Original Article

Persistent Impacts to the Deep Soft-Bottom Benthos One Year After the Deepwater Horizon Event

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ABSTRACT

In fall 2010, several months after the Deepwater Horizon blowout was capped, zones of moderate and severe impacts to deep-sea,

soft-bottom benthos were identified that together extended over an area of 172 km^2. A subset of stations sampled in 2010 was resampled in May and June 2011, 10 to 11 months after the event, to determine whether the identified adverse effects were persisting. The design compared 20 stations from the combined moderate and severe impact zone to 12 stations in the reference zone that were sampled in both years. There were no statistically significant differences in contaminant concentrations between the impact and nonimpact zones from 2010 to 2011, which indicates contaminants persisted after 1 y. Whereas there were some signs of recovery in 2011 (particularly for the meiofauna abundance and diversity), there was evidence of persistent, statistically significant impacts to both macrofauna and meiofauna community structure. Macrofaunal taxa richness and diversity in 2011 were still 22.8% and 35.9% less, respectively, in the entire impact zone than in the surrounding nonimpact area, and meiofaunal richness was 28.5% less in the entire impact zone than in the surrounding area. The persistence of significant biodiversity losses and community structure change nearly 1 y after the wellhead was capped indicates that full recovery had yet to have occurred in 2011.

Keywords: Deepwater Horizon oil spill, Deep-sea infaunal benthos, Benthic macrofauna and meiofauna, Sediment quality, Oil-spill impacts, Gulf of Mexico

INTRODUCTION

The Deepwater Horizon (DWH) blowout at the Macondo well drilling site occurred in the northern Gulf of Mexico on 20 April 2010. The spill, which occurred at a water depth of 1525 m, in Mississippi Canyon Block 252, released approximately 3.19 million barrels (507 million L) of oil over a 3-month period (DWH Natural Resource Trustees 2016). Ryerson et al. (2012) have estimated that a majority of the oil was removed by cleanup operations, other natural mechanisms, or was present at the surface in oil slicks, but up to 35% of the hydrocarbons were trapped and transported in persistent deep-sea plumes. The large volume of oil trapped in the deep-sea plumes indicates that large amounts of oil were likely transported to offshore, deep-water sediments. It is now known that this

transport could occur via several potential pathways, for example, sinking of dispersed oil droplets that were absorbed onto suspended particles, incorporation of oil droplets into copepod fecal pellets, transport of oil-laden particles, sinking of heavier residues that resulted from burning oil during the oil spill response activities, or settling of oil–mud complexes from the injection of drilling mud during the failed top-kill operations (UAC 2010). In addition, drill cuttings, drill fluids, and other containment fluids, commonly used during offshore oil-drilling operations (Neff et al. 1987, Neff 2005) were likely released from the well blowout and deposited to the bottom after the blowout.

In September and October 2010, after the Macondo wellhead was capped in July, 2 DWH response cruises (aboard *RV Gyre* and *RV Ocean Veritas*) were deployed to sample sediments for the purpose of determining whether substantial amounts of oil were on the deep-sea floor. Initial results from analysis of 65 deep-water (>200 m) stations sampled in 2010 demonstrated significant impacts to the macrofauna and meiofauna benthos based on a spatial interpolation of a principal components analysis (PCA) of combined benthic and abiotic variables (Montagna, Baguley, Cooksey, Hartwell et al. 2013). The PCA produces a first principal component (PC1) that accounts for the largest possible variance in the data attributable to oil-spill impacts. The new PC1 variable was mapped in a geographic information system (GIS) to identify the footprint of DWH-related benthic impacts. The most severe reductions in macro- and meiofaunal abundance and diversity occurred within 3 km of the wellhead and covered an area approximately 24 km^2. Moderate impacts were observed in an area of 148 km^2 that had elliptical shape, which extended up to 17 km toward the southwest and 8.5 km toward the northeast of the wellhead. Thus, the total area that was adversely impacted covered 172 km^2. Adverse benthic effects were strongly correlated with concentrations of total petroleum hydrocarbons (TPHs), PAHs, and Ba, as well as distance to the wellhead, but there were no correlations with distance to hydrocarbon seeps. Thus, the observed correlations of biological effects were most likely due to the oil spill and not to natural hydrocarbon seepage.

A study was initiated in May 2011 to determine the effects of the DWH oil spill on sediments and benthic fauna in the deep sea of the Gulf of Mexico (Montagna, Baguley, Cooksey, Hyland 2013). This study was performed under the direction of the DWH Natural Resource Damage Assessment (NRDA) Deepwater Benthic Communities Technical Working Group (NRDA Deep Benthic TWG), which was composed of scientists, managers, and representatives from the trustees and British Petroleum (BP). The purpose of the study was to assess potential spatial and temporal impacts of the DWH oil spill on sediments and benthic fauna in deep-water areas of the Gulf. One objective of this NRDA study was to resample 32 stations that were sampled in 2010 on the 2 response cruises. In the present article, results from the NRDA field sampling effort in May and June 2011 are presented and statistically compared to 2010 data to test for persistence of impacts or alternatively for any signs of recovery from the oil spill.

METHODS

A cruise was conducted 23 May–11 June 2011 aboard the *M/V Sarah Bordelon*. A total of 38 stations were sampled; 32 coincided with stations that were sampled in fall 2010 (Figure 1A and B). An OISL mega-multicorer was used to collect 12 sediment core samples with each deployment. Cores from each drop were apportioned for analysis of macrofauna, meiofauna, hydrocarbons, metals, porewater chemistry (oxidation potential [Eh], sulfides, ammonia), and other basic sediment properties (total C, TOC, total inorganic C, total N, grain size). The details of the methods are described in Montagna, Baguley, Cooksey, Hartwell et al. (2013) and Montagna, Baguley, Cooksey, and Hyland (2013).

The experimental approach was to test for significance of mean differences in total community response between the 2 sampling years (fall 2010 and spring 2011). The stations were divided into 2 main DWH effects zones as defined by Montagna, Baguley, Cooksey, Hartwell et al. (2013): “impacted” (high to moderate impacts, the red- and orange-coded stations in Figures 2 and 3 of Montagna, Baguley, Cooksey, Hartwell et al. 2013) versus ”nonimpacted” (unlikely impacts, the light green- and green-coded stations). A 3rd zone (the yellow-coded stations), which is the border between moderate and unlikely impacts, but may have suffered impacts, was

included in the nonimpact zone for the purposes of the current analysis. For the present analysis, 20 impact stations were included (ALTNF001, ALTNF015, D031S, D034S, D038SW, D040S, D042S, D044S, D050S, FF010, LBNL1, LBNL14, LBNL3, LBNL7, NF006MOD, NF008, NF010, NF011, NF012, NF013) and 12 nonimpact stations were included (2.21, D002S, D019S, D024S, D043S, D062S, FF005, FFMT3, FFMT4, LBNL17, LBNL4, NF014).

Macrofauna were collected from 3 core samples from a single multicore drop at each station. Stations are thus nested within the zones, so the experimental design is a partially hierarchical, 2-way analysis of variance (ANOVA) that can be described by the following statistical model,

$$Y_{ijkl} = \mu + \alpha_j + \beta_k + \alpha\beta_{jk} + \gamma_{k(l)} + \alpha\beta\gamma_{jk(l)} + e_{(i)jkl}\ , \quad (1)$$

where Y_{ijkl} is the dependent response variable; μ is the overall sample mean; α_j is the main fixed effect for year, where j = 1 or 2 for either 2010 or 2011; β_k is the main fixed effect for sampling zone, where k = 1 or 2 for either the impact or nonimpact zone; $\alpha\beta_{jk}$ is the main fixed effect for the interaction between year and zone; $\gamma_{k(l)}$ is the main effect for stations that are nested (or unique) to the zones and are thus a random effect as denoted by the parentheses around the subscript l, which represents the 32 stations, all of which are nested unique to 1 of the 2 zones; $\alpha\beta\gamma_{jk(l)}$ is the interaction term for year–zone–station cells; and $e_{(i)jkl}$ is the random error term for each of the i replicate measurements within cells.

For meiofauna, only 1 core sample was collected from each single multicorer drop at each station. Because only 10 of 12 cores from each multicorer drop were usually successful, there were not enough cores available from each drop to collect more than 1 meiofauna sample in addition to remaining cores allocated for analysis of macrofauna, chemistry, and sediment properties. Also, our rationale for replicating macrofaunal and not meiofaunal samples was that smaller-sized meiofauna were assumed to vary less on the spatial scales of the size of the coring device than were larger-sized macrofauna. Because there are no replicate cores within multicorer drops, the

triple-interaction term does not exist, thus the model is reduced to

$$Y_{ijk} = \mu + \alpha_j + \beta_k + \alpha\beta_{jk} + \gamma_{k(l)} + e_{(i)jkl}. \quad (2)$$

Macrofauna were identified to the family level, and meiofauna were identified to the major taxa level (order or higher). Macrofaunal community structure was analyzed using nonmetric multidimensional scaling (MDS) by first creating a Bray-Curtis similarity matrix among stations and then an MDS plot (Clarke 1993; Clarke and Warwick 2001). Differences in community structure between years and zones were tested using analysis of similarities (ANOSIM) and SIMPER in Primer software (Clarke 1993). Data were $\log_e(x + 1)$ transformed prior to multivariate analysis in Primer to decrease the effect of numerically dominant species on the community composition, as presented in the MDS bivariate plots (Clarke and Gorley 2006).

Benthic response variables presented here include total faunal abundance, number of taxa, Hill's N1 diversity (Hill 1973), and nematode to copepod ratios (N:C). Hill's N1 is the exponentiated form of the Shannon-Weiner H' diversity index, $N1 = e^{H'}$, and was selected because it is easily interpreted as the effective number of dominant species (i.e., taxa for the present study) and N1 trends to 1 as diversity decreases. The N:C ratio first proposed by Raffaelli and Mason (1981) has been widely used as an indicator of pollution exposure, and it is sensitive to hydrocarbon exposure (Peterson et al. 1996; Montagna et al. 2013). Abiotic environmental variables consist of TPHs, total PAHs, Ba, and selected natural habitat characteristics (depth, sediment grain size, and TOC).

All ANOVA tests were performed using SAS 9.3 software. PROC MIXED was used for the ANOVA, and the statistical model was implemented with the following SAS code: Y = Year Zone Year*Zone Station(Zone) Year*Station(Zone) for macrofauna. Because there were no replicate cores for meiofauna or for sediment chemistry, the model was reduced to: Y = Year Zone Year*Zone Station(Zone). The RANDOM statement was used to declare Station(Zone) and Year*Station(Zone) random effects and calculate the expected mean squares (EMS) to construct appropriate *F*-tests. For macrofauna, the appropriate error term for the *F*-tests for the Year

and Year*Zone terms is the Year*Station(Zone) interaction term, and the appropriate error term for the Zone test is the Station(Zone) interaction term. For chemistry and meiofauna, the appropriate error term for the *F*-tests for the Year and Year*Zone terms is the residual error term, and the appropriate error term for the Zone test is the Station(Zone) interaction term.

RESULTS

There were no statistically significant differences in contaminant concentrations in the defined impact zone from 2010 to 2011 (Table 1, Figure 1). One station (D034S within 3 km of the wellhead) had a large increase in hydrocarbons (261-fold for total PAHs and 11-fold for TPHs) from low background levels in 2010 to levels equivalent to high impact sites in 2011. D034S was originally classified as a "yellow" station in 2010 with uncertain impacts, but would be classified as a "red" station in 2011 (Montagna, Baguley, Cooksey, Hartwell et al. 2013), so station D034S was reclassified as an impact station for analyses performed here (Table 1).

All 3 macrofaunal metrics ─ abundance (n), number of taxa (S), and Hill's diversity (N1) ─ increased from 2010 to 2011 within both the impact and nonimpact zones (Table 2A). However, the pattern of lower number of taxa and lower diversity in the impact zone compared to the nonimpact zone in 2010 persisted in 2011 (Figure 3), with results of ANOVA revealing significant between-zone differences in these metrics at $\alpha < 0.05$, and insignificant zone–year interaction terms (Table 2B). On average over both years, macrofaunal richness and diversity were 27% and 36% less in the impact zone than in the surrounding nonimpact zone. Whereas macrofauna abundance increased in the impact zone by 69% from 2010 to 2011, it increased only 4% in the nonimpact zone (Table 2A). Although this is an apparent interaction (Figure 2A), the year–zone interaction term was only marginally nonsignificant ($P = 0.0574$, Table 2B). Yet the large (69%) increase in macrofaunal abundance from 2010 to 2011 within the impact zone was notable (Table 2A). The highest abundance was 24 208/m^2 at station DO31S in 2011, but only 15 167/m^2 at station LBNL17 in 2010. Stations with the highest densities in 2011, in excess of the mean of 11 533/m^2 for both zones combined, are mostly from the impact zone (10 from impact sites, 5 from nonimpact sites).

Evidence of impacts to the meiobenthos in 2010 included higher total abundances, higher N:C, lower number of taxa, and lower N1 diversity within the impact zone compared to the nonimpact zone (Table 3A). In 2011, the higher total abundances and lower number of taxa in the impact zone persisted (Table 3B, Figures 4A and B). However, there were also signs of recovery observed in 2011, as reflected in significant interactions between zones and years for N1 diversity and N:C ratios (Table 4B), due to converging values of these variables between the 2 zones in 2011 (Figures 4C and D). Although there was a 23% reduction of total meiofauna in the impact zone from 2010 to 2011, and a 3% increase in the nonimpact zone (Table 4A, Figure 4A), the interaction between year and zone was not significant ($P = 0.3998$), so we conclude that there is no statistically significant difference in total meiofaunal abundance between the 2 zones over both years overall ($P = 0.0882$). However, one must always be aware of the increased likelihood of making a Type II error (failing to reject a false null hypothesis) when the *P*-value is less than 0.1. The interaction for the N:C ratio is strong, however ($P = 0.0046$, Table 3B, Figure 4D). There was a significant reduction of N:C in the impact zone from 2010 to 2011, largely because of reduced numbers of nematodes. However, the pattern of reduced numbers of meiofaunal taxa in the impact zone compared to nonimpact zone in 2010 (i.e., 23% less) still persisted in 2011 (28% less) with significant differences between zones ($P < 0.0001$, Table 3B) and insignificant zone-year interaction term ($P = 0.1518$, Figure 4B). The persistent impact to meiofaunal richness translates to 26% fewer taxa in the impact zone than in the surrounding nonimpact zone over both years overall.

Results of MDS plots suggest there were similar patterns in community structure among nonimpact stations between 2010 and 2011 for both macrofauna (Figure 5) and meiofauna (Figure 6): For both macrofauna and meiofauna, the open symbols group together on the left side of the plots. Community structure for macrofauna and meiofauna was different between impact and nonimpact stations in both years, and 2-way crossed ANOSIM indicates that community structure was significantly different between zones across all year groups ($P = 0.001$ for macrofauna; $P = 0.002$ for meiofauna). However, community structure within the impact zone was different between the 2 years, and ANOSIM confirmed year differences across the zone groups ($P = 0.002$, macrofauna; $P = 0.005$, meiofauna). This was supported further by the MDS plots, where there was a difference between the impact and nonimpact zones indicated by

solid (impact) symbols aggregating on the left side of the MDS plot and open (nonimpact) symbols aggregating on the right side of the plot for both macrofauna (Figure 5) and meiofauna (Figure 6). There was also a shift in community structure for both macrofauna and meiofauna within the impact zone between the 2 years, indicated by the blue-solid (2010 impact) symbols aggregating on the far left of the MDS plot followed by green-solid (2011 impact) symbols shifting to the right of the MDS plot. Finally, the blue (2010) symbols are spread across the entire MDS plots, compared to the concentrated aggregation of green (2011) symbols, which indicates that community structure for both macrofauna and meiofauna was more dissimilar in 2010 than in 2011.

DISCUSSION

Persistence of impacts

Chemical contaminants persisted at similar concentrations 1 y after the spill (Table 1). Total PAHs and TPHs were 40 and 34 times higher, respectively, in the impact zone relative to the nonimpact zone in 2011. These results demonstrate the persistence of spill-related contaminants in sediments in 2011 following the DWH event. However, the dramatic increase in PAH and TPH concentrations at station D034S is evidence of the patchy nature of contamination on small spatial scales.

There were differences between the years, and in particular the overall abundance increased in both the nonimpact and the impact sites from 2010 to 2011. This is likely a simple year-to-year variation, but we do not have sufficient ancillary data to speculate the cause of this, other than that the 2010 samples were collected in the fall and the 2011 samples were collected in the spring. So it is possible that this is simply a seasonal difference rather than an interannual difference. The difference between years is why the interaction terms are important to detect impacts, and this is analogous to a before-after-control-impact study.

There was evidence of persistent impacts to both the macrofauna and meiofauna as a result of the DWH event. In 2011, macrofaunal

richness and diversity remained 23% and 36% less, respectively, in the impact zone than in the nonimpact zone, and meiofaunal diversity was the same but richness was 28% less in the impact zone than in the nonimpact zone. The pattern of higher meiofaunal densities in the impact zone compared to the nonimpact zone in 2010 persisted in 2011 — that is, 36% higher in 2010 and 14% higher in 2011, or 26% higher overall for both years combined (Table 3A) — providing further evidence of lingering effects. These effects are largely due to higher densities of pollution-tolerant nematodes (Giere 1979; Montagna and Harper 1996; Montagna, Baguley, Cooksey, Hartwell et al. 2013; Baguley et al. 2015) in the impact zone in both years, although their elevated densities had declined by 2011 and between-zone differences were not statistically significant. On average overall, polychaete families made up 84% (±9% SD) of the individuals at impact sites, and 71% (±7%) at the nonimpact sites. These percentages were identical in both 2010 and 2011. Peterson et al. (1996) proposed that higher percentages of polychaetes also indicate impacts near offshore oil and gas platforms. There also was a large, though statistically insignificant, increase in macrofaunal abundances within the impact zone from 2010 to 2011. The most abundant taxa at these sites in 2011, with densities in excess of 5000/m^2, were the polychaete families Dorvelleidae and Acrocirridae, which were also dominant at nonimpact sites. Members of Dorvelleidae (e.g., the genus *Dorvillea*) and Acrocirridae (e.g., *Acrocirrus*) are recognized as pollution-tolerant taxa or opportunistic colonizers of disturbed sediment (Borja et al. 2000; Gillett et al. 2015; also see Pearson and Rosenberg 1978 regarding *Dorvillea*). Two other dominant, but less abundant, macrofaunal groups within both zones were the polychaete families Capitellidae and Maldanidae. Most capitellid species are known to be pollution tolerant, though maldanids are generally regarded as being pollution sensitive (Borja et al. 2000; Gillett et al. 2015).

The multivariate analyses on community structure also revealed persistent impacts to macrofauna and meiofauna in 2011, because of the persistent dissimilarity between the impact and nonimpact zones. This dissimilarity is manifested as the preponderance of solid symbols (representing stations in the impact zone) in the left part of Figures 5 and 6, and a preponderance of open symbols (representing stations in the nonimpact zone) on the right. The persistence of these community structure effects nearly 1 y after the wellhead was capped indicates that full recovery had yet to occur as of 2011.

Meiofauna N1 diversity and N:C ratios showed some signs of recovery. Total meiofaunal density within the impact zone from 2010 to 2011 was reduced by 23%, consistent with a significant 77% reduction in N:C ratios in the impact zone. By 2011, meiofauna N1 diversity showed no significant difference between the impact and nonimpact zones (both zones had means of 1.86), indicating a recovery of this metric since 2010, when N1 diversity was 30% lower in the impact zone (Figure 4C).

Interpretations in relation to current knowledge of the deep-sea benthos

In general, benthic recovery depends on infaunal community recruitment and succession rates following a disturbance. This is especially true for a disturbance with lasting contamination of the substrate. The deep sea poses a special circumstance because recruitment and succession rates may be extremely slow. For example, in situ experiments indicate that in the deep sea, recolonization of clean, azoic sediments by macrofauna can take a year or longer (Grassle 1977). One potential mechanism for recovery in the impact zone will be degradation or burial of DWH-derived contaminants (Valentine et al. 2014). Recovery of soft-bottom benthos after previous shallow-water oil spills in the Bay of Morlaix has been documented to take years to decades (Boucher 1985; Dauvin 1998). Given that metabolic rates of the deep-sea benthos are very slow and turnover times are very long (Baguley et al. 2008; Rowe et al. 2008), it is possible that full recovery of the initial impacts in the vicinity of the DWH blowout may take decades or longer.

Significance of findings

Oil that ultimately makes its way to the seafloor can pose significant risks to benthic macrofauna and meiofauna living within or in close association with bottom substrates, because these organisms have relatively limited ranges and are sedentary (Giere 1979). Potential benthic infauna losses are of concern because these organisms serve vital functions in the deep-sea ecosystem, including sediment bioturbation and stabilization, organic matter decomposition, nutrient regeneration, secondary production, and energy flow

to higher trophic levels (Tenore 1977; Gray 1981; Gage 2003; Thistle 2003; Danovaro et al. 2008). The deep-sea benthos may also represent an important source of marine biodiversity (e.g., Hessler and Sanders 1967; Jumars 1976; Gage 1979; Hecker and Paul 1979; Rex 1981; Rowe et al. 1982; Grassle and Morse-Porteous 1987; Grassle and Maciolek 1992; Blake and Grassle 1994). Maximum benthic species diversity in ocean waters of the northeastern Gulf of Mexico occurs at mid- to upper-continental slope depths between 1200 and 1600 m (Tyler 2003; Wei and Rowe 2006; Rowe and Kennicutt II 2008, 2009; Haedrich et al. 2008; Wei et al. 2010), which coincides with the depths of the DWH wellhead and impact zone. Danovaro et al. (2008) provide evidence linking the loss of benthic biodiversity to an exponential decline in deep-sea ecosystem functioning.

Assessing impacts to the benthos is also important from a legal perspective. Under the Oil Pollution Act of 1990 (OPA90, 33 U.S.C.), federal and state trustees are given authority to assess damages to natural resources caused by an oil spill as part of the NRDA process. Quantifying the persistence of damages to these deep-benthic fauna, in addition to more familiar species such as seabirds and marine mammals, all of which are considered to be natural resources in the public trust, is an important step in the NRDA process and for understanding what is needed to achieve full ecological restoration and to ensure the long-term ecological health of the Gulf.

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REFERENCES

Blake JA, Grassle JF. 1994. Benthic community structure on the U.S. Atlantic slope off the Carolina: Spatial heterogeneity in a current-dominated system. *Deep-Sea Res II* 41:835–874.

Baguley JG, Montagna PA, Cooksey C, Hyland JL, Bang HW, Morrison C, Kamikawa A, Bennetts P, Saiyo G, Parsons E, Herdener M, Ricci M. 2015. Community response of deep-sea soft-sediment metazoan meiofauna to the Deepwater Horizon blowout and oil spill. *Mar Ecol Prog Ser* 528:127–140.

Baguley JG, Montagna PA, Hyde LJ, Rowe GT. 2008. Metazoan meiofauna biomass, grazing, and weight-dependent respiration in the Northern Gulf of Mexico deep sea. *Deep-Sea Res II* 55:2607–2616.

Borja A, Franco J, Pérez V. 2000. A marine biotic index to establish the ecological quality of soft-bottom benthos within European estuarine and coastal environments. *Mar Pollut Bull* 40:1100–1114.

Boucher G. 1985. Long term monitoring of meiofauna densities after the *Amoco Cadiz* oil spill. *Mar Pollut Bull 16*:328–333.

Clarke KR. 1993. Non-parametric multivariate analyses of changes in community structure. *Australian J Ecol* 18:117–143.

Clarke KR, Gorley RN. 2006. PRIMER v6: User Manual / Tutorial. Plymouth (UK): PRIMER-E.

Clarke KR, Warwick RM. 2001. Change in marine communities: An approach to statistical analysis and interpretation. 2nd ed. Plymouth (UK): PRIMER-E.

Danovaro R, Gambi C, Anno AD, Corinaldesi C, Fraschetti S, Vanreusel A, Vincx M, Gooday AJ. 2008. Exponential decline of deep-sea ecosystem functioning linked to benthic biodiversity loss. *Current Biol* 18:1–8.

Dauvin JC. 1998. The fine sand *Abra alba* community of the Bay of Morlaix twenty years after the Amoco Cadiz oil spill. *Mar Pollut Bull* 36:669–676.

DWH Natural Resource Trustees. 2016. Deepwater Horizon oil spill: Final programmatic damage assessment and restoration plan and final programmatic environmental impact statement. [cited 2016 Apr 5]. Available from: http://www.gulfspillrestoration.noaa.gov/wp-content/uploads/Chapter-2_Incident-Overview_508.pdf

Gage JD. 1979. Macrobenthic community structure in the Rockall Trough. *Ambio Special Report* 6:43–46.

Gage JD. 2003. Food inputs, utilization, carbon flow, and energetics. Chapter 11. In: Tyler PA, editor. Ecosystems of the deep. Amsterdam (NL): Elsevier. p 313–380.

Giere O. 1979. The impact of oil pollution on intertidal meiofauna. Field studies after the La Coruña-spill, May 1976. *Cahiers de Biologie Marine* 20:231–251.

Gillett DJ, Weisberg SB, Grayson T, Hamilton A, Hansen V, Leppo EW, Pelletier MC, Borja A, Cadien D, Dauer D, et al. 2015. Effect of ecological group classification schemes on performance of the AMBI benthic index in US coastal waters. *Ecol Ind* 50:99–107.

Grassle JF. 1977. Slow recolonisation of deep-sea sediment. *Nature* 265:618–619.

Grassle JF, Maciolek NJ. 1992. Deep-sea species richness: Regional and local diversity estimates from quantitative bottom samples. *Am Nat* 139:313–341.

Grassle JF, Morse-Porteous L. 1987. Macrofaunal colonization of disturbed deep-sea environments and the structure of deep-sea benthic communities. *Deep-Sea Res* 34:1911–1950.

Gray JS. 1981. The ecology of marine sediments. Cambridge (UK): Cambridge Univ. 185 p.

Haedrich RL, Devine J, Kendal V. 2008. Predictors of species richness in the deep-benthic fauna of the northern Gulf of Mexico.

Deep-Sea Res II 55:2650–2656.

Hecker B, Paul AZ. 1979. Abyssal community structure of the benthic infauna of the eastern equatorial Pacific: DOMES sites A, B, and C. In: Bischoff JF, Piper DZ, editors. Marine geology and oceanography of the Pacific Manganese Nodule Province. *Mar. Sci.* 9 New York (NY): Plenum. p 287–308.

Hessler RR, Sanders HL. 1967. Faunal diversity in the deep sea. *Deep-Sea Res* 14:65–78.

Hill MO. 1973. Diversity and evenness: A unifying notation and its consequences. *Ecology* 54:427–432.

Jumars PA. 1976. Deep-sea species diversity: Does it have a characteristic scale? *J Mar Res* 34:217–246.

Montagna PA, Baguley JG, Cooksey C, Hartwell I, Hyde LJ, Hyland JL, Kalke RD, Kracker LM, Reuscher M, Rhodes ACE. 2013. Deep-sea benthic footprint of the Deepwater Horizon blowout. *PLOS One* 8(8)/e70540:1–8.

Montagna, PA, Baguley JG, Cooksey C, Hyland JL. 2013. Deepwater Horizon oil spill: Assessment of potential impacts on the deep soft-bottom benthos. Charleston (SC): National Oceanic and Atmospheric Administration (NOAA), National Centers for Coastal Ocean Science. 32 p. Interim Data Summary Report. NOAA Technical Memorandum NOS NCCOS 166.

Montagna PA, Harper Jr DE. 1996. Benthic infaunal long-term response to offshore production platforms. *Can J Fish Aquat Sci* 53:2567–2588.

Neff JM. 2005. Composition, environmental fates, and biological effect of water based drilling muds and cuttings discharged to the marine environment: A synthesis and annotated bibliography. Duxbury (MA): Battelle. 73 p. Prepared for Petroleum Environmental Research Forum (PERF) and American Petroleum Institute.

Neff JM, Rabalais NN, Boesch DF. 1987. Offshore oil and gas development activities potentially causing long-term environmental effects. Chapter 4. In: Boesch DF, Rabalais NN, editors. Long-term environmental effects of offshore oil and gas development. London (UK): Elsevier. p 149–174,

Pearson TH, Rosenberg R. 1978. Macrobenthic succession in relation to organic enrichment and pollution of the marine environment. *Oceanogr Mar Biol Annu Rev* 16:229–311.

Peterson CH, Kennicutt II MC, Green RH, Montagna P, Harper Jr DE, Powell EN, Roscigno PF. 1996. Ecological consequences of environmental perturbations associated with offshore hydrocarbon production: A perspective from study of long term exposures in the Gulf of Mexico. *Can J Fish Aquat Sci* 53:2637–2654.

Raffaelli DG, Mason CF. 1981. Pollution monitoring with meiofauna, using the ratio of nematodes to copepods. *Mar Pollut Bull* 12:158–163.

Rex MA. 1981. Community structure in the deep-sea benthos. *Ann Rev Ecol System* 12:331–353.

Rowe GT, Kennicutt II MC, editors. 2008. The deep Gulf of Mexico benthos program. *Deep-Sea Res Part II* 55 (Issues 24–26).

Rowe GT, II Kennicutt MC, editors. 2009. Northern Gulf of Mexico continental slope habitats and benthic ecology study. New Orleans (LA): US Dept of Interior, Minerals Management Service, Gulf of Mexico OCS Region. 456 p. Final report. OCS Study MMS 2009-039.

Rowe GT, Polloni PT, Haedrich RL. 1982. The deep-sea macrobenthos on the continental margin of the northwest Atlantic Ocean. *Deep-Sea Res* 29:257–278.

Rowe GT, Wei C, Nunnally C, Haedrich R, Montagna P, Baguley JG, Bernhard JM, Wicksten M, Ammons A, Escobar Briones E, et al. 2008. Comparative biomass structure and estimated carbon flow in food webs in the deep Gulf of Mexico. *Deep-Sea Res II* 55:2699–2711.

Ryerson TB, Camilli R, Kessler JD, Kujawinski EB, Reddy CM, Valentine DL, Atlas E, Blake DR, de Gouw J, Meinardi S, et al. 2012. Chemical data quantify Deepwater Horizon hydrocarbon flow rate and environmental distribution. *Proc Natl Acad Sci USA* 109:20246–20253.

Tenore KR. 1977. Food chain pathways in detrital feeding benthic communities: A review, with new observations on sediment resuspension and detrital recycling. In: Coull BC, editor. Ecology of the marine benthos. Columbia (SC): Univ South Carolina. p 37–54.

Thistle D. 2003. The deep-sea floor: An overview. Chapter 2. In: Tyler PA, editor. Ecosystems of the deep. Amsterdam (NL):

Elsevier. p 5–38.
Tyler PA. 2003. Introduction. Chapter 1. In: Tyler PA, editor. Ecosystems of the deep. Amsterdam (NL): Elsevier. p 1–4.
Valentine DL, Burch Fisher G, Bagbya SC, Nelson RK, Reddy CM, Sylva SP, Wood MA. 2014. Fallout plume of submerged oil from Deepwater Horizon. *Proc Nat Acad Sci* 111:15906–15911.
[UAC] Unified Area Command. 2010. NOAA cumulative subsurface trajectory for deepwater. In: Deepwater Horizon MC 252 response, Unified Area Command, strategic plan for sub-sea and sub-surface oil and dispersant detection, sampling, and monitoring, November 13, 2010. New Orleans (LA): UAC. p 27–28.
Wei C, Rowe G. 2006. Zonation in the macrobenthos of the deep northern Gulf of Mexico. ICES Annual Meeting, Maastricht, Holland, September 2006.
Wei C, Rowe GT, Hubbard GF, Scheltema AH, Wilson GDF, Petrescu I, Foster JM, Wicksten MK, Chen M, Davenport R, et al. 2010. Bathymetric zonation of deep-sea macrofauna in relation to export of surface phytoplankton production. *Mar Ecol Prog Ser* 399:1–14.

<abstract type=”short”>Key Points
Contaminants from the Deepwater Horizon blowout persisted for 1 y in the deep sea.</B1>
Persistent community structure change occurred for both macrofauna and meiofauna communities within 1 to 3 km of the wellhead.</B1>
Biodiversity loss persisted for both macrofauna and meiofauna taxa richness.</B1>
Persistent contaminants, biodiversity losses, and community structure change in the impact zone indicate little recovery in the deep sea 1 y after the spill.</B1></BL>

Figure 1. Station locations overlayed with impact zones as represented by principal components analysis (PCA) scores as defined by Montagna, Baguley, Cooksey, Hartwell et al. (2013): All stations within impact and reference zones (**A**); zoomed to within 10 km from the MC252 wellhead with distance rings at 1 and 3 km from wellhead (**B**).

Figure 2. Interactions between year and zone treatments for sediment chemistry, including mean and SDs for cells: Total PAHs for 44 priority compounds (PAH44) (**A**); total petroleum hydrocarbons (TPHs) (**B**); Ba (**C**); mud, that is, total silt plus clay content (**D**). Means and SDs are back-transformed for PAH44, TPH, and Ba.

Figure 3. Interactions between year and zone treatments for macrofauna metrics, including mean and SDs for cells: Abundance, n m^{-2}, based on log back-transformed values (**A**); taxa richness (R, number of taxa) (**B**); diversity, Hill's N1 (**C**). TPH = total petroleum hydrocarbon.

Figure 4. Interactions between year and zone treatments for meiofauna metrics, including mean and SDs for cells: Abundance, n m^{-2} based on log back-transformed values (**A**); taxa richness (R, number of taxa) (**B**); diversity, N1 (**C**); nematode to copepod ratio (**D**).

Figure 5. Nonmetric multidimensional scaling plot of macrofauna community structure with respect to the 2-way design (year vs zone).

Figure 6. Nonmetric multidimensional scaling plot of meiofauna community structure with respect to the 2-way design (year vs zone).

Table 1. Chemistry: Mean values of targeted abiotic environmental variables by year and zone, and ANOVA results[a]

Response variable	**Cell means**				**ANOVA**[b]		
	2010		2011		Probability *F*		
	Impact	Non-impact	Impact	Non-impact	Year	Zone	Year*Zone
Total PAHs (μg/kg)	3270	68	4020	25	0.4302	<0.0001[c]	0.2242
TPH (μg/g)	520	20	760	9	0.5906	<0.0001[c]	0.1023
Ba (μg/g)	1250	420	850	400	0.0414[c]	0.0016[c]	0.0807
Silt + clay (%)	97	98	95	97	0.0158[c]	0.0766[c]	0.1981

ANOVA = analysis of variance; TPH = total petroleum hydrocarbon.

[a] Average water depth is 1344 m at impact stations and 1520 m at nonimpact stations.

[b] PAH, TPH, and Ba ANOVAs are based on log-transformed values, and means are back-transformed.

[c] Significant *P* value.

Table 2. Macrofauna: Mean values of targeted response variables by zone and year, and results of ANOVA

Response variable	**Cell means**				**ANOVA**		
	2010		2011		Probability *F*		
	Impact	Nonimpact	Impact	Nonimpact	Year	Zone	Year*Zone
Density[a] (n/m^{-2})	7000	8600	11 800	8900	0.0314[b]	0.7583	0.0567
Richness (# taxa sample^{-1})	17	25	20	26	0.0134[b]	0.0034[b]	0.2666
Diversity (Hill's N1 sample^{-1})	11	17	11	18	0.0978[b]	0.0007[b]	0.6656

ANOVA = analysis of variance.

[a] Density was log-transformed prior to analysis and back-transformed for presentation here.

[b] Significant *P* value.

Table 3. Meiofauna: Mean values of targeted response variables by zone and year, and results of ANOVA

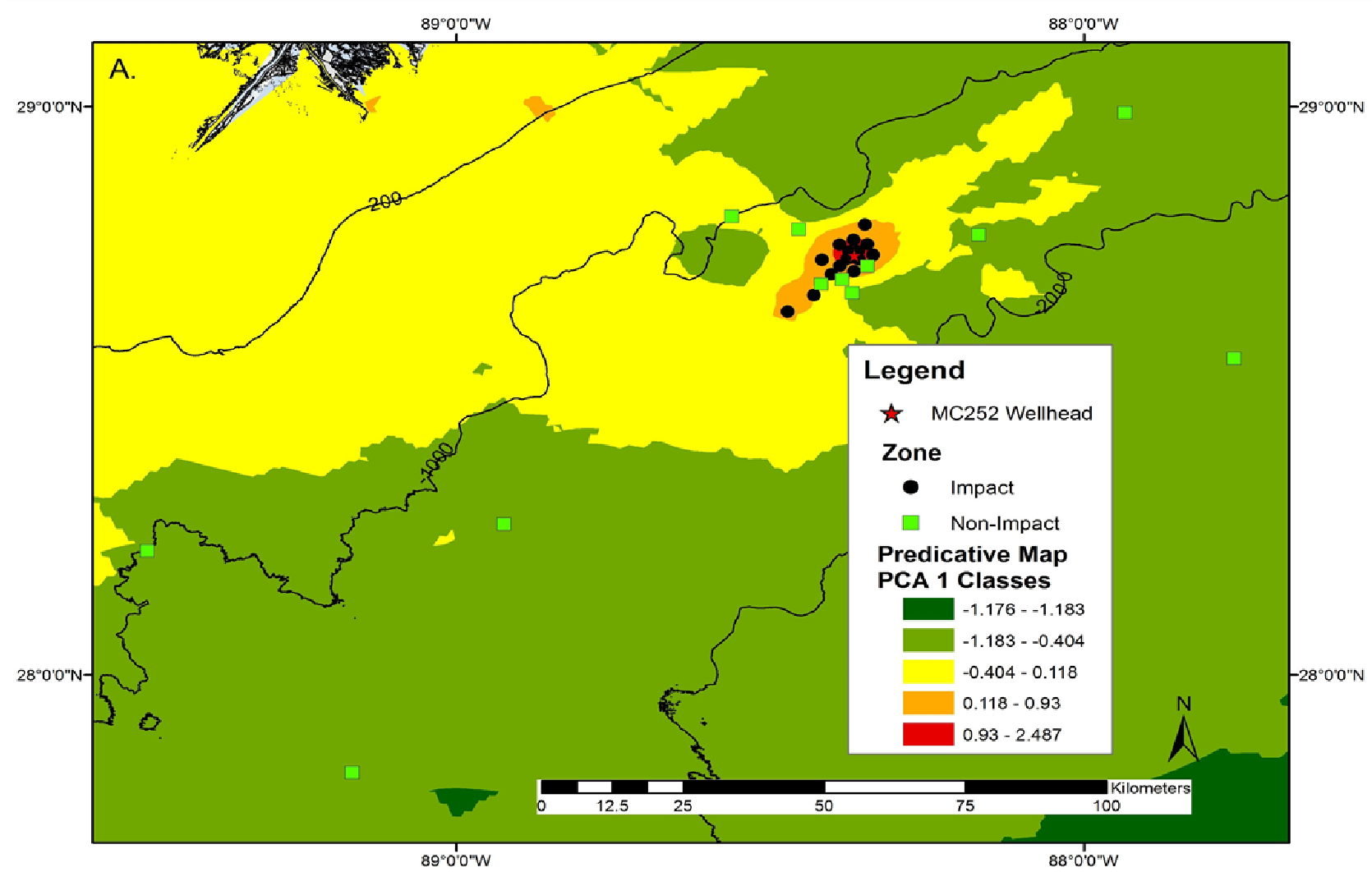
Response variable	**Cell means**				**ANOVA**		
	2010		2011		Probability *F*		
	Impact	Nonimpact	Impact	Nonimpact	Year	Zone	Year*Zone

Density[a] (n/m^{-2})	2 710 000	1 728 000	2 080 000	1 783 000	0.1976	0.2005	0.1094
Richness (# taxa sample^{-1})	8	10	10	14	<0.0001[b]	0.0002[b]	0.0072[b]
Diversity (Hill's N1 sample^{-1})	1.41	2.01	1.86	1.86	0.0536[b]	0.0002[b]	0.0008[b]
N:C	29.4	5.7	6.9	7.8	0.0212[b]	0.0109[b]	0.0064[b]

ANOVA = analysis of variance; N:C = nematode to copepod ratio.

[a] Density was log-transformed prior to analysis and back-transformed for presentation here.

[b] Significant *P* value.



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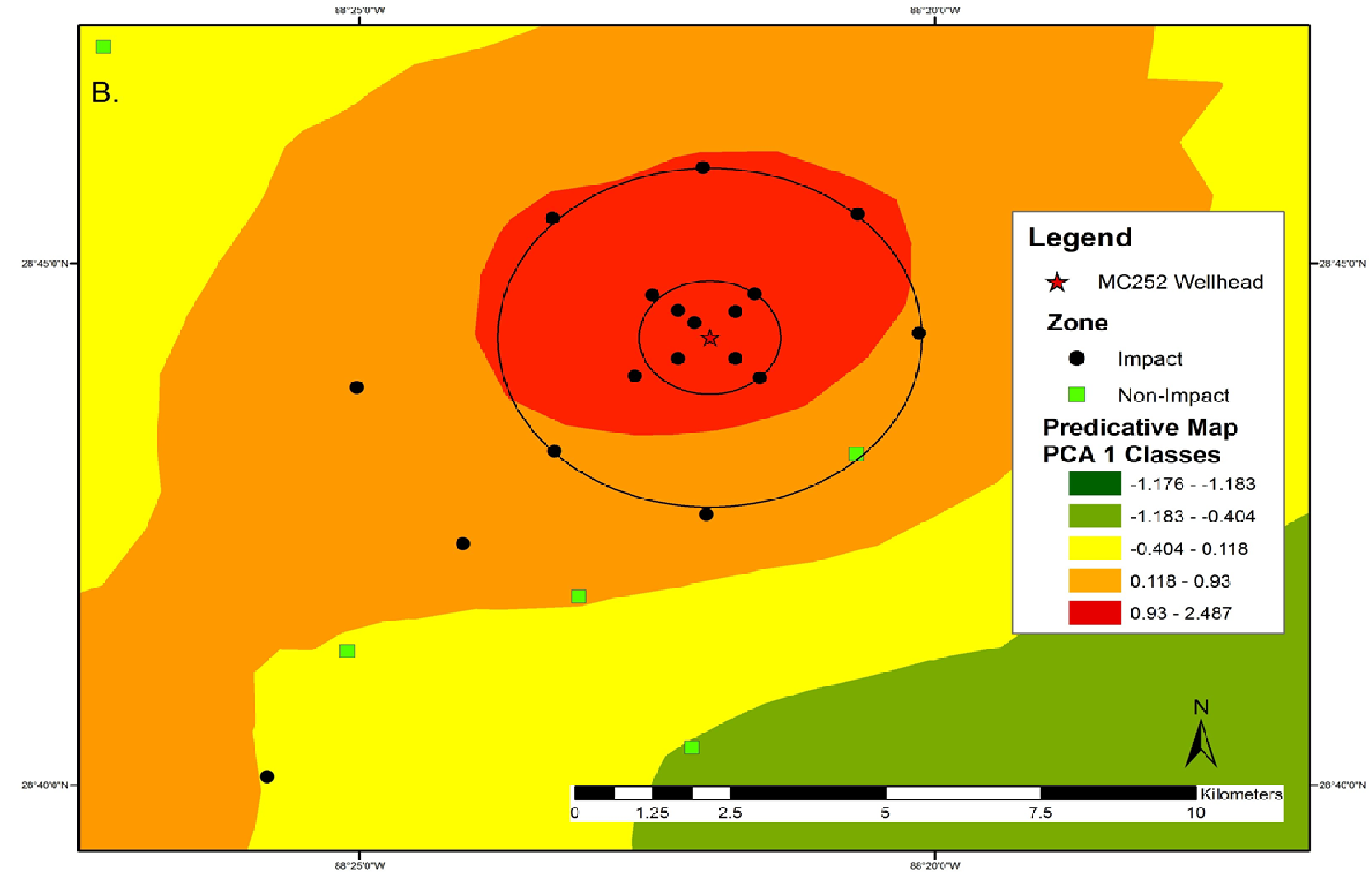

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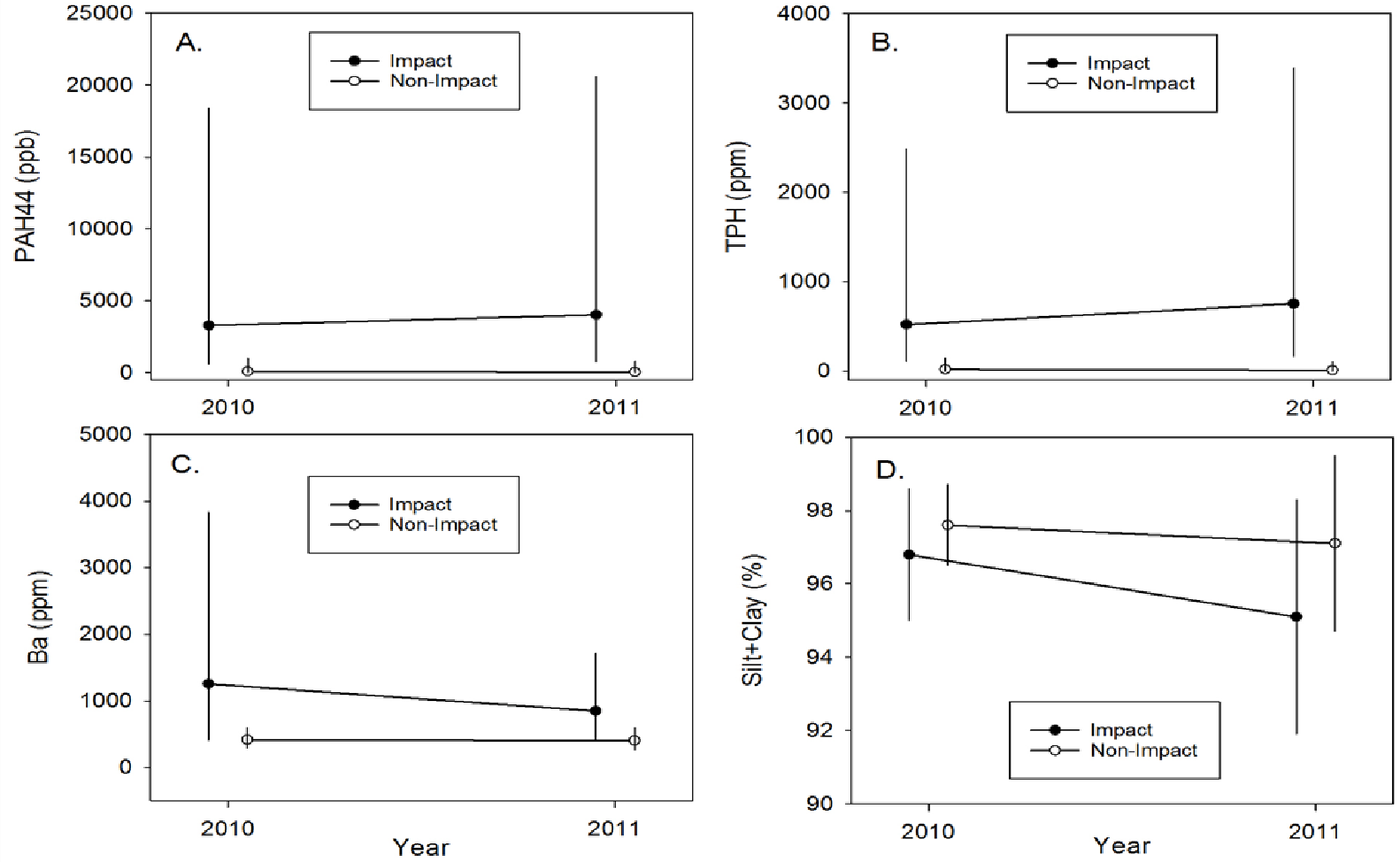

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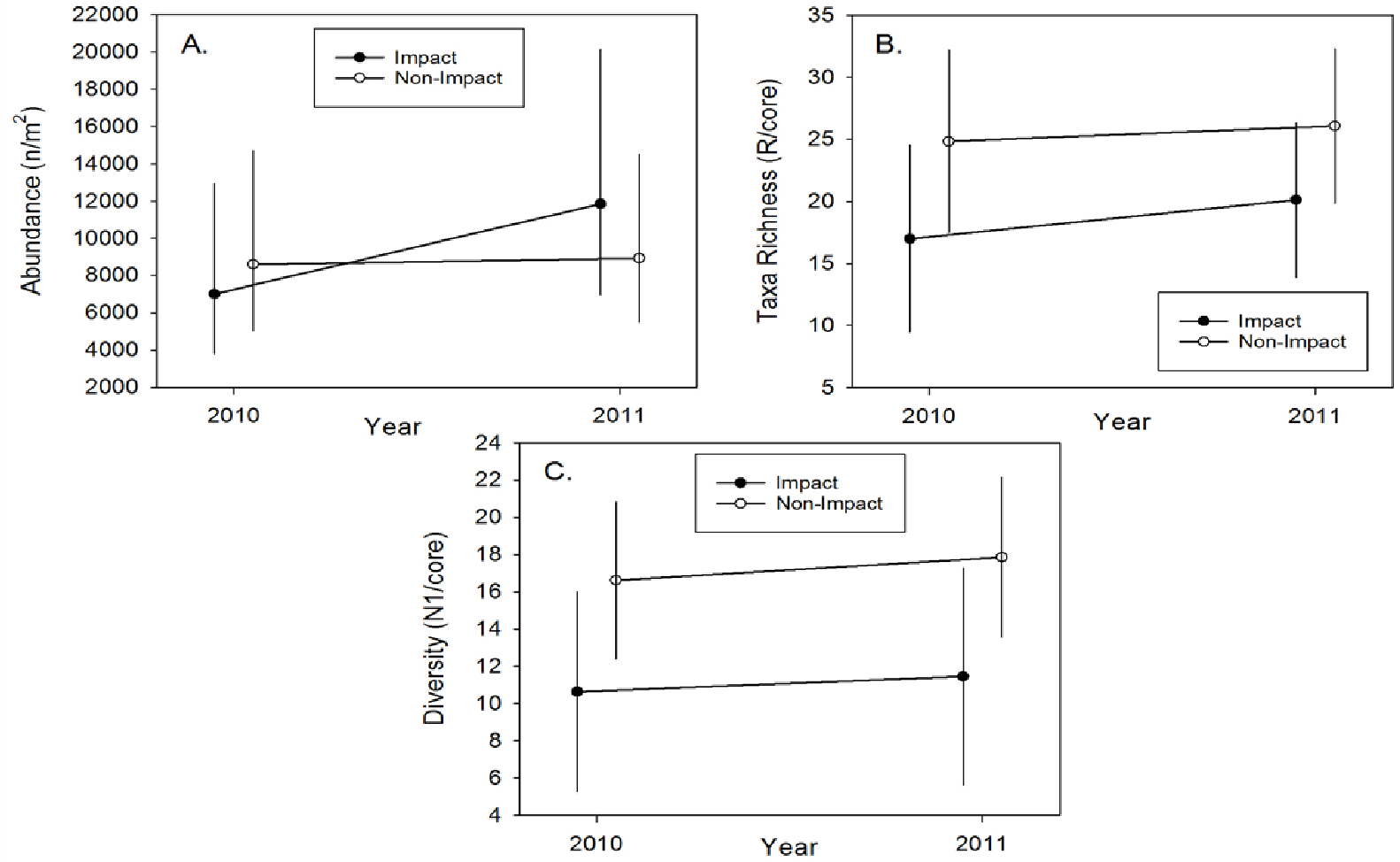

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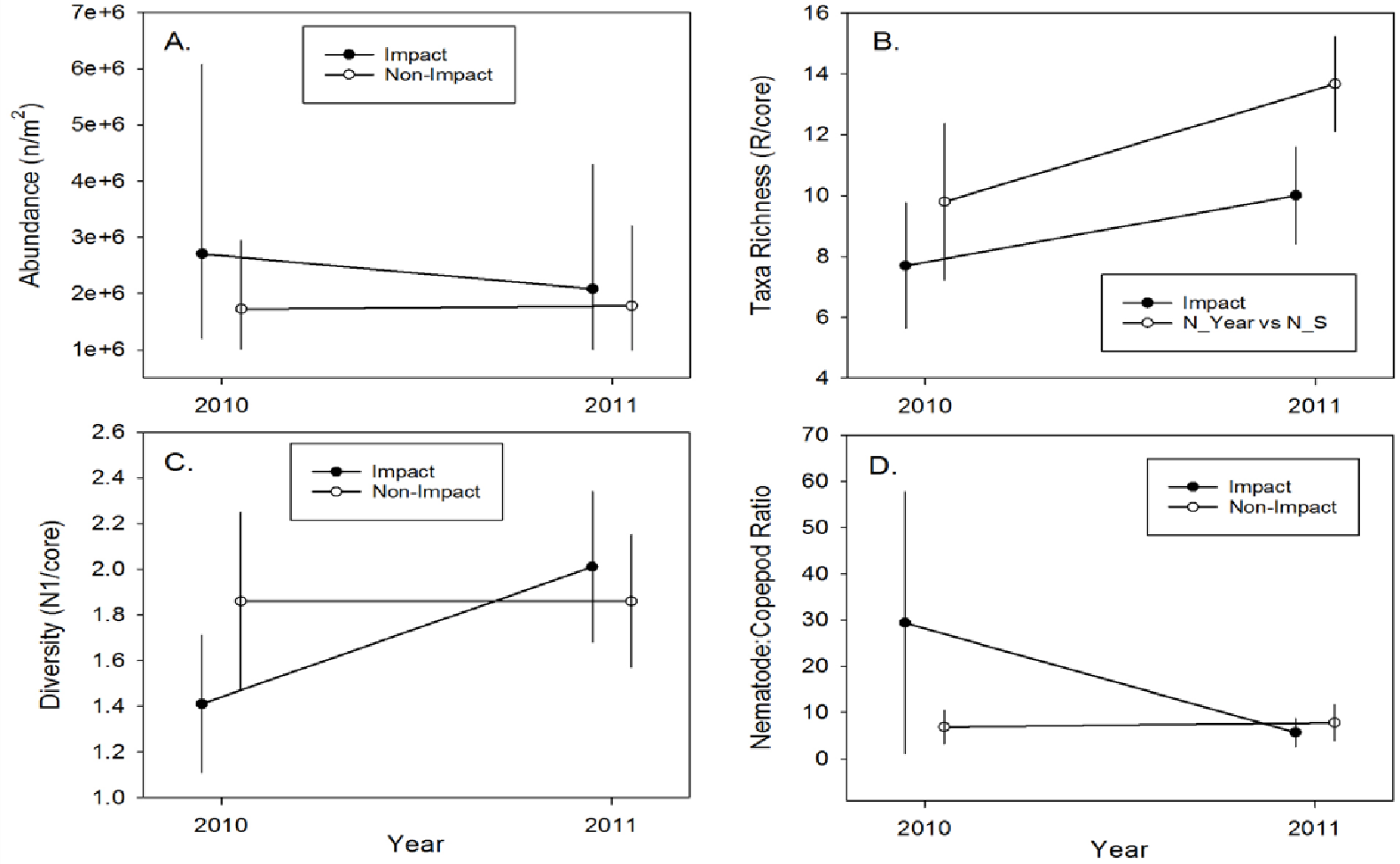

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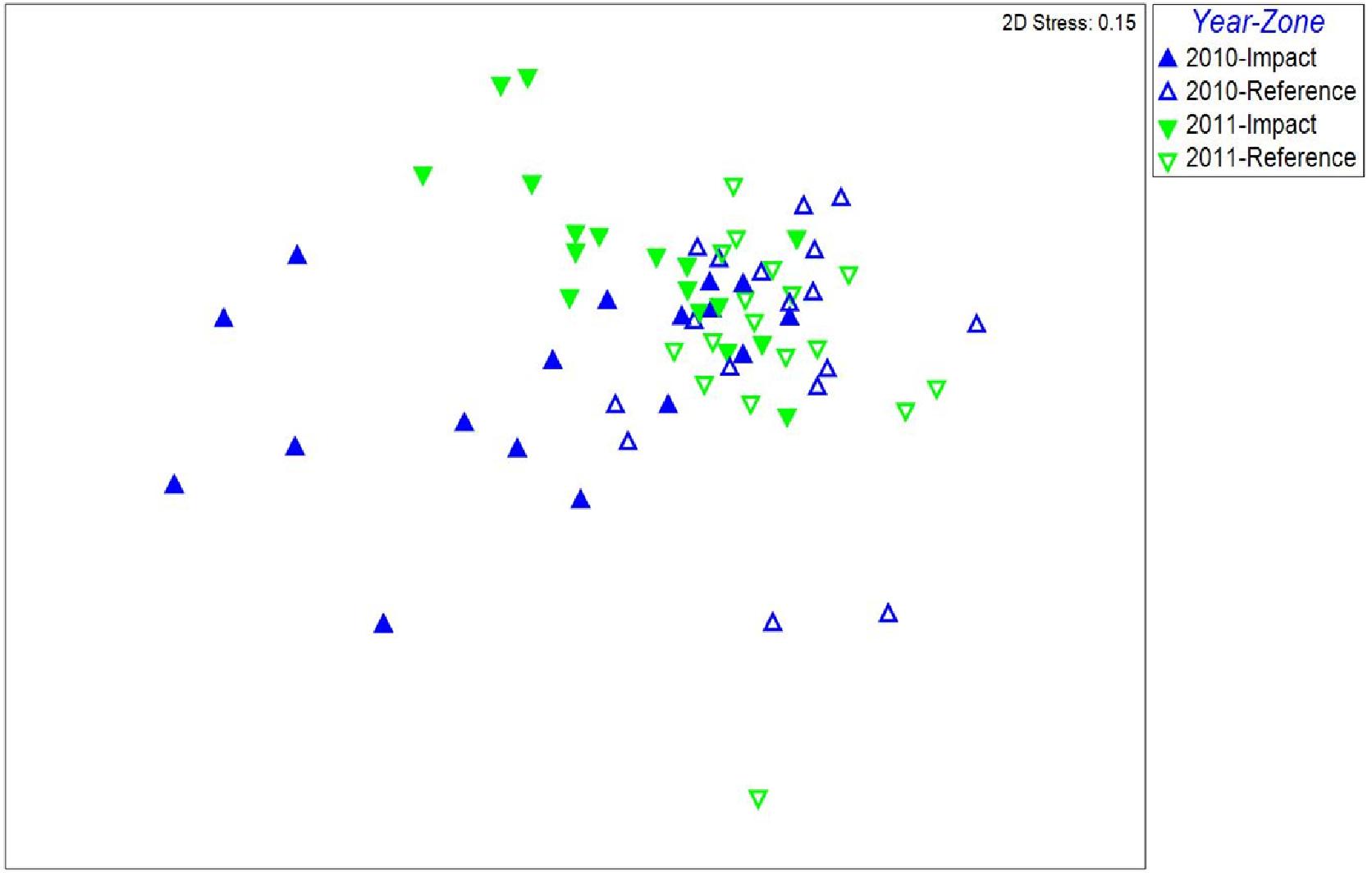

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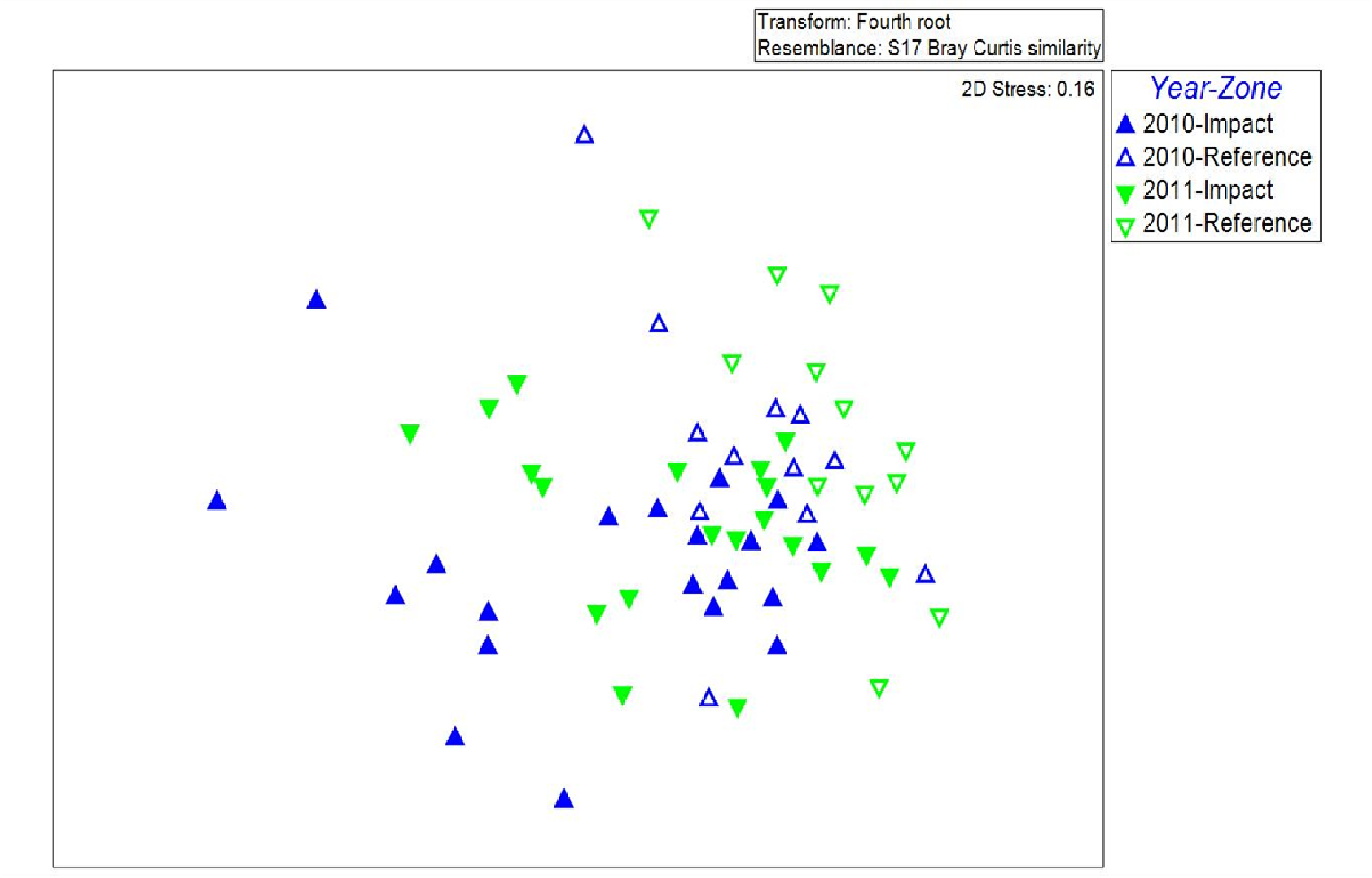

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