CANON update – March 2012

Robotic Sampling Method Comparison for Zooplankton

Platform overview:

ESP
sample volume (standard): 1 L
sample volume (high volume): 4 L
sampling duration: ~ 30–60 minutes
sampling style: "sipping"
sample number: several 10s per deployment
sample processing: near-real time, onboard

AUV Dorado
sample volume: ~ 1.8 L
sampling duration: < 2 seconds
sampling style: "gulping"
sample number: 10 per deployment
sample processing: shore-side benchtop sandwich hybridization assay (SHA)

Sampling Considerations

Point Sampling — sampling a discrete volume of water in an instantaneous manner (e.g. "gulping") is preferable to "sipping" in that it does not integrate zooplankton targets (nor environmental data) collected over a period of time. For this reason, point sampling provides the highest resolution (in both space and time) sampling possible.

Sample Volume — generally, more is better. However, observations from 2008-2011 for zooplankton sampling with Dorado indicate that 1.5 L of water is sufficient for assessing zooplankton variation with the benchtop SHA. This volume does not include water necessary for other assays (e.g., nutrients, chl, pigments, microscopy — Chavez lab and others). The latter need typically equals 100–200 mL per sample. Ideal sample volume is therefore 2–4 L.

Gulper Mechanism — despite their substantial benefits, gulpers are energetically costly to prime for repetitive sampling during a single mission. It is also possible cross contamination among consecutive samples would occur because gulpers can difficult to fully drain — tens of mLs residual seawater per sample.

A Theoretical Flow-Through System — a chamber (2–4 L) that allowed constant flow-through of ambient seawater inward at forward vents and outward through aft exhaust vents. Both sets of vents could be instantaneously shuttered to collect each sample. Each sample could then be "sipped" from this chamber, onto a filter of choice, by an ESP-like sampling mechanism. With a 4 L chamber volume, any volume up to 4 L could be

filtered and the remainder, if any, washed out upon re-opening of both forward and aft chamber vents. Continual flow-through would also 'clean' the chamber for the next sample.

Sample Preservation — RNAlater has been successfully tested with both mussel larvae and copepods for on-filter RNA preservation.

2008–2010 ESP and AUV Dorado Results:

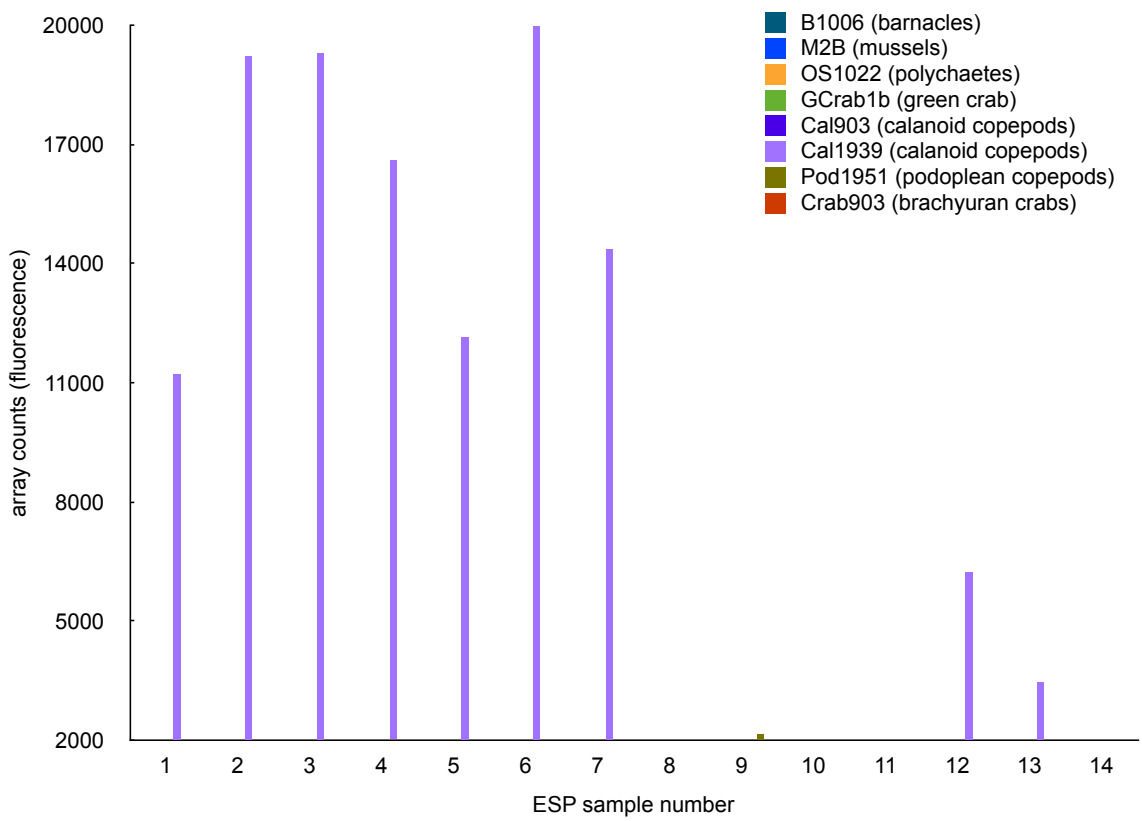
- At standard sampling volume of 1 L (2008 and prior deployments, data not shown) the ESP was only occasionally successful in detecting invertebrate larvae targets with taxonomically specific probes, but was consistent in detecting very broad taxonomic groups (e.g., eukaryotes, crustaceans).
- ESP high volume sampling (4 L) and new probes of moderate taxonomic specificity (e.g., calanoid copepods), resulted in far more consistent detection of zooplankton targets (Figure 1A, 2A).
- Benchtop SHA processing of AUV Dorado water samples (1.5 L), in contrast, has consistently detected zooplankton targets matching probes of both moderate (copepods) and fine (invertebrate larvae) taxonomic specificity (Figure 2A, 2B).
- The bottom line: the combination of "gulping" water samples with benchtop SHA consistently detects the relative abundance of multiple zooplankton targets while "sipping" followed by microarray SHA onboard the ESP has consistently detected fewer targets.
- We have yet to necessarily determine the relative contributions of mechanical sampling, platform behavior (mobile vs. stationary), and SHA chemistry styles (plate vs. microarray), to the observed zooplankton sampling differences between these two platforms.

Figure 1. Zooplankton sampling results in 2009, SHA probe signals are differentiated by color. A) Three ESP deployments April–October. Y-axis cut-off of 2000 microarray fluorescence counts is arbitrary — a few target signals above background did occur with values between 0–2000 counts but are not considered sufficiently strong to convey reliable presence-absence or relative abundance data. B) AUV Dorado sample results from one mission in April. Benchtop SHA optical density (OD), at wavelength = 450 nm, is indicated on y-axis. [Harvey et al. (2012) Journal of Experimental Marine Biology and Ecology 413 (2012) 60–70]

Figure 2. Zooplankton sampling results during BloomEX/BIOSPACE 2010, SHA probe signals are differentiated by color. A) Two ESP deployments 1–2 of November. B) AUV Dorado sample results from missions spanning 27–28 of October (closest in date to the above ESP samples). [unpublished data]

Figure 1

A


ESP deployments: April - October 2009
B1006 (barnacles)
M2B (mussels)
OS1022 (polychaetes)
GCrab1b (green crab)
Cal903 (calanoid copepods)
Cal1939 (calanoid copepods)
Pod1951 (podoplean copepods)
Crab903 (brachyuran crabs)
array counts (fluorescence)
20000
17000
14000
11000
8000
5000
2000
1
2
3
4
5
6
7
8
9
10
11
12
13
14
ESP sample number

B

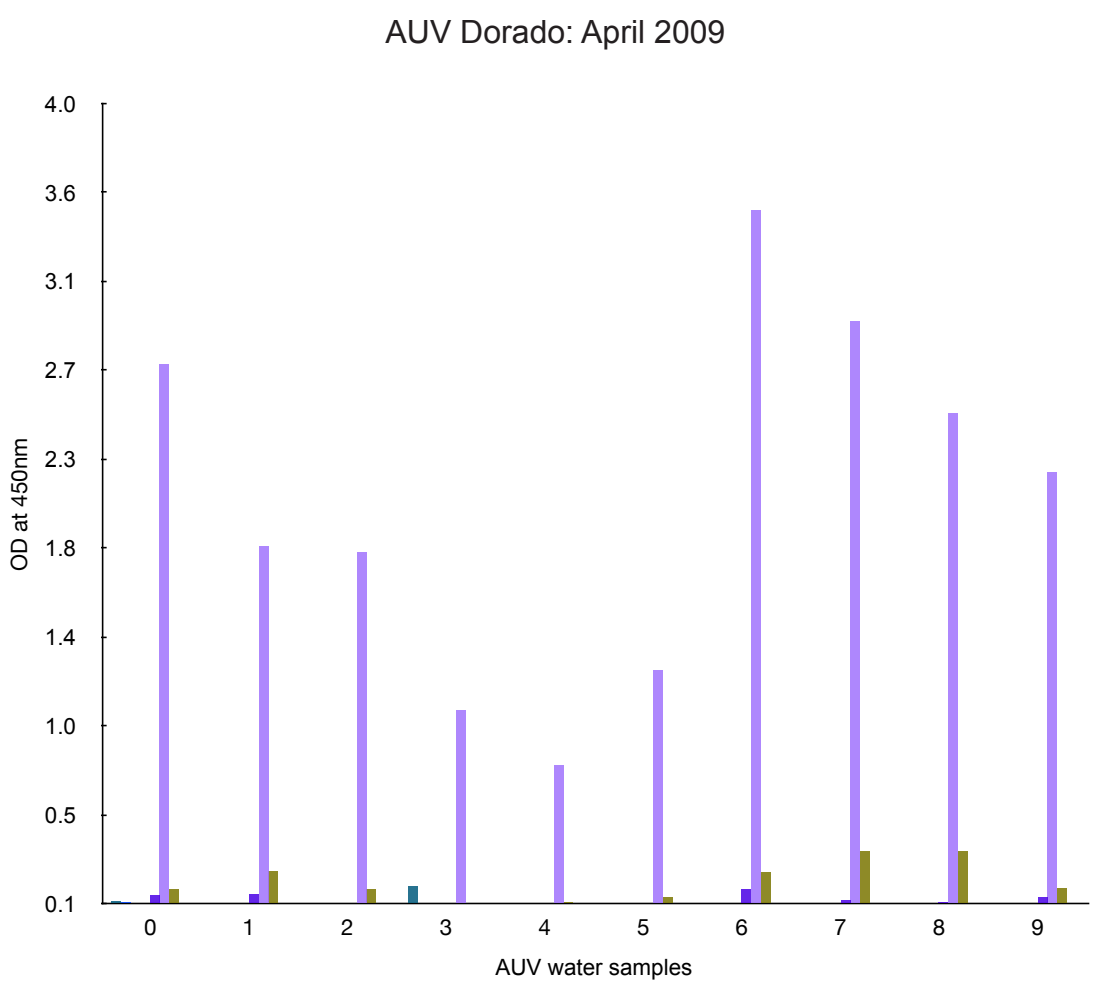
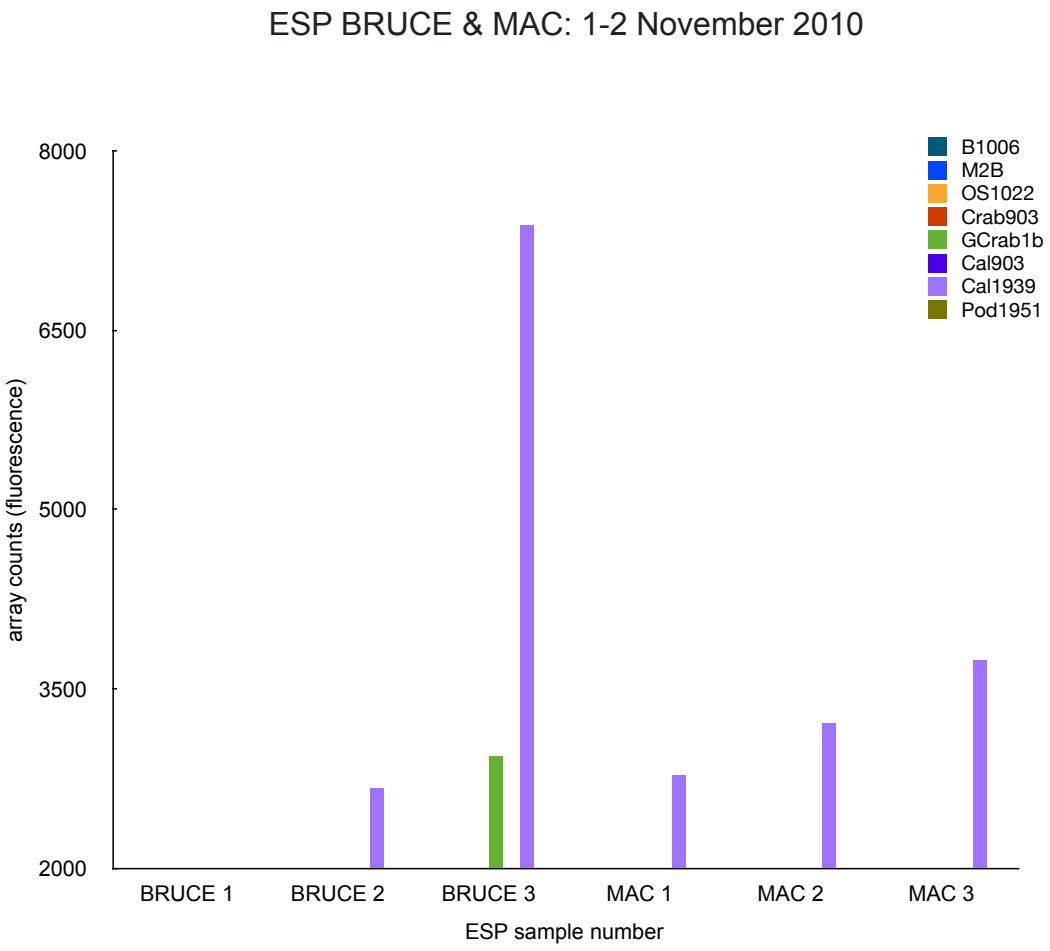
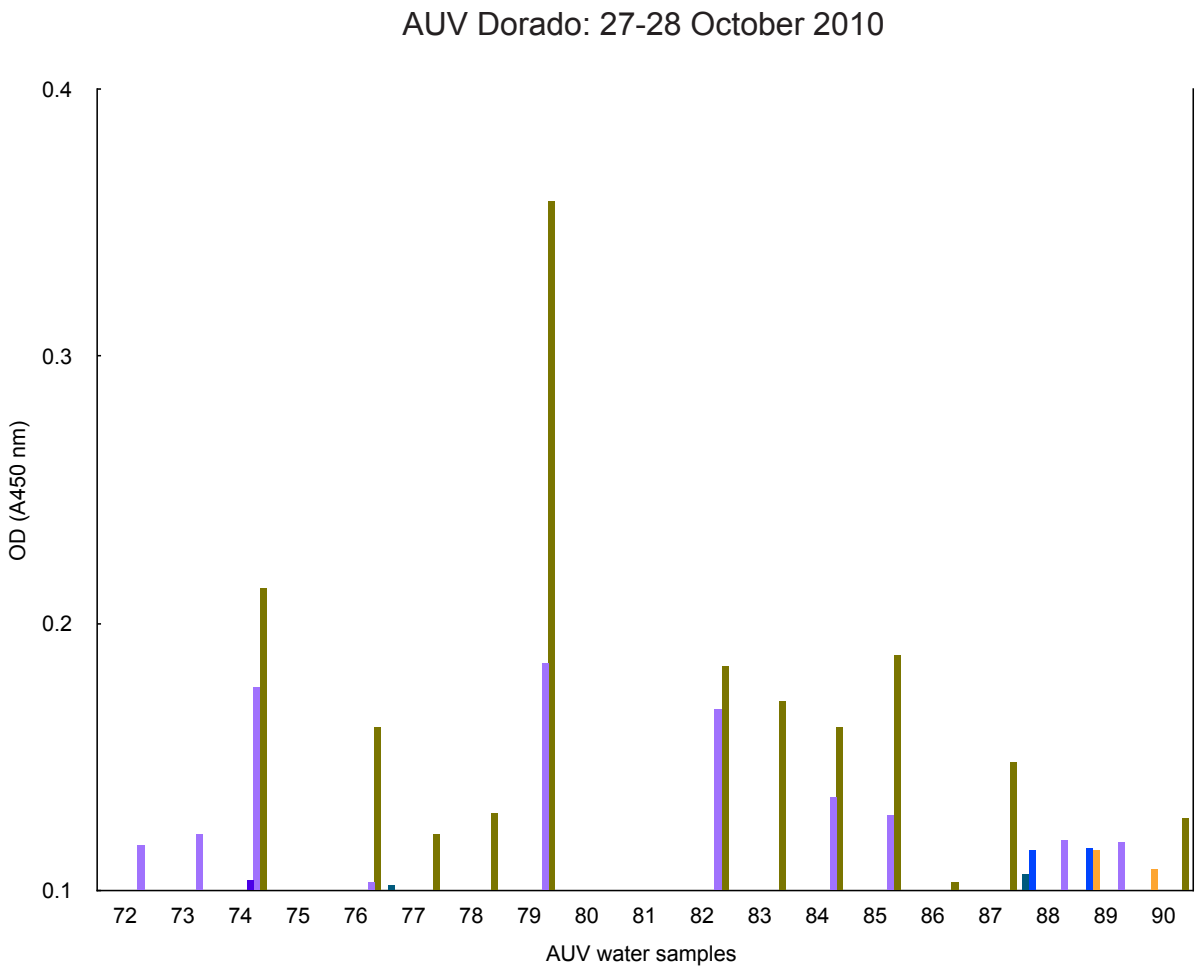

AUV Dorado: April 2009
OD at 450nm
4.0
3.6
3.1
2.7
2.3
1.8
1.4
1.0
0.5
0.1
0
1
2
3
4
5
6
7
8
9
AUV water samples

Figure 2

A


ESP BRUCE & MAC: 1-2 November 2010
B1006
M2B
OS1022
Crab903
GCrab1b
Cal903
Cal1939
Pod1951
8000
6500
5000
3500
2000
array counts (fluorescence)
BRUCE 1
BRUCE 2
BRUCE 3
MAC 1
MAC 2
MAC 3
ESP sample number

B


AUV Dorado: 27-28 October 2010
0.4
0.3
0.2
0.1
OD (A450 nm)
72
73
74
75
76
77
78
79
80
81
82
83
84
85
86
87
88
89
90
AUV water samples