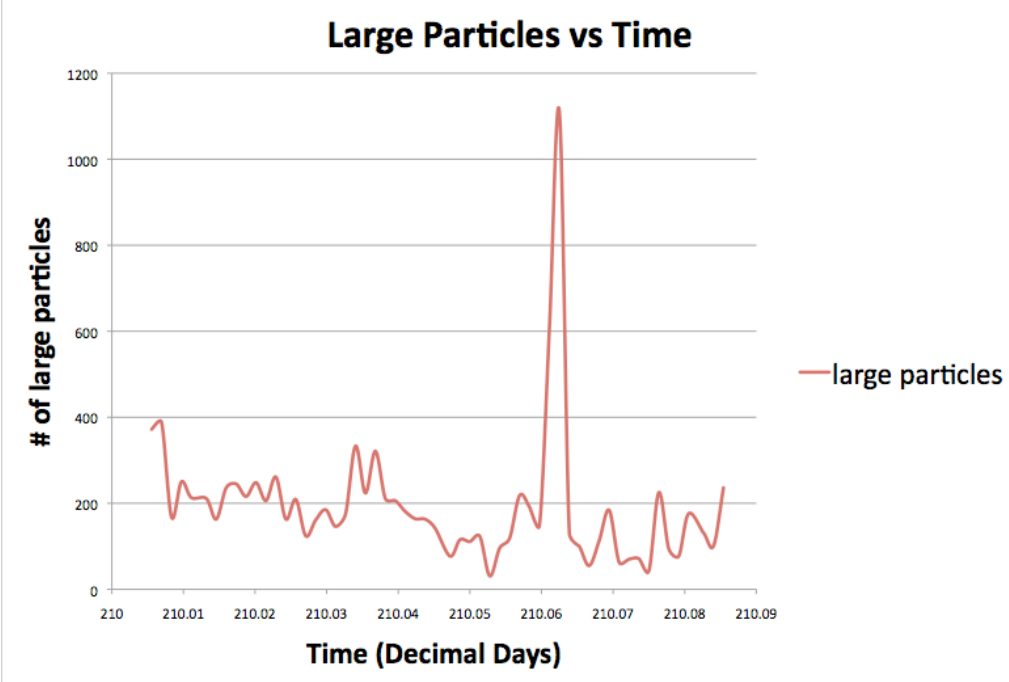

Figure 3: Smooth lined scatter plot in which each point depicts the total # of large particles in each 30 sec video at a given time.

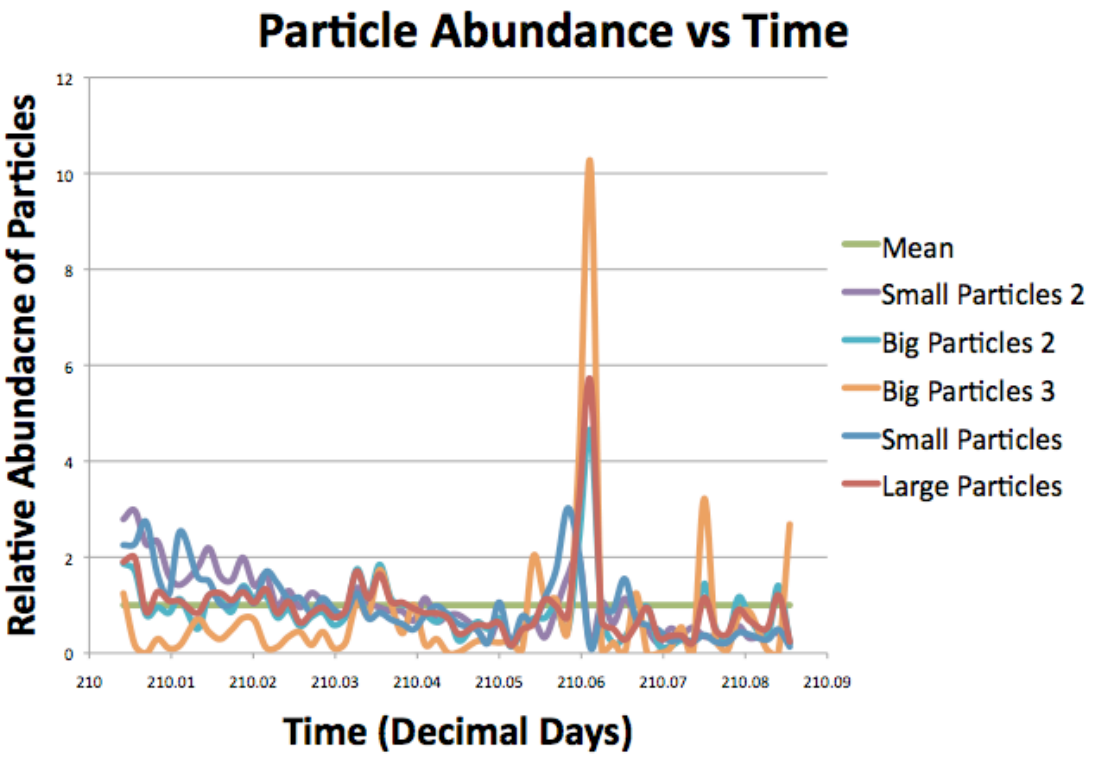

Figure 4: Similar to figure 3 above, however this graph includes various particles size ranges (by using different settings in the "Find Particles" filter) and relative rather than absolute particle counts. All absolute counts were divided by their respective means and therefore peaks and troughs represent deviations from mean distributions in each count. As shown, there seems to be some similarity between peeks and troughs of particles of varying sizes.

Discussion:

Preliminary results seem promising as they appear show some correlation between particles of various sizes, however further video analysis and fractal analysis will be required. Video analysis also requires a large amount of computing power, however, given the limitations of the laptop used only a fraction of the video obtained could be analyzed. Upon further analysis of larger sets of video the analysis settings could be further fine-tuned and fractal self-similarities between groups could be quantitatively ascertained.

An equally interesting result in this research however was the viability of the materials and methods used to obtain data. For a low cost, of only a few hundred dollars, we were able to create a submersible imaging system with the ability to analyze hours of video in order to ascertain the counts of various organisms of different size classes. With further time and fine-tuning the specificity of analysis could be greatly increased and further information about organism patchiness could be elucidated. This low cost, but high tech imaging system could open doors to affordable broad scale studies with multiple data collection points, greater ecological research in developing countries, and reduced workloads for both terrestrial and marine researchers through automated video analysis.

References:

Lasker, R. 1975. Field criteria for survival of anchovy larvae: the relation between inshore chlorophyll maximum layers and successful first feeding. *Fishery Bulletin,* 73 :453-462

Raechel Waters, and James Mitchell, "Centimetre-scale spatial structure of estuarine in vivo fluorescence profiles," *Marine Ecology Progress Series*, 237 (2002): 51-63,

Edward Codling, and Alex Dumbrell, "Mathematical and theoretical ecology: linking models with ecological processes," *Interface Focus*, no. 2 (2012): 144-49,

Pascual, Mercedes, Fortunato Ascioti, and Hal Caswell. "Intermittency in the plankton: a multifractal analysis of zooplankton biomass variability." *Journal of Plankton Research. 17. no.6*(1995): 1209-1232.

Greene, C.H., P.H. Wiebe, and J.E. Zamon 1994. Acoustic visualization of patch dynamics in oceanic ecosystems. *Oceanography* 7:4-12.

Grunbaum, Daniel. "The logic of ecological patchiness." *Interface Focus.* doi: 10.1098/rsfs.2011.0084. (2012).

Seuront, Laurent, Francois Schmitt, Yvan Lagadeuc, Daniel Schertzer, Shaun Lovejoy, and Serge Frontier. "Multifractalanalysis of phytoplankton biomass and temperature in the ocean." *Geophysical Research Letters.* 23. no. 24 (1996): 3591-3594.

Menden-Deur, Susanne. (2004). *Linking individual foraging strategies with ecological dynamics: Quantifying zooplankton movements in heterogeneous resource distributions.* Retrieved from ProQuest Dissertations and Theses. (Accession Order No. [3131200])

Ortner, P.B., Hill, L.C., Edgerton, H.E., 1981. In-situ silhouette photography of Gulf Stream zooplankton*. Deep-Sea Research* 28A, 1569–1576.