# SHORT COMMUNICATION

# DEPURATION OF MACONDO (MC—252) OIL FOUND IN HETEROTROPHIC SCLERACTINIAN CORALS (TUBASTREA COCCINEA AND TUBASTREA MICRANTHUS) ON OFFSHORE OIL/GAS PLATFORMS IN THE GULF

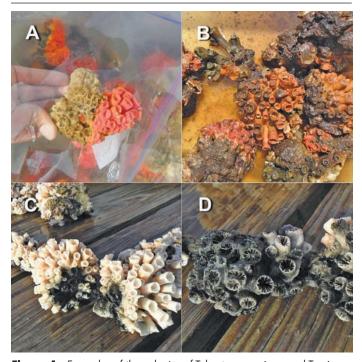
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## INTRODUCTION

A cluster of offshore platforms (90, 93, and 94) in the Gulf of Mexico (GOM) located within the Grand Isle block (hereafter, called GI 90 block platforms) possess a diverse invertebrate community, including two species of hard corals. Our dive team conducted more than 30 underwater surveys to observe and collect corals from the GI 90 block oil and gas platforms between 10 July 2005 and 21 October 2010 and found only healthy colonies of corals (Sammarco et al. 2009, 2010, 2012).

The BP/Deepwater Horizon MC-252 well blowout oc-



**Figure 1.** Examples of the colonies of Tubastrea coccinea and T. micranthus collected from the GI 90 block platforms. **A.** One half of this colony of T. coccinea was dead, as indicated by the brownish/black color and loss of tissue, when collected from GI 90 block on 8 May 2011. **B.** T. coccinea and T. micranthus colonies that showed visible signs of contamination. **C.** and **D.** T. coccinea colonies that showed visible signs of contamination.

curred on 20 April 2010 and lasted 84 d. It was the second largest marine spill in recorded history (Joye et al. 2011). It leaked 7.0 x  $10^5$  m<sup>3</sup> of crude oil from the sea floor into the northern GOM (Crone and Tolstoy 2010), but was reported to have been capped on 15 July 2010. The GI 90 block platforms are located 168 km WSW of BP's MC–252 spill site, and 60 km south of Port Fourchon, LA.

The impetus for this study began on 8 May 2011, when we noted sporadic mortality of colonies of orange-cup corals, Tubastrea coccinea (Lesson, 1829), and green sun corals, Tubastrea micranthus (Ehrenberg, 1834; Cnidaria, Anthozoa, Scleractinia) among colonies of coral that appeared to be in good health at the GI 90 block platforms. Colonies of T. coccinea which appeared to be healthy displayed markedly less tissue regression and were bright orange in color. The morbid polyps were dark brown or black (Figure 1A–D). The corals were collected, transported to the laboratory, and segregated; healthy-appearing corals were placed in guarantine tanks. After 12 h in guarantine tanks, the coral exudate turned the tank water black and the colonies died within 48 h. After observing this unusual behavior, we decided to document subsequent collection events and analyze the discharged oil to determine its source.

# Materials and Methods

#### Survey Sites

The GI 90 block platforms (28°32'37.61"N, -90°03' 56.06"W) are located in 64 m of water. Corals were collected on 11 June and 4 August 2011. The upper 20 m of the 64 m tall sub–surface rig support structures were surveyed via SCUBA and coral samples were scraped from the pilings at 12 and 20 m depths. The survey area was ~6,000 m<sup>2</sup>. Upon returning to the research vessel, the coral samples were placed in travel containers with seawater (salinity of 30), kept at 21°C, and transported to shore.



**Figure 2.** Oil slicks observed over a 10 km wide area during a sampling trip to GI 90 block platforms on 14 September 2011, 13 mo after the BP well had been capped. The foamy beige surface oil was similar in appearance to the oil discharge from coral skeletons (see Figure 4C).

We returned to the GI 90 block platforms on 14 September 2011 for additional coral collections; however, the collection trip was cancelled due to the presence of large crude—oil slicks near the GI 90 block (Figure 2).

#### **Aquarium Studies**

#### 11 June 2011 Analysis

Corals were examined and the specimens that appeared to be contaminated with oil were excluded from further observation. About 40 colonies of *T. coccinea* and 10 colonies of *T. micranthus* were placed in the quarantine tanks. The stocking density was about one coral colony per 35 L of seawater and the colonies were maintained in the quarantine tanks for 8 wk. The tank water was changed with GOM seawater (salinity 30) 3 times the first day, twice daily for the following week, and then once daily for the next 4 wk. Water changes were reduced to once weekly from weeks 5 through 8.

#### 4 August 2011 Analysis

Colonies of coral were examined and any that appeared to be contaminated (possessing dead black or brown polyps) were excluded from further observation. About 20 colonies of *T. coccinea* were placed in 57 L quarantine tanks. The stocking density was about one colony per 35 L of seawater. Routine water changes of  $\geq$ 50% of the tank's volume were performed after 2, 8, 16, and 48 h and on day 5. The salinity of the tank water was maintained at 30.

After day 5, the water within the aquaria was filtered using mechanical filter floss and a 0.25 x 0.25 m hydrocarbon adsorbent cloth (Dynamic Adsorbents<sup>®</sup>). The filter was wrapped in aluminum foil, sealed in a plastic bag, and stored at  $-20^{\circ}$ C to preserve the sample for future analysis.

A sample of the filtered tank water was sent to an analytical chemistry laboratory specializing in crude—oil and related substances (ALS Environmental, Edmonton, Canada) for determination of biomarker profiles. Biomarkers

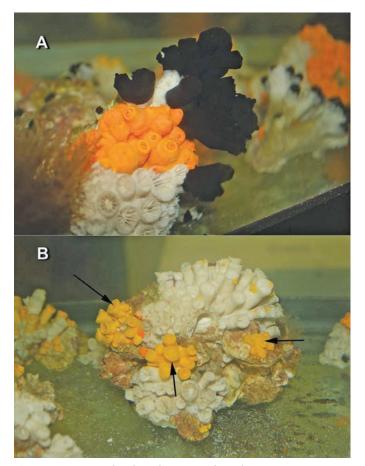
are hydrocarbon molecules derived from formerly living organisms and are present in crude oils at low concentrations (<100 ppm; Wang et al. 2007). The sample biomarker chromotagrams were matched with biomarkers from a BP MC–252 pipeline riser sample. Environmental applications of biomarker fingerprinting have been extensively reviewed elsewhere (Peters and Moldowan 1993; Peters et al. 2005a, 2005b; Wang et al. 2006). The methodology is described in Wang et al. (2007) and Hansen et al. (2007), and has been revised by the European Committee for Standardization (ECS) (2012). The sample was not analyzed for the presence of dispersants.

On day 7, all surviving corals were sacrificed by placing them in fresh water. Three of the colonies were removed from the saltwater quarantine tanks and placed in a 113 L container of fresh water for 28 d. The purpose of the freshwater soaking was to determine if hydrocarbon substances would continue to exude from the deteriorating coral tissues. All photographs were recorded using Sony FX High— Definition video camera and an 18 megapixel Cannon Rebel digital camera.

#### RESULTS

# 11 June 2011

Of the 125 coral colonies collected from the platform pilings, 60% showed visible signs of polyp mortality and were excluded from further observation. When the healthy colonies were placed in the quarantine tanks, they released a cloudy white material for 14 h. Several hours later, the aquarium water turned brown and then black. After water changes, the water would change from light brown to black as the corals depurated oil. The corals appeared to purge most of the material within 7 d, but continued to display retracted polyps, exuded mucus, and showed signs of tissue loss for another 5 d. By the end of week 2, the water in



**Figure 3. A.** Coral polyps that survived 4 wk in quarantine: orange colony = Tubastrea coccinea; dark green colony = Tubastrea micranthus. **B.** "Budding" yellow polyps (arrows), evidence of asexual reproduction, found growing on 10% of the colonies by week 6. Previously, during weeks 1 and 2, the corals had sloughed off muscle tissue from their stomach and calyx, and the skeletons were bleached.

the tanks remained clear overnight and the surviving corals began to extend their tentacles and started feeding on shredded shrimp.

By day 14, 80% of the coral polyps sloughed off injured tissue and transformed into bleached skeletons. The remaining 20% maintained their orange tissue (*T. coccinea*) or dark green tissue (*T. micranthus*) and regained their vigor. After 4 wk in the quarantine tanks, the surviving corals were feeding regularly and exhibiting normal color and behavior (Figure 3A).

By week 6, evidence of asexual reproduction could be seen in *T. coccinea* on the coral skeletons. The yellow coral polyps (Figure 3B) exhibited "budding" or asexual reproduction on the skeletons of recently morbid coral.

4 August 2011

Of the 80 coral colonies transported to the laboratory, 70% showed visible signs of polyp mortality. The remaining 30% that appeared healthy were placed in quarantine tanks. The water turned black after 9 h in the aquaria and corals continued to discharge oil after each water change for 7 d. The black oily water in quarantine tanks (Figure 4A) was present 48 h after the corals were placed in the aquaria. The foamy surface oil (Figure 4B) continued to appear after 5 d of water changes.

After 7 d in quarantine, 90% of the corals showed signs of substantial tissue regression (>50%), and the cultivation was then terminated for study purposes. Several of the largest colonies of *T. coccinea* were placed in fresh water for 28 d where they continued to discharge crude oil and slough off tissue postmortem (Figure 4C).

Chemical analysis of the hydrocarbon exudate from the corals matched crude oil signatures characteristic of oil from the MC–252 field. ALS Laboratories performed a critical–difference analysis of the calculated ratios of biomarker which revealed that 7 out of 8 ratios obtained from the coral samples matched those obtained for MC–252 crude oil. This suggests, from a weight–of–evidence approach, that the crude oil found in the water of the coral quarantine tanks was a positive match with the crude oil from the MC–252 well.

# DISCUSSION

Coral samples have been collected at the GI 90 block platforms since 10 July 2005, including eight occasions between 14 June and 21 October 2010, during and shortly after the MC–252 blowout period. Prior to 8 May 2011, we



**Figure 4.** Examples of quarantine aquaria. **A.** The oil ingested by the corals turned the water black after 48 h (10 August 2011). **B.** Day 5 in quarantine tanks (13 August 2011), the corals continued to purge oil. Brownish black foam appeared on the surface. **C.** The dead coral continued to leach beige foamy crude oil after 28 d in fresh water (8 September 2011). The surface oil in the container is similar to crude oil slicks observed offshore (Figure 2).

found no visual evidence of oil discharge in tank water or high polyp mortality among corals during the previous 7 yr of coral collection for research, including during and after the BP oil–well blowout. In the past, we noted that colonies of *T. coccinea* occasionally discharged orange materials during and shortly after transport from the platform to the laboratory, likely from the coelom or muscle tissue. *Tubastrea* spp. are known to respond to stress by producing tubastrine, a mucus material that protects the colony from biological and viral threats (Meyer et al. 2009). Prior to 8 May 2011, the corals in quarantine tanks occasionally discharged orange materials, requiring one water change to clear from the tanks; however, the oil discharges in June and August of 2011 were greater and lasted longer.

Coral behaviors during the June and August 2011 observation periods were similar in that the coral discharged oil for 7 d; however, the first group of corals discharged a white milky material for the first 14 h whereas the second group of corals initially discharged oil that turned the water in the tank black within 9 h. The second observation period was shorter due to the high rate of mortality (90%) of coral polyps. During the 4 August 2011 quarantine period, concentrations of oil were noticeably different from tank to tank; 2 of the 57 L tanks were substantially darker than other tanks.

In addition to scleractinian corals, gorgonians, sponges, tunicates, barnacles, bryozoans, hydrozoans, and oysters in-

habit the offshore platforms (Gallaway et al. 1981, Lewbel et al. 1987, Dokken et al. 2000, Carney 2005, Rouse 2009). White et al. (2011) investigated deepwater coral reef habitats (1,370 m) and found crude oil impacts on scleractinian, gorgonian, and antipatharian corals 11 km SW of BP's MC–252 well 3 mo after its reported capping. They describe the appearance of a "brown flocculent material" on the surface of the invertebrates which resemble the materials on the corals found here (see Figure 1B). The coral samples (n = 5) were subjected to chemical analysis and biomarkers were found to match MC–252 oil (White et al. 2012).

The heterotrophic coral on the GI 90 block platforms appear to be assimilating subsurface crude oil. About 3,500 major structures are scattered across the northern GOM (BOEM 2012) and we suggest that some heterotrophic and filter—feeding organisms on the pilings of offshore platforms can be used to determine the source (Wang et al. 2006; Hansen et al. 2007) as well as the horizontal and vertical distributions of BP's MC–252 oil spill. Therefore, by collecting corals from platforms in other areas of the GOM and analyzing the organisms for hydrocarbons, one might be able to determine the geographic range of the MC–252 crude oil plume. The combination of these data could provide geographic insight into the BP MC–252 spill and assist in determining recent and historical exposure to subsurface crude oil.

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