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## MEMORANDUM

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**DATE:** MARCH 12, 2018

**To:** Mat Fuzie  
Deputy Director  
California State Parks  
Off-Highway Motor Vehicle Recreation Division  
1725 23<sup>rd</sup> Street, Suite 200  
Sacramento, CA 95816

**FROM:** Will Harris  
Senior Engineering Geologist

**SUBJECT:** Scripps Study of Marine Contributions to Aerosol Particulates  
Oceano Dunes State Vehicular Recreation Area, San Luis Obispo County, California.

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Deputy Director Fuzie,

Attached herein, please find the March 6, 2018 report entitled “Marine Contributions to Aerosol Particulates in a Coastal Environment.” This document was prepared by Dr. Brian Palenik of the Scripps Institution of Oceanography at UC San Diego (Scripps) as part of his investigation of particulates found on ocean-facing structures at the Oceano Dunes State Vehicular Recreation Area (Oceano Dunes).

As you know, every spring in south San Luis Obispo County (south county), strong prevailing winds blowing from over the ocean, out of the west-northwest, provide the key force that gives shape to the coastal sand dunes in the south county. Because of the prevailing winds, conditions offshore from Oceano Dunes and vicinity are ideal for large planktonic blooms, particularly during the spring (Walter, Armenta et al., 2018; Garcia-Reyes and Largier, 2012; Tognazzini, 2009). The west-northwest winds run parallel to the coast from Point Buchon to Point San Luis, pushing warm surface water towards and along that part of the shore that faces west (e.g., Oceano Dunes). Deeper, colder, nutrient-rich seawater upwells offshore from the west-northwest line of coast in response to the displaced surface water. When this water reaches the surface, sunlight warms it causing various planktonic species to grow. The amount of plankton varies based on seasons and conditions, but humpback whale and other cetacean feeding activity in the area provides anecdotal evidence that the production of plankton can be prolific (<https://www.youtube.com/watch?v=GeD5i0DadVA>). Overall, the amount of upwelling that occurs may be increasing because the strength of the northwesterly winds during the spring and summer has slowly increased over time based on a review of wind data from the Diablo Canyon nuclear power plant, which is located at Point Buchon (Lindsey, 2013).

During prevailing wind episodes, as observed at Oceano Dunes, a film of very fine grit accumulates on the ocean-side surface of just about anything the stands above the dunes. Posts, signs, fencing, even vegetation, become coated with this material (Figure 1). Under microscope the material appears to be biologic, possibly planktonic (Figure 2).

Sea/air interactions related to plankton blooms have been demonstrated by several researchers, including those at the Scripps Institution of Oceanography. For example, aerosolized planktonic algae have been shown to influence the formation of coastal clouds and fog (Prather, Bertram et al., 2013).

Dr. Brian Palenik, professor of marine biology at Scripps, has examined these interactions. He was intrigued by the aforementioned grit at Oceano Dunes and so designed a study that examined the biological signatures in seawater offshore from Oceano Dunes and that of the very fine grit material (less than 10 microns in diameter) collected from a variety of media, including filter tapes from EBAM air samplers deployed in the sand dunes. Samples were collected in 2014, 2015, and 2016 and examined for the presence of marine prokaryotes (bacteria) and marine eukaryotes, such as phytoplankton diatoms, using DNA sequence analysis.

From the analyses performed on samples of seawater, dune sand, surf foam, and the airborne grit, Dr. Palenik was able to determine that “nearby coastal seawater is contributing biological material to PM10 aerosols (10 microns or less) detected and captured inland at monitoring sites using EBAMs. This likely includes whole microbes, ranging from the one micron sized bacteria to small eukaryotes less than 10 microns such as small diatoms.” Further, Dr. Palenik notes, “We did find diatom DNA on EBAM filters from *Chaetoceros calcitrans* and *Thalassiosira pseudonana*, two small marine species that are less than 10 microns. DNA from these diatoms was found on some near-beach fencing but not in dune sand samples, suggesting they are transported as whole-cell aerosols from seawater to the EBAM filters.”

The findings from this work have bearing on the ongoing efforts to mitigate airborne PM10 detected in Nipomo Mesa (Mesa), which is approximately two miles downwind from Oceano Dunes. At present, all mitigation efforts are focused on the reduction of saltation-derived dust emanating from locations within the 1500 acre off-highway vehicle (OHV) riding area of Oceano Dunes. Additionally, PM10 air dispersion modeling undertaken collaboratively with the California Air Resources Board and the San Luis Obispo County Air Pollution Control District considers saltation-derived dust from the OHV riding area of Oceano Dunes as the sole source of PM10 ultimately detected on the Mesa. As demonstrated by the Scripps investigation, the sourcing of airborne PM10 detected in this coastal setting is more complex, a function of several phenomena that occur on land, the ocean, and in the air. An ocean source of planktonic and related bacterial PM10 is potentially far greater than other sources because the area of ocean surface where phytoplankton blooms occur offshore from Oceano Dunes is tens of thousands of acres larger than the OHV riding area of the State Park.

In the cover letter to his report, Dr. Palenik notes that the findings of his investigation have been presented at the 2018 Ocean Sciences Meeting, which is sponsored by the American Geophysical Union, among others. Dr. Palenik also anticipates publishing the work in a scientific journal, and he intends to conduct further work in the area to more completely quantify the marine biological signature of PM10 dust detected in the south county.

I am available for any questions you may have regarding the Scripps investigation or related topics.

Respectfully submitted,

Will J. Harris, PG 5679, CEG 2222, CHg 750  
Senior Engineering Geologist



Concur:

William R. Short, PG 4576, CEG 1429, CHg 61  
Supervising Engineering Geologist



Attachments: Figures 1 and 2;  
Scripps Investigation – Marine Contributions to Aerosol Particulates in a Coastal Environment by Dr. Brian Palenik and Maitreyi Nagarkar, March 6, 2018.

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Figure 1. Grit on plasticized screen, Plover Exclosure fence, Oceano Dunes SVRA, May 2011 (photo by Will Harris).

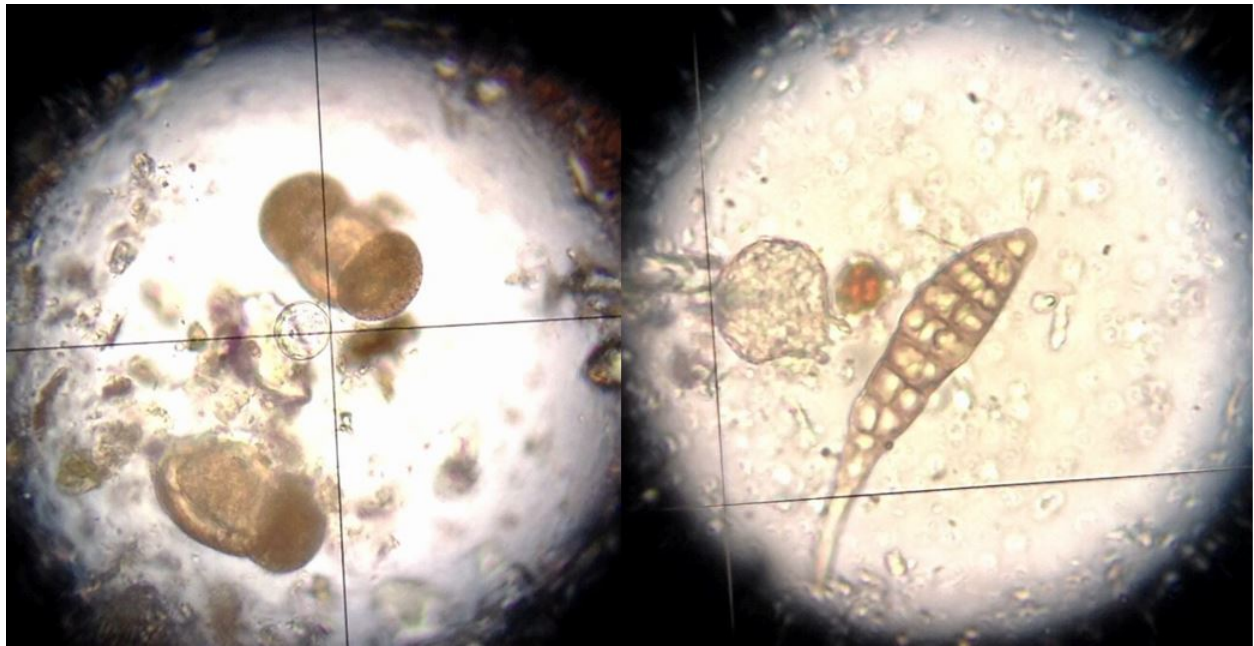


Figure 2. Photomicrographs of grit particles that were collected from plasticized screen shown in Figure 1 above (photomicrographs by Will Harris).



Will Harris  
Senior Engineering Geologist  
California Geological Survey  
801 K Street, MS 12-30  
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March 6, 2018

Dear Mr. Harris,

Please find attached our report of findings regarding the marine contribution to aerosolized particulates in south San Luis Obispo County (south county). As stated in the report, our analysis shows nearby coastal seawater is contributing biological material to PM10 aerosols (10 microns or less) detected and captured inland at EBAM monitoring sites within the dune environment of the south county. Detected marine-derived materials within the PM10 fraction include prokaryotes (bacteria) and eukaryotes, such as small diatoms.

Our findings from this investigation have been presented at the 2018 Ocean Sciences Meeting co-sponsored by AGU, ASLO and TOS societies. Additionally, we are in the process of submitting this work to a scientific journal for publication.

We would like to extend our appreciation to the California Geological Survey and to the California Department of Parks and Recreation for the assistance and access that made our investigation possible. We look forward to continued collaboration in future endeavors.

Sincerely,

A handwritten signature in cursive script that reads "Brian Palenik".

Brian Palenik  
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858-534-7505

## **Report: Marine Contributions to Aerosol Particulates in a Coastal Environment**

**Brian Palenik**  
**Maitreyi Nagarkar**

**Scripps Institution of Oceanography**  
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**La Jolla, CA 92093-0202**

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**Submitted 3/6/2018**

### **Introduction**

There is increasing interest in the contribution of marine-derived aerosols to particulates found over land. These marine aerosols may contain inorganic, organic, or biological materials. In studies of the marine atmosphere, about 10% of particulate matter by volume has been attributed to biological material, introduced either from land or through mechanisms such as bubble bursting at the sea surface (Matthias-Maser, Obolkin et al. 2000) (Aller, Kuznetsova et al. 2005) (Matthias-Maser, Brinkmann et al. 1999), with the latter mechanism likely dominant.

Once marine-derived aerosols move over land they can have multiple effects such as contributing to nucleating water droplet formation (cloud condensation nuclei) and thus fog/cloud formation or even potentially on human health (Prather, Bertram et al. 2013). As an example of the latter, some toxic marine phytoplankton have been found to produce aerosolized toxins and subsequent health effects (Hoagland, Jin et al. 2009, Kirkpatrick, Fleming et al. 2011). There has been an increasing appreciation that marine aerosols can contain microbes both prokaryotic and eukaryotic and these may vary with seawater conditions and distance from land (Cochran, Laskina et al. 2017, Rastelli, Corinaldesi et al. 2017) (Hiraoka, Miyahara et al. 2017) (Mayol, Arrieta et al. 2017) (Xia, Wang et al. 2015) (Woo, Brar et al. 2013) (Dueker, O'Mullan et al. 2012) (Dueker, Weathers et al. 2011) (Urbano, Palenik et al. 2011).

Here we determined the potential link between local marine waters and the composition of airborne material blown shoreward by prevailing winds to the coastal dunes in south San Luis Obispo County. This site was chosen as it shows seasonally high winds thought to promote local phytoplankton blooms and associated bacteria and is involved in local sand dune formation at Oceano Dunes State Vehicular Recreation Area (Oceano Dunes). Oceano Dunes is a California state park in San Luis Obispo (SLO) County that lies within the northern end of the Guadalupe Nipomo Coastal Dune Complex. The dune complex consists of more than 18,000 acres of coastal dunes that stretch from southern San Luis Obispo County into northern Santa Barbara County. The microbial community of the dunes has not been explored. Oceano Dunes SVRA consists of about 3,600 acres, with about 1,500 of those acres devoted to camping and off-highway vehicle (OHV) recreation.

We used DNA sequence analysis as a qualitative measure of compositional information, with 16S rRNA gene sequences as a measure of bacterial aerosols and 18S rRNA gene sequences as a measure of eukaryotic-derived aerosols. DNA sequences from marine microbes typically found in seawater was used as an indicator of a marine component of local waters finding its way into the total pool of airborne particulates. Initially we compared the sequences from the airborne particulates only to sequences found from our seawater samples, but later on we also incorporated marine sequences from public databases. We included analysis of a range of particulates including those caught on dune fencing to the very fine material (approximately 10 microns in diameter, also called PM10) that could affect air quality. We show that a commercial environmental beta particle attenuation monitor (EBAM) system (Met One Instruments, Inc) often used for air quality monitoring can be analyzed for microbial components using DNA sequence analyses of their sampling “tapes”.

## **Results**

32 samples were collected from seawater, beach foam, beach and dune sand, or air filters (EBAM tapes) at several different sites on different dates (Table 1). Total DNA was extracted from each of these and the Illumina platform was used to sequence either the 16S (for bacterial community composition) or 18S (for eukaryotic community composition) amplicon. Sequences were then clustered into predicted species (referred to as operational taxonomic units, or OTUs) based on sequence similarity and classified using the Silva v123 database.

Significantly, DNA from marine microbes in seawater samples was found translocated shoreward. It was caught on different particle collecting systems including dune wind fencing and screens and on commercial E-BAM systems, thus providing information about the contribution of marine aerosols to inland particle composition (Table 1, Supplemental Table 1)

We used the OTUs found in the three seawater samples (from two different dates) to identify “marine” bacterial and eukaryotic OTUs, against which we compared the sequences found at the other sites. From the 16S data, we found that between all the seawater samples there were 1348 OTUs with at least two sequences present and 1596 OTUs with a least one sequence present. Many of these OTUs were known marine species such as *Pelagibacter*.

We then quantified the presence of these seawater OTUs in the EBAM air samples and samples from the other sites. We calculated the proportion of total sequences from each site that represented marine OTUs (OTUs found in the seawater samples) by adding the number of sequences represented by marine OTUs and dividing by the total number of sequences from the sample. Marine bacterial sequences collected on the EBAM filters varied from 0 to 44 % of the total sequences obtained from a specific filter, showing a large component of marine bacteria to be likely present in the EBAMs. EBAM data is shown in bold in Table 1.

We combined the sequences across all the air samples to determine the top 25 most abundant bacterial OTUs on the air filter samples from the EBAMs (Table 2). The most abundant non-marine bacterial taxa were typical soil microbes such as *Dyella*. Four of the twenty five top OTUs were putative marine bacteria: *Fluviicola*, *Amylibacter*, *Paracoccus*, and a *Pseudomonas* seen in seawater, with *Fluviicola* and *Amylibacter* being the two most clearly of marine origin due to their strong representation in the seawater samples. Other marine bacteria, including *Vibrio* species, were clearly present in some, but not all, air samples (Supplemental Table 1).

The most abundant marine microbe on the air filters was a *Fluviicola like species* from the family Cryomorphaceae and this microbe was also the most abundant microbe in the beach foam sample (Table 1). In addition, two other beach foam samples from San Diego were also analyzed and found to be dominated by similar bacteria (data not shown).

We took the same approach with 18S rRNA amplicon sequences and used the marine seawater sample eukaryotes as an indicator of marine eukaryotic components of airborne particulates. There were 3907 OTUs found in seawater, similar to numbers found in coastal seawater in other studies (Nagarkar et al in review). Marine eukaryotic sequences collected on EBAM filters varied from 0 to 54% of the total sequences when determined using this approach.

Interestingly, no marine eukaryotic OTUs that were present in the seawater samples were found on EBAM samples from 2016, so the above analysis was not sufficient for determining the translocation of marine eukaryotic species to the air filters. However, the abundant OTUs in the 2016 EBAM samples included a marine diatom species. This OTU was also found on fencing (PS-2) very near the ocean, suggesting a seawater source. We believe that while these diatoms were not abundant in the reference seawater sample collected by lifeguards beyond the surf zone, but they may have been abundant in the seawater closer in to the beach.

Since the seawater samples we analyzed for 18S sequences did not provide a complete reference for all possible marine OTUs, we performed a second analysis where we determined if abundant aerosol 18S OTUs were of marine origin based on their DNA sequence similarity to that of common marine phytoplankton reported by others in published analyses. We used mothur (Schloss, Westcott et al.) and BLAST for this comparative sequence analysis. We added these newly marine-annotated OTUs into OTUs already identified as being in seawater samples based on their presence in the reference seawater samples. This analysis showed that all EBAM samples, including ones in 2016, contained eukaryotic marine phytoplankton DNA. The most abundant eukaryotic OTUs on EBAM filters are shown in Table 3 and include marine diatoms. It is significant that diatoms, in contrast to many other microbes, will contribute frustule silica in addition DNA to the aerosol. They will thus contribute, along with sand, to the Si content of aerosols.



In contrast to seawater and even marine sands and sediments (Probandt, Eickhorst et al. 2017) (Gobet, Böer et al. 2011), little is known about the microbiology of dune sands and whether this is a unique ecosystem or one largely similar to terrestrial soils. The microbial community of dune sand specifically from Oceano Dunes has not been studied. We thus examined the microbial composition of dune sand from areas protected from dune riding and from areas where dune off road vehicle riding is allowed. The bacterial composition of these two were largely similar. The ten most abundant 18S OTUs in four dune samples (Table 4) were not abundant in EBAMs, suggesting the dune was not a large contributor of microbes to EBAMs (but could still contribute inorganic material). However, we don't believe we have sufficient information about dune microbes to determine a specific percentage contribution of the dune microbes to aerosol sample DNA.

We also found OTUs from eukaryotic microalgae (found in BLAST but not in the seawater sample) to be present in dune sand. These were from *Trebouxia* sp. and *Apatococcus lobatus*. Their DNA sequences were similar to reported sequences from algae growing on rocks and buildings (Gorbushina and Broughton 2009, Gustavs, Schumann et al. 2016) so this may be an endemic dune population of microalgae or part of a larger terrestrial population. In addition, we found what appears to be sand mite DNA (closest relative *Micropsammus*) and nematode DNA (*Meloidogyne hapla* strain MeloHap11 ) suggesting also that the dune may have an interesting “ecosystem” of organisms. We also found DNA from sunflower (*Helianthus pauciflorus* subsp. *Subrhomboideus*) and slash pine (*Pinus elliottii*) presumably from pollen. From our small sample set, we found no evidence that recreational vehicles were affecting the community composition of this potential ecosystem as similar eukaryotes were found in riding and undisturbed areas.

## **Discussion and Conclusions**

Nearby coastal seawater is contributing biological material to PM10 aerosols (10 microns or less) detected and captured inland at monitoring sites using EBAMs. This likely includes whole microbes, ranging from the one micron sized bacteria to small eukaryotes less than 10 microns such as small diatoms. Potentially fragments of larger eukaryotic phytoplankton including their DNA can be found on EBAM filters. For example, dinoflagellates are typically larger than 10 microns, but we did find some dinoflagellate DNA. The use of DNA as a marker of this contribution is largely a qualitative one, because the copy number of the 16S region can vary among bacteria (marine and soil). In some cases, we did not detect marine bacterial DNA in EBAM filters while in others the contribution appeared to be very large. The same was true of the eukaryotic contribution. Thus, the processes determining when marine aerosol contributions are large are still unknown, but are likely dependent on wind speed, the presence of phytoplankton blooms in coastal waters and possibly their susceptibility to aggregation or lysis.

We did find diatom DNA on EBAM filters from *Chaetoceros calcitrans* and *Thalassiosira pseudonana*, two small marine species that are less than 10 microns. DNA

from these diatoms was found on some near-beach fencing but not in dune sand samples, suggesting they are transported as whole-cell aerosols from seawater to the EBAM filters. Interestingly, DNA from the diatom *Pseudo-nitzschia* was found in some EBAM samples. Some *Pseudo-nitzschia* species produce the toxin domoic acid with known toxicity to marine life (Ryan, Kudela et al. 2017, Zhu, Qu et al. 2017). It would be interesting to see whether domoic acid can be found in water and aerosols during blooms of this diatom species. These species are monitored on the California coast at specific monitoring sites (<http://www.sccoos.org/data/habs/>). It is not clear what the impact of aerosolized domoic acid would be.

We found that bacteria from the family Cryomorphaceae ((Muramatsu, Takahashi et al. 2012) ) were present in beach foam and in EBAM filters. Foam was very abundant during sampling in 2016. Cryomorphaceae have been associated with phytoplankton blooms (Pinhassi, Sala et al. 2004) and diatoms (e.g. Flo-31) in (Grossart, Levold et al. 2005). This particular bacterial group and its possible association with foam and high abundance in aerosols warrants further attention. There is no information for example on whether this group could be a human allergen, although endotoxins from bacteria have been measured in aerosols (Lang-Yona, Lehahn et al. 2014).

One intriguing possibility suggested by our results is that beach foam is a causative agent of aerosols in addition to direct aerosol formation of seawater, eg sea spray. Presumably both of these mechanisms are occurring depending on the presence of phytoplankton blooms and foam formation from their decomposition. This could be further explored by sampling on dates with varying degrees of foam present.

## **Methods**

EBAM air filters (Met One Instruments, Inc.) were cut to obtain the circular area containing the aerosols and then cut into smaller pieces and processed for standard DNA isolation from filters. Sand containing materials such screens, beach sand, and dune sand were washed with Phosphate buffered saline (PBS) containing 0.5M NaCl. The washes were filtered onto 2 x 25mm 0.2 um Supor filters and both filters were used for DNA extraction (Tai and Palenik 2009). For “orange” and “black” fencing, the fencing was washed with TE and the TE wash was used for DNA isolation. Foam was diluted with an equal volume of TE before DNA isolation.

DNA was checked for PCR quality by running a standard 16S or 18S rRNA amplification with the primers listed below. Total DNA was then sent to Research and Testing Laboratories (RTLGenomics, Lubbock, TX) for Illumina amplicon sequencing of the 16S or 18S rRNA gene. Primers used for 16S amplification were 28F (GAGTTTGATCNTGGCTCAG) and 388R (TGCTGCCTCCCGTAGGAGT). Primers used for 18S amplification were TAREukF (CCAGCASCYGC GGTAATTCC) and TAREukR (ACTTTCGTTCTTGATYRA) (Stoeck, Bass et al. 2010).

Sequences were clustered into OTUs using the `pick_open_reference_OTUs.py` pipeline in Qiime (Caporaso, Kuczynski et al. 2010), which assigns species-level OTUs at the 97% sequence similarity cutoff. The resulting biom file was converted into an OTU table and

sequences were classified using mothur (Schloss, Westcott et al. 2009) against the Silva v128 database (Quast, Pruesse et al. 2013).

## Figures and Tables

Figure 1. Map of Oceano Dunes sampling sites.

Table 1. Samples descriptions and fraction (of 1) of total DNA sequence reads from each sample that were likely of marine origin, based on comparison to either only the seawater OTUs (“SW only”) or those and database-derived sequence (“SW + DB”). “Singlets removed” refers to the same sample but only including sequences present two or more times. ND, not determined. The data shows that marine microbes make substantial contributions to EBAMs, as seen in the DNA recovery from EBAM filters (bolded).

Table 2. Top 25 most abundant bacterial OTUs in EBAM samples (with all EBAM samples combined). Bold shows predicted marine bacteria (based on presence of those OTUs in the seawater samples), with the exception of *Pseudomonas*, which may be due to dune input. Dunes may be contributing some bacteria such as FFCH11085 given the abundance of this sequence in the dune reads.

Table 3. Top 25 most abundant eukaryotic OTUs found in EBAM samples. Bold shows taxa predicted by presence in seawater sample. Other marine taxa were found by taxonomic knowledge and BLAST annotation.

Table 4. The most abundant eukaryotic OTUs in four dune sand samples. These OTUs were not typically found on EBAMs. A few appear to be microbial residents in the sand while others are from nearby plants.

## Acknowledgements:

This project received sampling and DNA processing help from Dr. Bianca Brahmsha (SIO), Emy Daniels (SIO), and Karl Hong (SIO). Sequencing was provided by RTLGenomics, Lubbock, TX.

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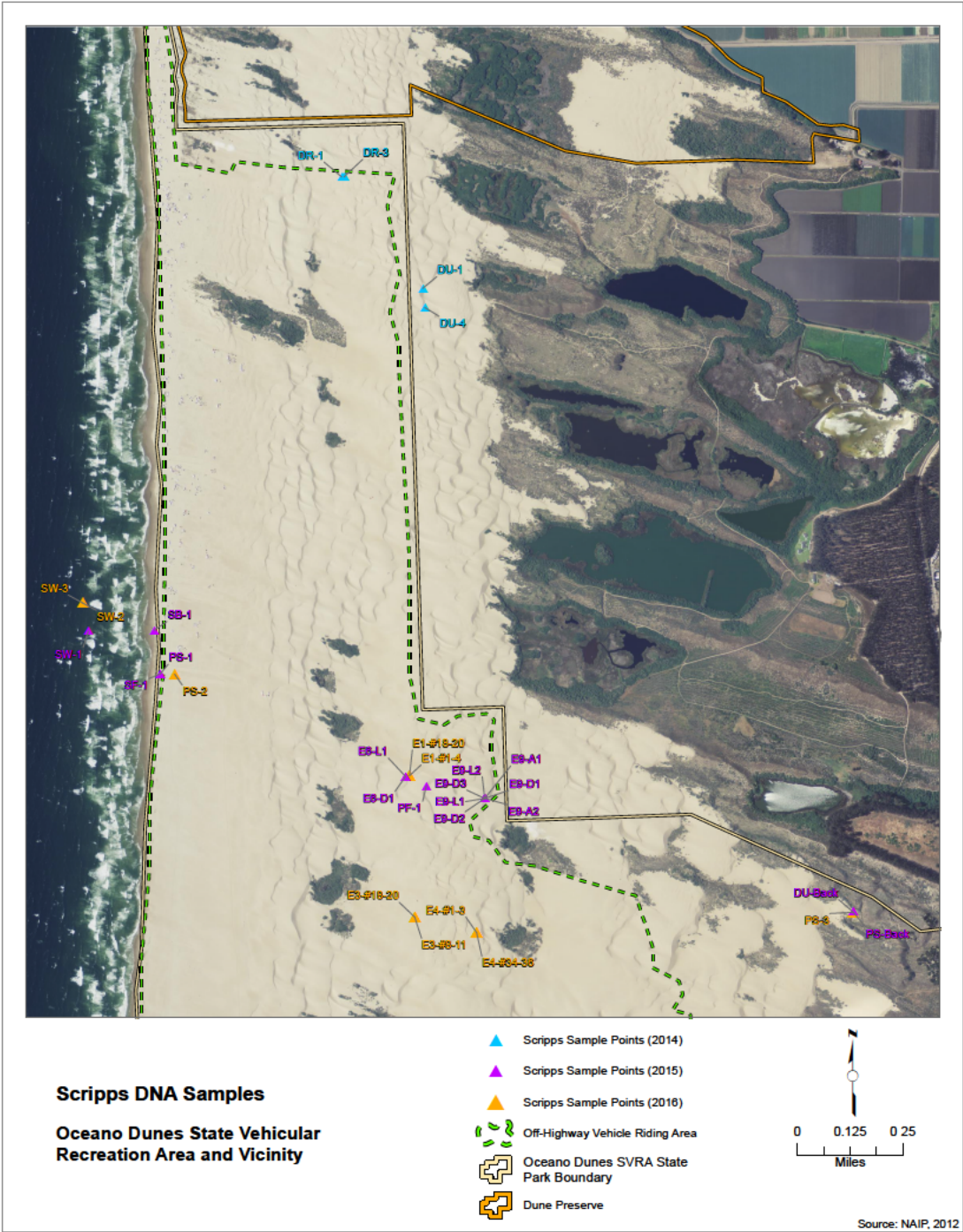


Figure 1. Map of Oceano Dunes sampling sites.

Sample Name	Sample Type	Date	Marine Fraction of Eukaryotic OTUs, all seqs, SW only	Marine Fraction of Eukaryotic OTUs, singlets removed, SW only	Marine Fraction of Eukaryotic OTUs, all seqs, SW + DB	Marine Fraction of Bacterial OTUs, all seqs, SW only	Marine Fraction of Bacterial OTUs, singlets removed, SW
DU-1	Dune	7/26/14	0.00	0.00	0.00	0.04	0.04
DU-4	Dune	7/26/14	0.00	0.00	0.00	0.00	0.00
DR-1	Dune	7/26/14	0.00	0.00	0.00	0.00	0.00
DR-3	Dune	7/26/14	0.00	0.00	0.00	0.03	0.02
SW-1	Seawater	4/7/15	1.00	1.00	1.00	1.00	1.00
SB-1	Beach Sand	4/7/15	0.37	0.37	0.37	0.57	0.49
<b>E6-11</b>	<b>Air</b>	<b>2015</b>	<b>0.03</b>	<b>0.03</b>	<b>0.03</b>	<b>0.45</b>	<b>0.44</b>
<b>E6-D1</b>	<b>Air</b>	<b>2015</b>	<b>0.55</b>	<b>0.54</b>	<b>0.55</b>	<b>0.05</b>	<b>0.05</b>
<b>E9-A1</b>	<b>Air</b>	<b>2015</b>	<b>0.19</b>	<b>0.19</b>	<b>0.19</b>	<b>0.20</b>	<b>0.18</b>
<b>E9-A2</b>	<b>Air</b>	<b>2015</b>	<b>0.71</b>	<b>0.34</b>	<b>0.71</b>	<b>0.45</b>	<b>0.41</b>
<b>E9-L1</b>	<b>Air</b>	<b>2015</b>	<b>0.27</b>	<b>0.25</b>	<b>0.30</b>	<b>0.13</b>	<b>0.13</b>
<b>E9-L2</b>	<b>Air</b>	<b>2015</b>	<b>0.17</b>	<b>0.17</b>	<b>0.47</b>	<b>0.12</b>	<b>0.09</b>
<b>E9-D1</b>	<b>Air</b>	<b>2015</b>	<b>0.26</b>	<b>0.22</b>	<b>0.32</b>	<b>0.15</b>	<b>0.14</b>
<b>E9-D2</b>	<b>Air</b>	<b>2015</b>	<b>0.00</b>	<b>0.00</b>	<b>0.20</b>	<b>0.06</b>	<b>0.05</b>
<b>E9-D3</b>	<b>Air</b>	<b>2015</b>	<b>0.01</b>	<b>0.01</b>	<b>0.01</b>	<b>0.07</b>	<b>0.06</b>
PF-1	Fencing	4/7/15	0.76	0.74	0.76	0.68	0.63
DU-Back	Dune	4/7/15	0.00	0.00	0.00	0.01	0.01
PS-Back	Fencing	4/7/15	0.38	0.37	0.38	0.75	0.73
SF-1	Foam	4/7/15	0.92	0.91	0.92	0.90	0.88
PS-1	Fencing	4/7/15	0.00	0.00	0.00	ND	ND
SW-2	Seawater	6/20/16	1.00	1.00	1.00	1.00	1.00
SW-3	Seawater	6/20/16	1.00	1.00	1.00	1.00	1.00
PS-2	Fencing	6/20/16	0.19	0.19	0.68	0.15	0.08
PS-3	Fencing	6/20/16	0.08	0.08	0.13	0.10	0.09
<b>E1-#1-4</b>	<b>Air</b>	<b>6/16/16</b>	<b>0.00</b>	<b>0.00</b>	<b>0.72</b>	<b>0.10</b>	<b>0.06</b>
<b>E1-#18-20</b>	<b>Air</b>	<b>6/16/16</b>	<b>0.00</b>	<b>0.00</b>	<b>0.91</b>	<b>0.08</b>	<b>0.06</b>
<b>E3-#8-11</b>	<b>Air</b>	<b>6/16/16</b>	<b>0.00</b>	<b>0.00</b>	<b>0.96</b>	<b>0.35</b>	<b>0.15</b>
<b>E3-#18-20</b>	<b>Air</b>	<b>6/16/16</b>	<b>0.00</b>	<b>0.00</b>	<b>0.99</b>	<b>0.03</b>	<b>0.00</b>
<b>E4-#1-3</b>	<b>Air</b>	<b>6/13/16</b>	<b>0.00</b>	<b>0.00</b>	<b>0.95</b>	<b>0.12</b>	<b>0.04</b>
<b>E4-#34-36</b>	<b>Air</b>	<b>6/13/16</b>	<b>0.00</b>	<b>0.00</b>	<b>0.61</b>	<b>0.11</b>	<b>0.00</b>
<b>E5-#1-3-Mesa2</b>	<b>Air</b>	<b>6/13/16</b>	<b>0.00</b>	<b>0.00</b>	<b>0.69</b>	<b>0.06</b>	<b>0.03</b>
<b>E5-#34-36</b>	<b>Air</b>	<b>6/13/16</b>	<b>0.00</b>	<b>0.00</b>	<b>0.84</b>	<b>0.10</b>	<b>0.07</b>

Table 1. Sample descriptions and fraction (of 1) of total DNA sequence reads from each sample that were likely of marine origin, based on comparison to either only the seawater OTUs (“SW only”) or those and database-derived sequences (“SW + DB”). “Singlets removed” refers to the same sample but only including sequences present two or more times. ND, not determined. The data shows that marine microbes make substantial contributions to EBAMs, as seen in the DNA recovery from EBAM filters (bolded).





O	U	taxonomy	SUM OF SW READS	SUM OF EBAM READS	SUM OF DUNE READS	Likely marine	Land Plant	Fungi	Unknown	opblast hit	%ident	opblasthit	
New.ReferenceO	U163	Diatomeal(100);Bacillariophyta(100);	0	339703	0	yes				KY852256.1	100	Chaetoceros calcitrans strain CCMP1315	
New.ReferenceO	U26	EDiatomeal(100);Bacillariophyta(100);	0	77758	0	yes				KU900218.1	100	Thalassiosira pseudonana strain CCAP 1085/12	
New.ReferenceO	U129	Dilayal(100);Ascomycota(100);	0	36026	20		yes*			KR336841.1	100	Candida austromarina	
New.ReferenceO	U1	Eumetazoa(43);Bilateria(43);	0	22363	0			yes		F 459746.1	90 *	Gregarina kingi	
New.ReferenceO	U108	Diatomeal(100) Bacillariophyta(100)	0	8376	0	yes				KY852256.1	95	Chaetoceros calcitrans strain CCMP1315	
New.ReferenceO	U91	Pragmoplistophyta(100);Streptophyta(100);	0	7114	42		yes			A 272181.1	99	Triticum aestivum	
New.ReferenceO	U117	Dilayal(95);Basidiomycota(95);	0	6724	0		yes			L 603011.1	97	Hyphodroma orphanellum	
New.ReferenceO	U79	Phragmoplistophyta(100) Streptophyta(100)	0	6197	0		yes			K 459714.1	98	Vicia villosa (Vetch)	
New.ReferenceO	U28	Eumetazoa(94);Bilateria(94);	0	5930	0			yes		F 808261.1	93	Nepalomyia sp. 3 SNK-2009	
New.ReferenceO	U24	Dilayal(77);Basidiomycota(77);	0	5944	0		yes			MF112031.1	99	Sporobolomyces roseus	
New.ReferenceO	U158	unclassified;unclassified;	0	4951	0			yes		O245081.1	95	Centroheliozoa sp. ATCC 50535	
New.ReferenceO	U148	Diatomeal(16);ME Euk FW10(16);	0	3804	0	yes				KY054619.1	100	Uncultured eukaryote clone E035	
New.ReferenceO	U99	Dinophyceae(51);SGYB05(22);	0	3743	0	yes				AB686255.1	95	Lepidodinium sp. MH 360	
New.ReferenceO	U58	Chrysothecae(27);P.34.45(20);	0	3426	0	yes				L31799.1	85 *	Gregarina caledia	
New.ReferenceO	U140	Eumetazoa(96) Bilateria(96)	1	3316	0	yes				GU175705.2	100	Dolioletta geogenbaui (marine tunicate)	
New.ReferenceO	U176	Diatomeal(100);Bacillariophyta(100);	155	3231	0	yes				DQ514870.1	98	Thalassiosira oestrupii var. venickae strain CC03-15	
New.ReferenceO	U65	Dilayal(100);Ascomycota(100);	0	3027	0		yes			KU058168.1	100	Saccharomyces cerevisiae strain JCBASC23	
New.ReferenceO	U75	Diatomeal(100);ME Euk FW10(64);	65	2979	0	yes				KX229690.1	99	Pseudo-nitzschia fraudulenta strain UNC1413	
New.ReferenceO	U20	Diatomeal(97);Bacillariophyta(47);	0	2912	0	yes				FM877466.1	99	Diatom endosymbiont ex foraminifera MH-2008	
New.ReferenceO	U110	Dilayal(98);Basidiomycota(98);	0	2800	0	yes				L 603002.1	99	Hyphodontia rimosissima	
New.ReferenceO	U96	Rhodymeniophyceae(27);Melantherial(10);	0	2754	0		yes			F 459752.1	80 *	Leidyana erratica	
New.ReferenceO	U40	Dilayal(100) Basidiomycota(100)	0	2566	0		yes			AY336784.1	100	Antridia sitchensis strain HNB 5298SP	
New.CleanUp.ReferenceO	U2271	Conidiasida(94);Gregarinasina(77);	0	2561	0		yes			F 459755.1	99	Paraschmeidia metamorphosa	
New.ReferenceO	U145	Eumetazoa(100);Bilateria(100);	0	2287	0		yes			K 946005.1	100	Helicoverpa zea (moth)	
New.ReferenceO	U151	Colpodellida(23) Voromonasi(22)	0	1956	0		yes			A 697751.1	93	Cryptosporidium struthionis	

\*marine Fungi

Table 3. Top 25 most abundant eukaryotic OTUs found in EBAM samples. Bold shows taxa predicted by presence in seawater sample. Other marine taxa were found by taxonomic knowledge and BLAST annotation. *Triticum aestivum*, common wheat, may be coming from cereal straw bales used for erosion control.

OTU	Taxonomy	SUM O SW READS	SUM O EBAM READS	SUM O DUNE READS	Top Bas Accession	% de	Name
New Refe e ceOTU 07	Eume azoa( 00);B a e a( 00);	0	0	24638	K_32504	97	M c opsammus sp AR -20 5 (m e)
New Refe e ceOTU49	u c ass fed;u c ass fed;	0	0	0750	JQ24508	98	Ce o e ozoa sp ATCC50535
New Refe e ceOTU22	agmpas op y a( 00);S ep op y a( 00);	0	0	6580	KT_79677	98	He a us pauc fo us subsp sub ombo deus
New Refe e ceOTU5	T eboux op yceae( 00);T eboux a es(90);	0	5	6450	KM05629	99	T eboux a sp - 42 (m c oa ga)
New Refe e ceOTU86	T ecof oseal( 00);C yomo ad da( 00);	0	0	5667	KY905096	95	R ogos oma cy d ca so a eKD 020
New Refe e ceOTU 53	T eboux op yceae(93);C o e a es(49);	0	0	3300	JX_69829	99	Ap a ococcus oba us s a SAG 2 5 (m c oa ga)
New Refe e ceOTU55	C y d omyco a(99); ce ae Sed s(99);	0	0	2682	AH009023.2	98	Ge a omyces va ab s s a BK9 -
New Refe e ceOTU 62	C y sop yceae(24); ce ae Sed s( 9);	0	0	22	KU587705	98	Cyp obas d um ypogym co a vouc e S- 26467
New Refe e ceOTU 23	agmpas op y a( 00);S ep op y a( 00);	0	0	634	A_05_798	99	use o (sas p e)
New Refe e ceOTU 25	Eume azoa(92);B a e a(92);	0	0	495	KJ636268	99	Me o dogy e apa s a Me oHap ( ema ode)

Table 4. The most abundant eukaryotic OTUs in four dune sand samples. These OTUs were not typically found on EBAMs. A few OTUs such as the microalgae appear to be microbial residents in the sand while others are from nearby plants.