VOLUME 2: COST PROPOSAL

ONR BAA ANNOUNCEMENT # ONR-BAA-07-040

Title: Applications of the Environmental Sample Processor (ESP) to Harmful Algal Bloom Management: Field Deployment, Technology Transfer, and Commercial Evaluation

Prime Offeror:

Monterey Bay Aquarium Research Institute
Moss Landing, California

Subrecipients:

Woods Hole Oceanographic Institution
Woods Hole, Massachusetts

NOAA National Ocean Service
Charleston, South Carolina

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Duration of effort: 1 September 2008 – 31 August 2011

Funds Requested: Y1- $503,921 Y2 - $510,266 Y3 - $513,216 Total: $1,527,404

Employer ID: 77-0150850
Type of Applicant: Research Institute, Private, Non Profit 501(c)(3) Organization
Submitted to CFDA Number: 12.300

TABLE OF CONTENTS

NON – UNOLS SHIP USE

For year-2, limited IOOS funds will support only a single deployment and recovery in the coastal ocean at Buoy B (Vol.1, Fig. 6). To revisit the Buoy B site, we request 6 days of ship time on the R/V Tioga (WHOI) during year-2 to cover two trips to Buoy B for instrument deployment and ground-truth sampling near the mooring. Costs for that total $14.K, 6 days at $2,400 per day.

The R/V Tioga is a very capable coastal research vessel (60ft) with required lift capabilities from the A-frame, dedicated CTD/rosette, current profilers, and an underway hydrographic data collection system (see http://www.whoi.edu/page.do?pid=8159).

TABLE OF PARTNERSHIPS- MONTEREY BAY AQUARIUM RESEARCH INSTITUTE

Principal Investigator	Institution	FY 08 funds Requested	FY 09 funds Requested	FY10 funds Requested	TOTAL
C. Scholin Lead PI	MBARI (Non-profit)	$475,797	$287,107	$248,761	$1,011,666
D. Anderson B. Keafer D. McGillicuddy Co-Pis	WHOI (Non-Profit)	$28,124	$223,159	$264,455	$515,738
G. Doucette Co-PI	NOAA/NOS (Government)	$2,000	$49,380	$49,899	$101,279
C. Melancon Industry Rep	Applied Biosystems Inc. **(Business)**	$0	$0	$0	$0
C. Mathewson Industry Rep	McLean Research Laboratories Inc. **(Business)**	$0	$0	$0	$0

Budget

MBARI - SUMMARY BUDGET	Yr 1		Yr 2		Yr 3		Yr 1-3	
SALARIES/WAGES	# Months	2008 -2009	# Months	2009 - 2010	# Months	2010 - 2011	# Months	Total
Scholin - Principal Investigator	1.00		1.00		1.00		3.00	
Massion - Sr. Mechanical Engineer	1.00		1.00		0.50		2.50	
Jensen - Electrical Engineer	1.00		1.00		1.00		3.00	
Roman - Software Engineer	1.00		1.00		1.00		3.00	
Everlove - Mechatronics Engineer	3.00		6.00		4.00		13.00	
Schramm - Software Engineer	1.50		1.00		1.00		3.50	
Machinist	2.00						2.00	
Total Reg Salary		**91,638**		**99,044**		**82,446**	**30.00**	**417,886**
TOTAL SALARY & WAGES		91,638		99,044		82,446	30.00	417,886
BENEFITS (Rate-Reg) 53.00%		48,568		52,493		43,696		144,758
TOTAL BENEFITS		48,568		52,493		43,696		144,758
TOTAL SAL & BEN		**140,206**		**151,537**		**126,142**	**0.00**	**562,644**
EQUIPMENT Includes CIP Equip items >$5K								
1 Pressure housing		13,250						13,250
2 CTD/fluorometer/OBS/O		10,000						10,000
Subtotal Equipment		**23,250**						**23,250**
CIP EQUIPMENT - Fabrication								
Outside Service 2x 40K for mfg parts for ESP fab		80,000						80,000
COTS parts 2x 40K for ESP fab		80,000						80,000
COTS and mfg parts for ESP mooring		40,000						40,000
Subtotal CIP Equipment		**200,000**						**200,000**
SUBAWARD								
Subaward: WHOI		28,124		223,159		264,455		515,738
Subtotal Subaward		**28,124**		**223,159**		**264,455**		**515,738**
SUPPLIES: Instrument Spares		0		5,000		5,000		10,000
Mooring Hdwr/maint/spares				10,000		10,000		20,000
Expendable reagents for ESP probe chemistry		10,000		15,000		15,000		25,000
Subtotal Supplies		**10,000**		**30,000**		**30,000**		**70,000**
TRAVEL Domestic Travel: $300 air, $500 hotel, $200 meals,								
$200 rental car/truck = $1.2K per trip								0
Yr 1 - 2 to scope site, Yr2&3 3 to deps/turnarounds		3,600		3,600		3,600		
Subtotal Travel		**3,600**		**3,600**		**3,600**		**10,800**
SHIPPING/POSTAGE								
Shipping		5,000		5,000		5,000		15,000
Subtotal Shipping		**5,000**		**5,000**		**5,000**		**15,000**
Total Direct Costs		**410,180**		**413,296**		**429,197**		**1,252,674**
Modified Total Direct Costs Base		**183,806**		**190,137**		**164,742**		**538,686**
Total Indirect Costs Rate: 51%		**93,741**		**96,970**		**84,019**		**274,730**
TOTAL DIRECT & INDIRECT COSTS		**503,921**		**510,266**		**513,216**		**1,527,404**

Budget

MBARI - INSTITUTIONAL BUDGET	Yr 1		Yr 2		Yr 3		Yr 1-3	
SALARIES/WAGES	# Months	2008 -2009	# Months	2009 - 2010	# Months	2010 - 2011	# Months	Total
Scholin - Principal Investigator	1.00		1.00		1.00		3.00	
Massion - Sr. Mechanical Engineer	1.00		1.00		0.50		2.50	
Jensen - Electrical Engineer	1.00		1.00		1.00		3.00	
Roman - Software Engineer	1.00		1.00		1.00		3.00	
Everlove - Mechatronics Engineer	3.00		6.00		4.00		13.00	
Schramm - Software Engineer	1.50		1.00		1.00		3.50	
Machinist	2.00						2.00	
Total Reg Salary		**91,638**		**99,044**		**82,446**	**30.00**	**273,128**
TOTAL SALARY & WAGES		**91,638**		**99,044**		**82,446**	**30.00**	**273,128**
BENEFITS (Rate-Reg) 53.00%		48,568		52,493		43,696		144,758
TOTAL BENEFITS		**48,568**		**52,493**		**43,696**		**144,758**
TOTAL SAL & BEN		**140,206**		**151,537**		**126,142**	**0.00**	**417,886**
EQUIPMENT Includes CIP Equip items >$5K								
1 Pressure housing		13,250						13,250
2 CTD/fluorometer/OBS/O		10,000						10,000
Subtotal Equipment		**23,250**						**23,250**
CIP EQUIPMENT - Fabrication								
Outside Service 2x 40K for mfg parts for ESP fab		80,000						80,000
COTS parts 2x 40K for ESP fab		80,000						80,000
COTS and mfg parts for ESP mooring		40,000						40,000
Subtotal CIP Equipment		**200,000**						**200,000**
SUPPLIES: Instrument Spares		0		5,000		5,000		10,000
Mooring Hdwr/maint/spares				10,000		10,000		20,000
Expendable reagents for ESP probe chemistry		10,000		15,000		15,000		25,000
Subtotal Supplies		**10,000**		**30,000**		**30,000**		**70,000**
TRAVEL Domestic Travel: $300 air, $500 hotel, $200 meals, $200 rental car/truck = $1.2K per trip								0
Yr 1 - 2 to scope site, Yr2&3 3 to deps/turnarounds		3,600		3,600		3,600		
Subtotal Travel		**3,600**		**3,600**		**3,600**		**10,800**
SHIPPING/POSTAGE								
Shipping		5,000		5,000		5,000		15,000
Subtotal Shipping		**5,000**		**5,000**		**5,000**		**15,000**
Total Direct Costs		**382,056**		**190,137**		**164,742**		**736,936**
Modified Total Direct Costs Base		**183,806**		**190,137**		**164,742**		**538,686**
Total Indirect Costs Rate: 51%		**93,741**		**96,970**		**84,019**		**274,730**
TOTAL DIRECT & INDIRECT COSTS		**475,797**		**287,107**		**248,761**		**1,011,666**

MBARI Statement of Work

Year 1 September 2008-August 2009

- Procure parts for and fabricate two core Environmental Sample Processors (ESPs); calibrate and conduct instrument acceptance tests at MBARI.
- Convene ESP operations training workshop at MBARI for project participants. This will include instruction on laboratory use, set-up, programming, data management, basic service, preparation for deployment, at-sea deployment/recovery of the ESP.
- Install core ESPs at the NOAA/NOS Marine Biotoxins Laboratory (Doucette) and WHOI (Anderson lab), and provide a dedicated, custom-configured laptop for operating the ESP and processing data. Installation of the ESP requires travel to partner institutions for instrument set-up and preliminary testing to confirm the machines are operating properly.
- Produce custom probe arrays and coordinate production of quality-assured expendable reagents for use at MBARI, NOAA and WHOI (this requires moderate scale production runs, documenting QA/QC protocols, creating a tracking scheme for reagent lots, etc.).
- Coordinate inter-lab calibration experiments using standardized operating procedures to evaluate instrument performance against natural and synthetic samples.
- Fabricate one coastal mooring for ESP surface water operations (includes data telemetry, CTD package, 50m-rated pressure housing, power system, subsurface platform, rode anchor and shipping container). Deliver to WHOI ahead of year-2 field operations.
- Coordinate formation of industry partnerships; initiate opportunities for commercializing the ESP.

Year 2 September 2009-August 2010

- Produce quality-assured custom probe arrays and expendable reagents for use at MBARI, NOAA and WHOI in support of ESP operations. Coordinate inter-lab calibration experiments and logistics in preparation for Salt Pond deployment spring/summer 2010 (see Vol. 1).
- Deploy ESP at Salt Pond, establish data telemetry system. This includes travel to WHOI to assist with pre-deployment instrument configuration and remote operation, and to establish procedures data interpretation and dissemination.
- Participate in field and laboratory studies to validate performance of the ESP as described in Vol. 1.
- Provide support for integrating new toxin assays devised by Doucette's team for use on the ESP (see Vol. 1 or 2, Appendix 1).
- Summarize results for publication in peer-reviewed journals.
- Continue formation of industry partnerships and efforts to commercialize the ESP. Work towards definition of instrumentation products, accessories, expendable reagents, production costs, required customer support, pricing structure, and market assessment.

Year 3 September 2010-August 2011

- Produce and quality-assure custom probe arrays and expendable reagents for use at MBARI, NOAA and WHOI in support of ESP operations. Coordinate inter-lab calibration experiments and logistics in preparation for fielding three ESPs April-June 2011.
- Deploy an array of three ESPs in the Gulf of Maine (see Vol. 1), establish data telemetry, management and dissemination system. This requires travel to WHOI to assist with instrument configuration, deployment/servicing/recovery, and remote operation.
- Participate in cruises and laboratory work to validate performance of the ESP.
- Summarize results for publication in peer-reviewed journals.
- Complete plan for commoditizing the ESP system.

Declarations

MBARI will retain ownership of the ESPs and the mooring for fielding the instrument throughout the duration of this project. By the end of year-1, equipment constructed will be formally transferred to NOAA/NOS Marine Biotoxins Laboratory and WHOI and remain in receiving labs' custody for the duration of this project. During year-3, both ESPs produced under the terms of this proposal will be used for field operations in the Gulf of Maine. At the end of this project, all custom made hardware will be returned to MBARI in lieu of any future agreement to the contrary.

In the event the instrument malfunctions or is damaged, MBARI will service, repair or replace the equipment at its discretion.

NOAA and WHOI acknowledge that the ESP is an experimental device for research purposes only, and as such will not hold MBARI liable for any damages that may be incurred as a result of using the instrument or its mooring in the laboratory or field.

MBARI Budget Justification

Personnel

C. Scholin, P.I., will oversee project planning and implementation in collaboration with co-I's Doucette, Anderson, Keafer, and McGillicuddy. He will also oversee activities at MBARI to fabricate, test, ship to and install equipment at partner institutions in concert with the MBARI engineering and science support team. He will spend 1 mo per year on this project.

Roman Marin III, Senior Research Specialist in Scholin's lab, will participate up to 6mos per year at no cost to this project. Mr. Marin will be responsible for production of custom reagents and arrays, support inter-lab calibration experiments and participate in field and laboratory studies as required.

The engineering team consists of Gene Massion (Mechanical Engineer), Scott Jensen (Electrical Engineer), Brent Roman (Software Engineer; ESP operation), Cheri Everlove (Mechatronics Engineer), Rich Schramm (ESP data management, GUI) and a machinist (TBD). Gene Massion will devote 1 mo time in year-1 &2, 0.5 mo. in year-3. He will be responsible for configuring moorings/dockside installations and contribute to marine operations. Scott Jensen is the lead engineer for the core ESP and will devote 1 mo time to this project each year. He will be responsible for overall operational integrity of the instruments and engineering matters related to field deployments and laboratory operations. He will participate in the training workshop, as well as off-site visits to NOAA and WHOI as required for installing equipment in partner labs and instrument deployment/recoveries. Brent Roman will be responsible for configuring the ESPs and dedicated laptops, and will devote 1 mo time to this project each year. He will provide support for integrating new toxins assays that will come from the Doucette lab, and will provide support for lab and field operations. Cheri Everlove will be responsible for procuring parts to build the ESPs and mooring, undertake assembly/documentation of the instruments and mooring, calibrate the instruments, participate in the user workshop, and will travel to partner institutions as required for laboratory and field operations support. She will devote 3 mo in year-1, 6 mo in year-2, and 4 mo in year-3. Rich Schramm will oversee the data management (DM)/user interface (UI) application that project participants will use throughout this project. Development of the DM/UI application is currently sponsored by MBARI and will reach operational status in 2008. This system will allow users to configure the ESP for extended missions and manage the data (and associated metadata) that will stem from those deployments. As the lead on the DM/UI effort, he will work with project participants to establish standards for sharing ESP data and data products via web access, and will participate in off-site laboratory and field trials as required. He will devote to this project 1.5 mo in year-1 and 1 mo each in year-2 & 3. Finally, 2 mo of machinist time (from the MBARI pool of machinists) is requested in year-1 for assisting with custom fabrication of ESP instrument and mooring components.

Capital Equipment

Funds are requested to purchase a 50m-rated pressure housing for the ESP ($13.25K) and CTD package ($10K) for outfitting the ESP mooring that will be deployed in outlaying years of the project. The CTD/fluorometer/transmissometer (or OBS) will be bundled with the ESP to provide contextual environmental data during field deployments. This equipment is essential to ESP field operations.

Fabricated Equipment

Both commercial-off-the-shelf (COTS) and custom manufactured parts (MP) are needed to construct the core ESPs and mooring. Based on a production run of several units, and assuming some minor re-work that may be required during the assembly (e.g. particular COTS part that is no longer available, changes to all lead-free electronics, processor upgrades, etc) as well as purchase of an initial set of spares that are compliant with this production run, cost of COTS and MP are estimated as follows:

Type of part	ESP instrument	ESP mooring
COTS	$40K	$30K
MP	$40K	$10K

Over time we expect costs of the ESP and mooring to decline significantly as production volumes increase and the design matures with larger scale production in mind.

Supplies

Funds are requested for all materials used to produce custom probe arrays, reagents and cultured and synthetic samples for inter-lab calibration (DNA probes, expendable reagents, expendable plastics, culture supplies, probe array membrane support, filters, etc.). Supplies total $10K in year-1 and $15K each for year-2 & 3. In addition to science supplies, we request $5K each in year-2 & 3 for instrument spares (electronic components, syringes, valves, cables, etc). Finally, we request $10K each in year-2 & 3 for spares/equipment maintenance to support field operations (batteries, custom fabrication for dockside deployment, mooring spares, telemetry system, batteries, etc. – all COTS or MP components that could be lost or damaged under normal operating conditions and would require immediate replacement during field trials/cruises to avoid interruption of ESP service).

Travel

A total of $3.6K per year is requested for travel to partner institutions to install/service equipment at partner labs (year-1), deploy/recover ESPs (year-2 & 3), participate in laboratory and field trials, and to attend annual project meetings, relevant workshops or scientific meetings. Each trip is estimated as being multi-day with expenses roughly as follows: $300 airfare, $500 hotel, $200 meals, $200 car/truck rental. In practice, each trip may be more or less than the assumed $1.2K per person per trip depending on the specifics of what is required of the project team. We expect persons from MBARI will participate in all field expeditions, and it may be necessary for either science or engineer personnel to travel to partner institutions in the event problems arise and can not be solved via phone or e-mail exchange.

Shipping

Shipping expenses are estimated at $5k per year. This includes charges to transport samples, reagents, instrument and spares, mooring equipment, etc. It may be necessary to ship the ESPs or components therein between partner institutions and MBARI for upgrade, repair or replacement as the study progresses. In some cases, shipments will require overnight service (e.g., some reagents or samples that require temperature control).

Indirect charges

All charges associated with labor, fringe benefits, travel, and non-capital expenses are subject to MBARI's federally negotiated indirect cost rate of 49% MTDC. Total pre-production for materials is $223.25K. Of that amount, $13.25K is for a 50m-rated pressure housing for the ESP and is considered capital equipment. Capital equipment also includes a dedicated CTD package ($10K) for the mooring. No indirect costs are applied to capital equipment (i.e., greater than 5K).

MBARI's Indirect Rate Agreement is established with NSF. The current Rate Agreement is dated September 4, 2007. Subrecipient WHOI's rate Agreement is established with ONR on a Salary and Fringe Benefit base, as described on the following page. NOAA does not include indirect costs in the total budget as it is a federal entity.

Budget

WHOI	Yr 1		Yr 2		Yr 3		Yr 1-3	
SALARIES/WAGES	# Months	2008 -2009	# Months	2009 - 2010	# Months	2010 - 2011	# Months	Total
Anderson - Co Principal Investigator	0.00	0	2.00		1.00		3.00	0
McGillicuddy - Co Principal Investigator	0.00	0	0.00		0.53		0.53	0
Anderson - Sr. Scientist	0.00		2.00		1.00		3.00	0
Keafer - Res Associate	0.78		4.00		2.75		7.53	0
2 Other Professionals	1.00		3.64		8.71		13.35	
TOTAL SAL & BEN		**11,065**		**92,592**		**122,251**	**0.00**	**225,908**
SUPPLIES:								
Laboratory supplies		4000		7,000		15,000		26,000
Subtotal Supplies		**4,000**		**7,000**		**15,000**		**26,000**
TRAVEL								
Domestic		3,205		3,822		2,481		9,508
Subtotal Travel		**3,205**		**3,822**		**2,481**		**9,508**
OTHER DIRECT COSTS								
Guest Student Stipends				16,200				
Computer Services & other						1,187		1,187
Shipping/Postage				1,682		1,561		3,243
Insurance				5,000		13,500		18,500
Tioga =- full day				14,400				14,400
Subtotal Shipping		**0**		**37,282**		**16,248**		**53,530**
Total Direct Costs		**18,270**		**140,696**		**155,980**		**314,946**
Laboratory Overhead		**6,453**		**54,000**		**71,034**		**131,487**
General & Administrative		3,401		28,463		37,441		69,305
Total Indirect Costs		**9,854**		**82,463**		**108,475**		**200,792**
TOTAL DIRECT & INDIRECT COSTS		**28,124**		**223,159**		**264,455**		**515,738**

Cumulative Budget

	Approximate Labor Months			
	09/01/08 08/31/09	**09/01/09 08/31/10**	**09/01/10 08/31/11**	**Total**
A. Senior Personnel				
1. DONALD M. ANDERSON, Co-PI	0.00	2.00	1.00	3.00
2. BRUCE A. KEAFER, Co-PI	0.78	4.00	2.75	7.53
3. DENNIS J. MCGILLICUDDY, Co-PI	0.00	0.00	0.53	0.53
B. Other Personnel				
2. Other Professionals	1.00	3.64	8.71	13.35
C. Total Salaries & Benefits	**$11,065**	**$92,592**	**$122,251**	**$225,908**
E. Travel				
1. Domestic	3,205	3,822	2,481	9,508
Total Travel	**3,205**	**3,822**	**2,481**	**9,508**
G. Other Direct Costs				
1. Materials and Supplies	4,000	7,000	15,000	26,000
4. Computer Svc	0	0	660	660
6. Other	0	37,282	15,588	52,870
Total Other Direct Costs	**4,000**	**44,282**	**31,248**	**79,530**
H. Total Direct Costs	**$18,270**	**$140,696**	**$155,980**	**$314,946**
I. Indirect Costs				
1. Laboratory Overhead	6,453	54,000	71,034	131,487
2. General & Administrative	3,401	28,463	37,441	69,305
Total Indirect Costs	**9,854**	**82,463**	**108,475**	**200,792**
J. Total Direct and Indirect Costs	**$28,124**	**$223,159**	**$264,455**	**$515,738**
L. Amount of this Request	**$28,124**	**$223,159**	**$264,455**	**$515,738**

Anderson component

	Approximate Labor Months			
	09/01/08 08/31/09	**09/01/09 08/31/10**	**09/01/10 08/31/11**	**Total**
A. Senior Personnel				
1 . D. ANDERSON, Senior Sci	0.00	2.00	1.00	3.00
2 . B. KEAFER, Res Assoc III	0.78	4.00	2.75 *	7.53
B. Other Personnel				
2. Other Professionals				
J. KLEINDINST, Ctr Admin II	0.00	0.00	0.50	0.50
K. NORTON, Res Asst II	1.00	3.64	2.50*	7.14
C. Total Salaries & Benefits	**$11,065**	**$92,592**	**$69,638**	**$173,295**
E. Travel (see detail)				
1. Domestic	3,205	3,822	2,481	9,508
Total Travel	**3,205**	**3,822**	**2,481**	**9,508**
G. Other Direct Costs (see detail)				
1. Materials and Supplies	4,000	7,000	15,000	26,000
6. Other	0	37,282	15,061	52,343
Total Other Direct Costs	**4,000**	**44,282**	**30,061**	**78,343**
H. Total Direct Costs	**$18,270**	**$140,696**	**$102,180**	**$261,146**
I. Indirect Costs				
1. Laboratory Overhead	6,453	54,000	40,350	100,803
2. General & Administrative	3,401	28,463	21,268	53,132
Total Indirect Costs	**9,854**	**82,463**	**61,618**	**153,935**
J. Total Direct & Indirect Costs	**$28,124**	**$223,159**	**$163,798**	**$415,081**
L. Amount of this Request	**$28,124**	**$223,159**	**$163,798**	**$415,081**

Travel Detail

Period 1 09/01/08 - 08/31/09

1. Domestic

trip to MBARI (Boston, MA to Monterey, CA)

Lodging	Nightly Rate (inc. Tax)	1 Rooms	14 Nights	@125	1,750	
Airfare	Round Trip	1 Tickets		@600	600	
Ground	General Grnd Transport.	1		@225	225	
Per Diem	Inside Continental US	1 People	14 Days	@45	630	
				Trip Total:	3,205	

Total Domestic Travel - Period 1 **3,205**

Period 2 09/01/09 - 08/31/10

1. Domestic

Scientific meeting (Boston, MA to San Francisco, CA)

Lodging	Nightly Rate (inc. Tax)	1 Rooms	6 Nights	@125	750	
Misc. Expens	Meeting Registration	1 People		@400	400	
Airfare	Round Trip	1 Tickets		@600	600	
Ground	General Grnd Transport.	1		@200	200	
Per Diem	Inside Continental US	1 People	6 Days	@45	270	
				Trip Total:	2,220	

Salt Pond field work (Woods Hole, MA to Eastham, MA)

Ground	Mileage	10		@53	530	
Per Diem	Inside Continental US	1 People	10 Days	@12	120	
				Trip Total:	650	

NH field work (Woods Hole, MA to Portsmouth, NH)

Lodging	Nightly Rate (inc. Tax)	2 Rooms	2 Nights	@125	500	
Ground	Mileage	2		@136	272	
Per Diem	Inside Continental US	2 People	2 Days	@45	180	
				Trip Total:	952	

Total Domestic Travel - Period 2 **3,822**

Period 3 09/01/10 - 08/31/11

1. Domestic

Scientific meeting (Boston, MA to San Francisco, CA)

Lodging	Nightly Rate (inc. Tax)	1 Rooms	6 Nights	@125	750	
Misc. Expens	Meeting Registration	1 People		@400	400	
Airfare	Round Trip	1 Tickets		@600	600	
Ground	General Grnd Transport.	1		@200	200	
Per Diem	Inside Continental US	1 People	6 Days	@45	270	
				Trip Total:	2,220	

Narragansett field work (Woods Hole, MA to Narragansett, RI)

Ground	Mileage	3		@87	261	
				Trip Total:	261	

Total Domestic Travel - Period 3 **2,481**

Other Direct Costs - Detail

	09/01/08 08/31/09	09/01/09 08/31/10	09/01/10 08/31/11	Total
G. Other Direct Costs				
1. Materials and Supplies				
a. Laboratory Supplies	4,000	7,000	15,000	26,000
Total Materials and Supplies	**4,000**	**7,000**	**15,000**	**26,000**
6. Other				
a. Guest Student Stipend	0	16,200	0	16,200
b. Insurance	0	500	0	500
c. Over the Side Insurance	0	4,500	13,500	18,000
d. Shipping & Postage	0	1,682	1,561	3,243
e. Tioga - full day	0	14,400	0	14,400
Total Other	**0**	**37,282**	**15,061**	**52,343**
Total Other Direct Costs	**4,000**	**44,282**	**30,061**	**78,343**

McGillicuddy component

	Approximate Labor Months			
	09/01/08 08/31/09	**09/01/09 08/31/10**	**09/01/10 08/31/11**	**Total**
A. Senior Personnel				
1 . D. ANDERSON, Senior Sci	0.00	0.00	0.00	0.00
2 . D. MCGILLICUDDY, Senior Sci	0.00	0.00	0.53	0.53
B. Other Personnel				
2. Other Professionals				
K. SMITH, Res Assoc III	0.00	0.00	5.71	5.71
C. Total Salaries & Benefits	**$0**	**$0**	**$52,613**	**$52,613**
G. Other Direct Costs (see detail)				
4. Computer Svc	0	0	660	660
6. Other	0	0	527	527
Total Other Direct Costs	**0**	**0**	**1,187**	**1,187**
H. Total Direct Costs	**$0**	**$0**	**$53,800**	**$53,800**
I. Indirect Costs				
1. Laboratory Overhead	0	0	30,684	30,684
2. General & Administrative	0	0	16,173	16,173
Total Indirect Costs	**0**	**0**	**46,857**	**46,857**
J. Total Direct & Indirect Costs	**$0**	**$0**	**$100,657**	**$100,657**
L. Amount of this Request	**$0**	**$0**	**$100,657**	**$100,657**

Other Direct Costs - Detail

	09/01/08 08/31/09	09/01/09 08/31/10	09/01/10 08/31/11	Total
G. Other Direct Costs				
4. Computer Svc				
a. Technical Assistance (CIS)	0	0	660	660
Total Computer Svc	**0**	**0**	**660**	**660**
6. Other				
a. Communications	0	0	225	225
b. Photocopying	0	0	225	225
c. Shipping & Postage	0	0	77	77
Total Other	**0**	**0**	**527**	**527**
Total Other Direct Costs	**0**	**0**	**1,187**	**1,187**

Statement of Work – WHOI

Year 1

WHOI personnel will travel to MBARI and participate in a Monterey Bay, CA deployment of an ESP for training purposes. Following training, we will make standards and perform relevant molecular assays for future validation studies at WHOI. We will also collect field samples during blooms in Salt Pond, Eastham, MA and the Gulf of Maine (GOM) and archive these samples for later ESP testing.

Year 2

After delivery of the first NOPP ESP from MBARI to WHOI, the Anderson laboratory will bench test and operate the unit with additional training and oversight from MBARI. Initial experiments will focus on running standard calibration series using synthetic, cultured and natural samples analyzed with both laboratory-based (96 well plate SHA and whole-cell formats) and ESP methodologies.

The initial ESP field deployment will be in Salt Pond, with ground truthing conducted on a weekly basis during the bloom season. We will also maintain the IOOS-funded ESP deployed at the Buoy B site in the Gulf of Maine. This will entail additional ground truthing and servicing not provided by IOOS funding. McLane Research will be invited to participate in these laboratory and field activities for potential commercialization.

Year 3

Work will require preparation and deployment of three ESPs within the open coastal waters of the GOM. Three cruises are scheduled - one for initial deployment; one for turn-around of all three ESPs; and one for the final recovery of the instruments. A UNOLS request has been submitted for these three cruises (total request 18 days). Near real-time data will be analyzed and compared with samples from these cruises to validate ESP performance.

In addition to the major cruise effort, assimilation of ESP data into our models will also be a significant effort in this year as all three ESPs begin reporting data. McGillicuddy will lead the modeling team to process the data and assimilate it into the nowcast/forecast system using the ROMS adjoint model. Assimilated data products will be distributed to our resource management partners on a timely basis.

Budget Justification - WHOI

Personnel: D.M. Anderson, PI, will be responsible for project planning and coordination of the Gulf of Maine and Salt Pond components. B. Keafer, co-PI, will assist with project planning and execution, and will organize and conduct bench testing, field operations to deploy/recover/ground truth the ESP, and *A. fundyense* analyses of the samples and ESP data stream. K. Norton, Research Assistant, will assist with all lab and field-testing of ESP including participation in cruises and whole cell and SHA sample processing. J. Kleindinst, Administrator, will provide support in year three, assisting with reports and publications.

In year three, co-PI D.J. McGillicuddy will oversee the effort to assimilate ESP data into the near-real-time quasi-operational forecast model of *A. fundyense* currently operating in the region[1]. The actual hands-on work will be carried out by Research Associate K.W. Smith. Mr. Smith is an expert in data assimilation techniques, and is familiar with the ROMS-based adjoint methodology. Smith will be responsible for processing the ESP data, preparing it for assimilation into the model, executing data-assimilative simulations, and visualizing the results. McGillicuddy will oversee the design of the numerical experiments and participate in the analysis of the results.

The Anderson laboratory has established a successful collaboration with Northeastern University in which coop students come to the lab for six-month periods, gaining knowledge and experience by working on a variety of research topics. To assist with this project, we are budgeting for two students in year two (see Other Costs below).

Travel. In year one, we are requesting funds for B. Keafer to travel to MBARI for training prior to ESP deployments. In years two and three, we have budgeted travel for field work: year two includes ten trips to Salt Pond (Eastham, MA) and two trips to Portsmouth, NH; year three includes three trips to Narragansett, RI to load and unload gear for the cruises. (Although we are requesting R/V *Oceanus* time in year three, we are budgeting a modest amount of travel to RI in the event that our shiptime is assigned on the R/V *Endeavor*.) In years two and three, we are requesting funds for one PI to attend a national scientific meeting (such as ASLO) to present results of this project.

Materials and supplies. We are requesting funds for the whole cell and SHA sample collection and processing. Supplies needed include: oligo-nucleotide probes, SHA plates, microscope supplies, various sample containers, filters, and 1 Dewar for Liquid N_2 sample storage.

Other Direct Costs. Over-the-side insurance will be needed for one ESP deployment in year two, and three in year three. Inland marine insurance will also be needed for one ESP unit in year two, which will be located at Salt Pond, Eastham, MA. Shiptime on the R/V *Tioga* (WHOI vessel) is also requested – 6 days in year 2. Funds for shipping in years two and three are needed to cover the cost of shipping samples between MBARI, NOAA-NOS and WHOI for intercalibration. We also anticipate some samples from 'ships of opportunity' and are budgeting funds to cover the cost of shipping samples to us. In year three, modest funds are budgeted for communications and photocopying as well as technical assistance from the WHOI computer center. Funds are requested in year two to support stipend costs for two Northeastern University coop (undergraduate students) for six months each. This project will provide diverse training to co-op students. The Northeastern University cooperative program has proved to be an excellent source of good students for other projects that are ongoing in this laboratory. For a modest cost, we are able to provide a significant learning experience to students who would otherwise not be exposed to oceanography.

Budget Information: The Woods Hole Oceanographic Institution calculates overhead rates (both Laboratory Costs and General & Administrative Costs) as a percent of total direct salaries

[1] http://omgrhe.meas.ncsu.edu/Redtide/Redtide_07/

and benefits, as allowed by OMB Circular A-122. Direct salaries exclude overtime-premium pay. A proposed labor month is equal to 152 hours or 1824 hours annually versus 2080 hours (40 hours/week for 52 weeks). The difference is for vacations, holidays, sick time, and other paid absences, which are included in Employee Benefits.

The rates included in the proposal are negotiated with ONR or they are estimates. When estimated rates are finalized, costs will be in accordance with the rate agreement.

With the assistance of Pricewaterhouse Coopers, WHOI undertook a study to determine what our 1997 combined F&A overhead rate would be, if we used the MTDC method used by most Universities, which are subject to OMB Circular A-21. We repeated the study and determined that our rate for 2004 would be 51.31%--lower than most research institutions and research-intensive universities.

VOLUME 2: COST PROPOSAL

APPENDIX 1. FEDERAL ENTITY – NOAA/NATIONAL OCEAN SERVICE

COST DETAIL

Applications of the Environmental Sample Processor (ESP) to Harmful Algal Bloom Management: Field Deployments, Technology Transfer, and Commercial Evaluation

Co-Investigator Dr. Gregory J. Doucette
Marine Biotoxins Program
Center for Coastal Environmental Health & Biomolecular Research
NOAA/National Ocean Service
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Charleston, SC 29412
Tel: 843-762-8528; E-mail: greg.doucette@noaa.gov

This proposal is submitted pursuant to the Broad Agency Announcement ONR-BAA-07-040, dated 21 August 2007, entitled "National Oceanographic Partnership Program (NOPP) and Interagency Committee on Ocean Science and Resource Management Integration (ICOSRMI)," Topic 3.

Funding Requested: $101,279

Period of Performance: May 1, 2008 to April 30, 2011

Gregory J. Doucette
Co-Investigator
Marine Biotoxins Program
NOAA/NOS/NCCOS/CCEHBR

Geoffrey I. Scott
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The National Ocean Service (DOC/NOAA/NOS) is a federal government agency.
Employer ID (EIN or TIN): 52-08216908

Budget

NOS/NOAA		Yr 1		Yr 2		Yr 3		Yr 1-3	
SALARIES/WAGES		# Months	2008	# Months	2009	# Months	2010	# Months	Total
	G. Doucette	2.00	0	2.00	0	2.00	0	6.00	0
	C. Mikulski			4.00	19,600	4.00	20,580	8.00	40,180
	Total Salary & Wages		**0**		**19,600**		**20,580**		**40,180**
	Total Benefits 55% of salary		0		10,780		11,319		22,099
	TOTAL SALARY & BENEFITS		**0**		**30,380**		**31,899**		**62,279**
SUPPLIES	Expendable Reagents (includes custom antibodies				13,000		7,500		20,500
	and protein conjugates for domoic acid and saxitoxin)								
	Protein arrayer maintenance				6,000		6,000		12,000
	Subtotal Supplies		**0**		**19,000**		**13,500**		**32,500**
TRAVEL	Yr. 1: 1 wk to MBARI - ESP training/assay calibration		2,000						2,000
	Yr. 2: 2 trips (1wk/ea) to WHOI - ESP deployment/recovery				3,000				3,000
	Yr. 3: 3 trips (1wk/ea) to WHOI - ESP deployment/						4,500		4,500
	turnaround/recovery								
	Subtotal Travel		**2,000**		**0**		**4,500**		**6,500**
Total Direct Costs			**2,000**		**49,380**		**49,899**		**101,279**
Total Indirect Costs	**Rate:** **0%**		**0**		**0**		**0**		**0**
TOTAL DIRECT & INDIRECT COSTS			**2,000**		**49,380**		**49,899**		**101,279**

Statement of Work – NOAA/ NOS FEDERAL ENTITY

This statement of work outlines the tasks to be performed under the direction of Co-Investigator Dr. Gregory Doucette, representing the Federal entity DOC/NOAA/National Ocean Service. This work constitutes an integrated, coordinated element of the proposal being submitted by Principal Investigator Dr. Christopher Scholin of the Monterey Bay Aquarium Research Institute, and Co-Investigators Drs. Donald Anderson and Dennis McGillicuddy of the Woods Hole Oceanographic Institute. The primary contributions of this component will be the laboratory-based refinement and calibration of an assay for domoic acid (DA) formatted previously for the Environmental Sample Processor (ESP) platform, as well as the initial development and testing of a new assay for the PSP toxin class. Essential to achieving these aims will be delivery of a core ESP instrument from MBARI to Dr. Doucette's laboratory, as described in the main body of this proposal.

The ESP-formatted DA assay was designed and developed as part of a previous NOPP-funded project (NSF OCE-0314089). A protocol for the extraction of DA from phytoplankton samples acquired by the ESP (> 90% recovery) was also developed as part of this work. The DA assay employs a competitive ELISA (cELISA) approach and utilizes a membrane-based protein array with the same chemiluminescence reporting system as used in the LSU rRNA probe-based assay for HAB species detection, thereby permitting 'multiplexing' of several reagents. Briefly, a toxin extract produced onboard the ESP (generally representing material concentrated from 500 to 1000 mL of seawater) is combined with a DA-specific antibody and exposed to a membrane array spotted with a DA-protein conjugate; the toxin in solution competes with the toxin-protein conjugate immobilized on the array for the antibody available in solution; antibody bound to the array (inversely proportional to the toxin concentration in the sample extract) is visualized with a chemiluminescent signal that is captured by an onboard CCD camera (see Fig. 2 in main proposal). The DA assay has undergone several successful test deployments on the ESP in Monterey Bay, CA during 2006 and 2007, representing the first ever autonomous, sub-surface detection of a phycotoxin (manuscript in prep.).

The next critical steps needed to move the DA assay towards a more 'operational' status are further refinement of the assay to minimize changes in response over one to two month deployments, as well as extensive calibration to critically establish assay performance parameters and reproducibility. This labor-intensive effort will require constant access to a core ESP instrument in our laboratory. To date the majority of our assay development work has been carried out using a 'hand-held' assembly consisting of an ESP 'puck' (i.e., membrane array holder) and syringes, which is not capable of accurately reproducing the precise microfluidics of the ESP instrument. Two attempts at calibrating the DA assay on the core ESP unit, using a non-toxic *Pseudo-nitzschia* extract spiked with certified DA reference material (NRC/IMB, Halifax, NS, Canada) to account for matrix effects, have been conducted during visits to Dr. Scholin's

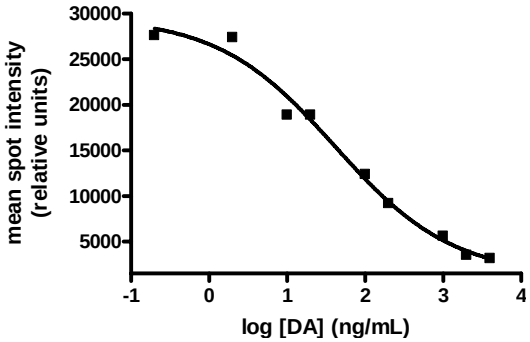

Fig. 1. Calibration curve for DA cELISA conducted onboard core ESP; EC50 (i.e., half-maximal assay response) is ~ 40 ng/mL.

laboratory at MBARI in 2006 and 2007. The most recent results (Fig. 1) indicate that this assay will permit detection of 0.2 ng DA/mL of seawater for a 0.5 L sample collected by the ESP, which is sufficiently sensitive to detect DA levels far below what is generally considered problematic or 'bloom' conditions. However, this result represents a single calibration, whereas it is essential to generate numerous curves to properly characterize assay reproducibility. Moreover, additional, iterative testing of modified assay reagents is required to further refine and assess the long-term stability of the DA test.

A second, equally important focus of our group during this project will be to initiate development and testing of an analogous assay for detection of PSP toxins onboard the ESP. This is especially relevant for ESP deployments planned for the western Gulf of Maine region, where PSP toxin-producing *Alexandrium* spp. will be targeted by the ESP arrays for HAB species detection. All of the information obtained during development and refinement of the DA cELISA for the ESP in terms of array spotting, reagent stability and reactivity, antibody/antigen ratios, etc. will be extremely valuable in designing the PSP toxin assay. We currently have direct access to antibodies against the PSP toxins, as well as saxitoxin-protein conjugates, through a collaborator on another project aimed at developing a surface plasmon resonance (SPR)-based detection method for this toxin class. These fully characterized, custom reagents will be provided at a minimal cost to the project. In fact, the DA antibody and DA-protein conjugate used in the ESP DA assay are currently employed in an SPR-based DA detection method run routinely in our laboratory (formatted as a cELISA; Traynor et al., 2006). We are confident that adopting this same strategy for development of the PSP toxin assay will bring a high probability for success. We should note that the PSP antibodies currently in hand show reduced cross-reactivity with the N-1 hydroxy saxitoxin derivatives (i.e., NEO, GTX1/4, C3/4); however, several parallel efforts outside the scope of this project are being pursued to address this issue and any antibodies developed will be freely available for the work proposed herein.

A primary component of proposal Year 1 essential for carrying out much of the work outlined above will be delivery of a core ESP instrument to our laboratory near the end of the year. Prior to this, our efforts during Year 1 will continue employ the hand-held apparatus described above and used for much of our assay development thus far; however, materials will be sent periodically to Dr. Scholin's laboratory for testing/evaluation on their available core ESP units. We can, to a certain extent, continue to refine the DA cELISA and, importantly, importantly, initiate the concurrent development of the PSP toxin extraction method and ESP-formatted assay using this hand-held device. We will also plan a visit to MBARI during Year 1 for training on laboratory-based operation of the core ESP (we do have some experience already from our two previous visits to perform assay calibrations) so as to minimize down-time following delivery of the instrument. We will likely field at least several DA arrays during the planned ESP training deployment in the western Gulf of Maine in cooperation with the NERACOOS Program (Buoy B). Additional ESP deployments will be conducted in Monterey Bay by Dr. Scholin during the Year 1 time frame (but apart from the current proposal) and we will take advantage of these opportunities to continue our field-based testing of the DA assay.

With the planned delivery of the core ESP instrument to our laboratory by the start of Year 2, much of our focus will turn to the intensive refinement, evaluation, and calibration of the DA cELISA on this unit. We have considerable experience (over 15 years) with the development and

validation of *in vitro* assays for algal toxins and the work required for this process will now be possible with constant access to an ESP instrument. We also expect that following our Year 1 effort, at least a preliminary version of the PSP toxin assay will also be ready for initial formatting/testing on the core ESP. Continued development of this assay should proceed with far greater efficiency given both the availability of an ESP instrument (as compared to use of the hand-held version) and the knowledge gained from our work on the analogous DA cELISA. In addition, we will plan to perform initial field tests of the PSP toxins assay during proposed test deployments of the ESP in the western Gulf of Maine (i.e., Salt Pond, MA; NERACOOS/Buoy B).

Work during Year 3 will shift exclusively to the PSP toxin assay, with our aim being to establish a functional assay for the planned field work in the western Gulf of Maine, which will involve the simultaneous deployment of three ESP instruments. Based on what is learned from the Year 2 deployment, we expect that additional refinements to reagents may be necessary to address the issue of assay stability over extended periods in the field. In addition, efforts will be focused on the rigorous calibration of this assay and establishing assay performance parameters, as will have been carried out for the DA assay during Years 1 and 2.

References

Traynor, I.M., Plumpton, L., Fodey, T.L., Higgins, C., Elliott, C.T. 2006. Immunobiosensor detection of domoic acid as a screening test in bivalve molluscs: comparison with liquid chromatography-based analysis. J. AOAC Int. 89, 868-872.

Budget Justification

Support for the NOAA/NOS portion of this project involving detection of domoic acid and PSP toxins onboard the ESP, including personnel, supplies, and travel, is being requested as follows: Yr. 1: $2,000; Yr. 2: $49,380; Yr. 3: $49,899.

Personnel

G. Doucette will dedicate two months per year to this component of the project - as a federal government employee, salary support is not required and is given as in-kind support. He will be responsible for application of the ESP for detection of domoic acid and PSP toxins by overseeing the assembly, quality control and provision of all dedicated reagents/supplies employed for this assay as well as the processing and evaluation of data generated. All activities will be coordinated closely with PI Dr. Christopher Scholin (MBARI) and Co-Investigators Drs. Anderson and McGillicuddy (WHOI) who will be responsible collectively for logistical components associated with configuration, deployment, and operation of the ESP.

Salary support is requested for C. Mikulski (Biol. Res. Tech.) at four months per project year during Years 2 and 3. She will be responsible for preparation, testing, and final quality assurance of all reagents/supplies (e.g., toxin arrays) required for conduct of the domoic acid and PSP toxin assays onboard the ESP. She will prepare for and carry out all ESP instrument/assay calibrations

ahead of all planned deployments in cooperation with the MBARI and WHOI groups. She will also be responsible for processing/initial evaluation of all raw toxin data generated by the ESP.

Supplies

Supplies requested for deployment of the domoic acid and PSP toxin assays onboard the ESP are as follows: Expendable Reagents - all commercially available chemicals, toxin certified reference standards, filters, and expendable plastics; custom DA antibody/DA-protein conjugate and custom saxitoxin antibodies/saxitoxin-protein conjugates are included in the Expendable Reagents category at a combined cost of $7,500 charged in Year 2 and $2,000 charged in Year 2 (PSP assay reagents only); Protein arrayer maintenance - parts and service for annual maintenance of Biodot protein arrayer used to print DA and saxitoxin arrays.

Travel

Support is requested for travel of one person from NOAA (Charleston, SC) to MBARI (Moss Landing, CA) for one week during Year 1 to participate in ESP training and to conduct DA assay calibration (this will take place prior to the delivery of a core ESP unit to NOAA). During Years 2 and 3, support is requested for travel of one person from NOAA to WHOI (Woods Hole, MA) to participate in ESP field deployments. In Yr. 2, this will involve two trips of one week duration for ESP deployment and recovery; in Yr. 3 three trips of one week each will be required for ESP deployment, turnaround, and recovery. The individual making the trips to WHOI in Yrs. 2 and 3 will also participate in the associated survey cruises as part of these deployments.

CERTIFICATION REGARDING RESTRICTIONS ON LOBBYING
(Proposals Greater than $100,000)

Proposal Title

Applications of the Environmental Sample Processor (ESP) to Harmful Algal Bloom Management: Field Deployments, Technology Transfer, and Commercial Evaluation

Proposal Number and/or Date 10 Dec 2007

The above referenced proposal was submitted in response to: (select one)

_____ The ONR Broad Agency Announcement (BAA) #08-001, as published in FedBizOpps on 5 Sep 07.

__X__ Other BAA (List Solicitation No., Project Title, and Date of Issuance):

ONR BAA-07-040

Applicants should review the instructions for certification requirement under 32 CFR Part 28, "New Restrictions on Lobbying." 32 CFR Part 25, The certification shall be treated as a material representation of fact upon which reliance will be placed when the Office of Naval Research determines to award the covered transaction, grant, or cooperative agreement.

CERTIFICATION REGARDING LOBBYING ACTIVITIES

The following certification applies only to actions exceeding $100,000:

Section 1352, Title 31, U.S.C. (PL 101-121, Section 319) entitled "Limitation on use of appropriated funds to influence certain Federal contracting and financial transactions."

(1) No Federal appropriated funds have been paid or will be paid by or on behalf of the undersigned, to any person for influencing or attempting to influence an officer or employee of an agency, a Member of Congress, an officer or employee of Congress, or an employee of a Member of Congress in connection with the awarding of any Federal contract, the making of any Federal grant, the making of any Federal loan, the entering into of any cooperative agreement, and the extension, continuation, renewal, amendment, or modification of any Federal contract, grant, loan, or cooperative agreement.

(2) If any funds other than Federal appropriated funds have been paid or will be paid to any person for influencing or attempting to influence an officer or employee of any agency, a Member of Congress, an officer or employee of Congress, or an employee of a Member of Congress in connection with the Federal contract, grant, loan, or cooperative agreement, the undersigned shall complete and submit Standard Form-LLL, "Disclosure Form to Report Lobbying," in accordance with its instructions.

(3) The undersigned shall require that the language of this certification be included in the award documents for all subawards at all tiers (including subcontracts, subgrants, and contracts under grants, loans, and cooperative agreements) and that all subrecipients shall certify and disclose accordingly.

This certification is a material representation of fact upon which reliance was placed when this transaction was made or entered into. Submission of this certification is a prerequisite for making or entering into this transaction imposed by Section 1352, title 31, U.S.C. Any person who fails to file the required certification shall be subject to a civil penalty of not less than $10,000 and not more than $100,000 for each such failure.

As the duly authorized representative of the applicant, I hereby make the above certifications on behalf of the applicant.

Applicant Institution:

MONTEREY BAY AQUARIUM RESEARCH INSTITUTE

Taxpayer Identification Number (TIN):

77-0150580

Printed Name, Title and Signature of Authorized Representative: X

Patrice A Carroll

PATRICE A. CARROLL, SR. GRANTS SPECIALIST

Date: 05 DEC 2007