

Montara Well Release Monitoring Study S4A –

Assessment of Effects on Timor Sea Fish **FINAL REPORT**

15 November 2011



Curtin University

Summary of Results

- Biopsies were collected on a total of 181 fish sampled during Phase I (November 2009), on 262 fish during Phase II (March 2010), and on 338 fish in Phase III (November 2010) with the majority of specimens being goldband snapper, red emperor, rainbow runner and Spanish mackerel.

- For each species, all individuals were in good physical condition at all sites, suggesting good health status.

- Results from Phase I indicated that in the short-term, fish were exposed to, and metabolised petroleum hydrocarbons, however no consistent adverse effects on fish health or on their reproductive activity were detected.

- In Phase II, continuing exposure to petroleum hydrocarbons was evidenced by elevated liver detoxification enzymes and PAH biliary metabolites in three out of four species collected close to the rig; in addition, red emperor collected close to the West Atlas had enlarged livers and elevated oxidative DNA damage.

- Phase III established that biomarkers of fish health showed a trend towards a return to reference levels with often, but not always, comparable biomarker levels in fish collected from reference and impacted sites.

- Liver integrity was preserved at all times, for all four species of fish collected.

- No reproductive impairment or structural alteration of gonadal tissues were observed in any of the species, up to a year following the end of the well release.

- Multivariate analysis suggests that overall differences between sites across all biomarkers were influenced mainly by biomarkers of exposure to petroleum hydrocarbons. Further, these analyses suggest that as the time post-release increases, the overall biomarker signals from different sites appear to converge.

- Overall, the fish collected initially showed evidence of exposure to petroleum hydrocarbons at sites close to the West Atlas drilling rig, increased liver size and occasionally, increased oxidative DNA damage in Phase I and II of the monitoring. While at some sites differences in biomarker levels were still observed one year after the end of the well release (Phase III), the magnitude of the differences had reduced relative to earlier samplings, suggesting a partial, ongoing trend toward a return to normal biochemistry/physiology following exposure to petroleum hydrocarbons.

Preface

This report was prepared by Drs Marthe Monique Gagnon and Christopher Rawson from the Department of Environment and Agriculture, Curtin University. The data referred to herein were collected from aboard the FV Megan M on three occasions, with a first field collection from 5th to 9th November 2009 (Phase I), a second collection from 4th to 18th March 2010 (Phase II), and a Phase III collection which took place on 9th to 30th November 2010. All biomarker analyses were conducted in the Aquatic Toxicology laboratories in the Department of Environment and Agriculture at Curtin University.

Acknowledgements

Special thanks to Grant Barker, captain of the FV Megan M and to the crew (Shane Ross, Beau Pieterman, Mitch Seelander and Matt Badart) for their assistance in the collection of fish samples. Thanks to Leif Cooper for assistance in the collection of sediment samples. The technical assistance of Tomoe Ota is sincerely thanked for the preparation of the histological slides of gonad tissues.

Document History

Rev	Date	Authors	Comments
A	5 May 2011	MM Gagnon CA Rawson	Initial Draft for comment to PTTEPAA
B	15 Sept 2011	MM Gagnon CA Rawson	Draft II following comments from SEER and PPR
C	8 Oct 2011	MM Gagnon CA Rawson	Draft III following comments from Robin Wright PTTEPAA
D	10 Oct 2011	MM Gagnon CA Rawson	Final Formal Version

For further information contact:

Associate Professor Marthe Monique Gagnon
Department of Environment and Agriculture
Curtin University
GPO Box U1987
Perth, Western Australia, 6845
Tel.: (08) 9266 3723
Email: m.gagnon@curtin.edu.au

Recommended Citation:

Gagnon M.M., Rawson C., 2011. Montara Well Release, Monitoring Study S4A – Assessment of Effects on Timor Sea Fish. Curtin University, Perth, Australia. 208 pages.

Disclaimer:

The views contained in this report are those of the authors and do not necessarily reflect the views of PTTEPAA, SEWPaC or any other party.

ISBN 978-0-9872223-0-5



Executive Summary

The Montara well release that occurred in the Timor Sea in 2009 discharged an estimated 23,000 barrels of oil and gas condensate in a high biodiversity marine environment, raising immediate concerns of economic, political and environmental significance. Of economic relevance are the short – and long-term impacts of the discharge on commercial fisheries, with the Northern Demersal Scale Fisheries exploiting stocks of goldband snapper (*Pristipomoides multidens*) and red emperor (*Lutjanus sebae*) in this area. Albeit to a lesser extent, rainbow runner (*Elegatis bipinnulata*) and narrow-barred Spanish mackerel (*Scomberomorus commerson*) are also fished recreationally in this environment. The well release occurred during the lead-up to the active reproductive season for these species.

The well release was controlled after seventy-four days after which a scientific team of ecotoxicologists was assigned to sample the commercially important fish species and evaluate the impacts of the hydrocarbon release on fish health. The FV Megan M commercial fishing boat was chartered in November 2009 for the first collection of biopsies on pelagic and demersal fish. In this first Phase of the study (Phase I), biopsies were collected on 145 demersal and 36 pelagic fish respectively. A second field collection occurred in March 2010, four months following the end of the well release. In the second Phase of the study (Phase II), biopsies were collected on 202 demersal and 60 pelagic fishes. Finally, a third sampling (Phase III) was conducted in November 2010, 12 months after the incident. During Phase III, 315 demersal and 23 pelagic fish were captured and sampled for biopsy collections. Biopsies were used to evaluate the short and long-term impacts of exposure to petroleum hydrocarbons on fish health, by the determination of physiological indices and biochemical markers (biomarkers).

The selected physiological indices were the condition factor, liver somatic index and the gonado-somatic index. Biochemical markers measured included liver detoxification activity, biliary polycyclic aromatic hydrocarbon (PAH) metabolites, liver integrity measured by serum sorbitol dehydrogenase activity (SDH), oxidative DNA damage and histological examination of the gonads.

Results from Phase I indicated that in the short-term, fish were exposed to, and metabolised, petroleum hydrocarbons however, no consistent adverse effects on fish health or on their reproductive activity were detected. The physiology of the fish was not affected by exposure to the hydrocarbon release, (i.e. the condition factor (fattiness) of the fish) and the liver and gonad sizes relative to their body weight were similar in fish collected within the impacted zone and in fish collected in reference areas. Similarly, liver integrity was maintained in fish from all areas, liver detoxification activity was similar at all sites, and there was no evidence of increased oxidative DNA damage at study sites near the West Atlas drilling rig. Histological examination of the gonads revealed no abnormalities immediately following the control of the well release.

Phase II investigated possible effects of exposure on fish health four months after the cessation of the hydrocarbon leak. In the longer term (4 months following the end of the well release), red emperor captured within 20 NM from the rig exhibited altered physiological indices such as larger liver size relative to body size, and higher condition factor, relative to the fish from reference areas. Female rainbow runner and male goldband snapper captured in the vicinity of the West Atlas rig had reduced gonad weight relative to body weight. Confirmation of ongoing exposure to PAHs was measured in demersal fish captured within 20 NM from the rig, as evidenced by high PAH biliary metabolites and high liver detoxification activity in these fish, relative to those collected in reference areas. While liver integrity was not compromised in any species, red emperor captured 20 NM from the rig had higher DNA damage than those fish originating from reference zones. Gonad histology did not identify reproductive abnormalities in any on the fish species.

The final phase of the study, Phase III study, was conducted 12 months following the end of the well release and sampled 338 fish from 3 impacted and 5 reference sites. For all fish species collected during Phase III, the condition factor from fish originating from the reference and impacted sites was comparable, indicating that all fish were in similar physical condition. Some variations were observed in liver size relative to body size in goldband snapper and red emperor, however these variations were modest and not consistent throughout impacted and reference areas. Similarly, the gonad size relative to body size showed some inter-site variability in male red emperor and female goldband snapper however no consistent pattern emerged. The onset of reproductive activities in these species is likely the main factor associated with these variations. One year following the end of the spill, no reproductive impacts could be detected in any of the fish species as histological examination of structural gonadal tissues revealed the absence of abnormalities or pathologies. The liver detoxification activity in goldband snapper and red emperor collected within 20 NM from the rig has returned towards reference levels, with the exception of fish of these two species collected at a single impacted site (Heywood shoal) located 53 NM south-west of the rig which appeared to have continuing high liver detoxification activity.

It is possible that a natural seepage occurs in the area, as one year after the spill fish collected from other impacted locations do not exhibit similar high liver detoxification activities. All fish originating at the impacted sites, including those with high detoxification activity, had levels of PAH metabolites in their biliary secretions which were comparable to those of fish collected at reference sites. Liver integrity in all fish species appeared to be intact. Oxidative DNA damage, which was previously found to be elevated in fish captured in close proximity of the well release, had returned to reference levels for all fish species, at all sites.

The results of a multivariate analysis indicates that over the entire sampling period, there were significant differences between impacted sites reference sites and that these differences were mostly due to the levels of PAH metabolites in the bile of the fish. Through time the impacted and non-impacted areas tended to converge towards a similar overall biomarker signal. This finding points to a reduction in the differences between the sites and, to a reduction in exposure to petroleum hydrocarbons at the “impacted” sites.

Overall, shortly following the control of the well release, fish captured in the oil-impacted area exhibited evidence of exposure to petroleum hydrocarbons and were showing, in the short term, very limited adverse health impacts. The investigation conducted four months after the spill revealed continuing exposure to petroleum hydrocarbons in demersal species captured close to the rig, with associated alterations to physiological indices consistent with chronic exposure to petroleum compounds. The survey conducted twelve months after the incident showed return towards reference levels of physiological and biochemical markers in fish captured close to the rig location. A small number of fish from one site however, returned some results that suggest exposure to natural or released hydrocarbons. Histological morphology of male and female gonads remained unaltered in all fish species from all locations.

Environmental factors such as the warm water temperatures found in the Timor Sea would favour weathering processes and biodegradation of the light Montara oil. Monitoring fish health at impacted and reference locations has demonstrated that fish were initially exposed to petroleum hydrocarbons, however, evidence of exposure and effects have significantly reduced one year following the end of the Montara well release. The reduced inter-site biomarker variability observed suggests an ongoing return towards reference levels.

Table of Contents

Executive Summary	v
Background	1
Toxicological Concepts.....	2
Physiological Parameters.....	2
Biochemical Markers (Biomarkers).....	3
Liver Detoxification Enzymes.....	3
Biliary Metabolites.....	3
Sorbitol Dehydrogenase (SDH).....	4
Oxidative DNA Damage.....	4
Project Aims	5
Sampling Timeline – Phase I	7
Sampling Timeline – Phase II	9
Sampling Timeline – Phase III	11
Sampling Sites	13
Field Methodology	17
Fish Sampling.....	17
Morphology.....	19
Biopsies Collected.....	19
Additional Sampling.....	19
Muscle Samples.....	19
Stomach/Intestinal Contents.....	20
Floating Material Samples.....	22
Water Samples.....	22
Sediment Samples.....	22
Laboratory Methods	23
Physiological Parameters.....	23
Liver Detoxification Enzymes (EROD Activity).....	23
Biliary Metabolites.....	24
Sorbitol Dehydrogenase (SDH) Activity.....	24
Oxidative DNA Damage (Serum 8-oxo-dG Concentration).....	24
Gonad Histology.....	24
Statistical Analyses.....	25

Results and Interpretation	27
Sampling Summary	27
Diversity	27
Biopsy Collection	29
General Fish Health	31
Physiological Parameters	31
Condition Factor	31
Liver Somatic Index (LSI)	34
Gonado-somatic Index (GSI)	36
Gonad Histology	41
Biochemical Parameters	48
Liver Detoxification Enzymes	48
Biliary Metabolites	51
Sorbitol Dehydrogenase (SDH) Activity	58
Oxidative DNA Damage (Serum 8-oxo-dG Concentration)	61
Multivariate Analysis	64
Conclusions	67
References	71
Appendix A – Statistical Results Phase I	73
Appendix B – Statistical Results Phase II	81
Appendix C – Statistical Results Phase III	89
Appendix D – Results of Floating Material Analysis	97
Appendix E – Results of Water Samples Analysis	109
(Phase I and Phase II)	109
Appendix F – Results of Sediment Analysis	145
(Phase III)	145

List of Figures

Figure 1. Map showing the location of the West Atlas drilling rig and the sampling sites. Concentric rings represent 20, 80 and 150 NM from the platform. Sites 1, 2, 3 and 4 were designated as impacted sites and Sites 5 and 6 were designated as reference sites in Phase I sampling. Site 7 is located at Scott Reef. Surface samples are plankton net trawls collecting observed floating material.	14
Figure 2. Method of demersal fish capture. Baited steel fish trap prior to deployment. In picture: Matthew Badart (left) and Beau Pieterman (right).	18
Figure 3. Left: flow-through live tank used to preserve the fish in good condition following capture; right: fish were initially measured and weighed prior to sacrifice.	18
Figure 4. Left: blood collection from the caudal artery on a goldband snapper; right: bile collection using a 1 mL syringe.	19
Figure 5. Parasitic cysts adhering to the stomach of a Spanish mackerel (top), and flat worm attached to the liver of a goldband snapper (bottom).	31
Figure 6. Condition factor of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. "Impacted" sites are in red and "reference" sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	32
Figure 7. Condition factor of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. "Impacted" sites are in red and "reference" sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	32
Figure 8. Condition factor of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. "Impacted" sites are in red and "reference" sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	33
Figure 9. Condition factor of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. "Impacted" sites are in red and "reference" sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	33
Figure 10. Liver somatic index of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. "Impacted" sites are in red and "reference" sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	34
Figure 11. Liver somatic index of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. "Impacted" sites are in red and "reference" sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	35
Figure 12. Liver somatic index of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. "Impacted" sites are in red and "reference" sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	35
Figure 13. Liver somatic index of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. "Impacted" sites are in red and "reference" sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	36

Figure 14. Gonado-somatic index of male goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	37
Figure 15. Gonado-somatic index of male red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	37
Figure 16. Gonado-somatic index of male rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	38
Figure 17. Gonado-somatic index of male Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	38
Figure 18. Gonado-somatic index of female goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	39
Figure 19. Gonado-somatic index of female red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	40
Figure 20. Gonado-somatic index of female rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	40
Figure 21. Gonado-somatic index of female Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	41
Figure 22. Stained (haematoxylin-eosin) slides of gonadal tissue of female goldband snapper collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.	42
Figure 23. Stained (haematoxylin-eosin) slides of gonadal tissue of male goldband snapper collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.	43
Figure 24. Stained (haematoxylin-eosin) slides of gonadal tissue of female rainbow runner collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.	44
Figure 25. Stained (haematoxylin-eosin) slides of gonadal tissue of male rainbow runner collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.	45
Figure 26. Stained (haematoxylin-eosin) slides of gonadal tissue of female red emperor collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.	46
Figure 27. Stained (haematoxylin-eosin) slides of gonadal tissue of male red emperor collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.	47

Figure 28. EROD activity (pmol/ mg protein/ min) in goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	49
Figure 29. EROD activity (pmol/ mg protein/ min) in red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	50
Figure 30. EROD activity (pmol/ mg protein/ min) in rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	50
Figure 31. EROD activity (pmol/ mg protein/ min) in Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	51
Figure 32. Naphthalene type metabolite concentrations in the bile of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	52
Figure 33. Naphthalene type metabolite concentrations in the bile of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	53
Figure 34. Naphthalene type metabolite concentrations in the bile of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	53
Figure 35. Naphthalene type metabolite concentrations in the bile of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	54
Figure 36. Pyrene type metabolite concentrations in the bile of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	54
Figure 37. Pyrene type metabolite concentrations in the bile of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	55
Figure 38. Pyrene type metabolite concentrations in the bile of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	55

Figure 39. Pyrene type metabolite concentrations in the bile of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	56
Figure 40. Benzo(a)pyrene type metabolite concentrations in the bile of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	56
Figure 41. Benzo(a)pyrene type metabolite concentrations in the bile of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	57
Figure 42. Benzo(a)pyrene type metabolite concentrations in the bile of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	57
Figure 43. Benzo(a)pyrene type metabolite concentrations in the bile of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	58
Figure 44. SDH activity in goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	59
Figure 45. SDH activity in red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	59
Figure 46. SDH activity in rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	60
Figure 47. SDH activity in Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	60
Figure 48. Oxidative DNA damage in goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	62
Figure 49. Oxidative DNA damage in red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).	62

Figure 50. Oxidative DNA damage in rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$). 63

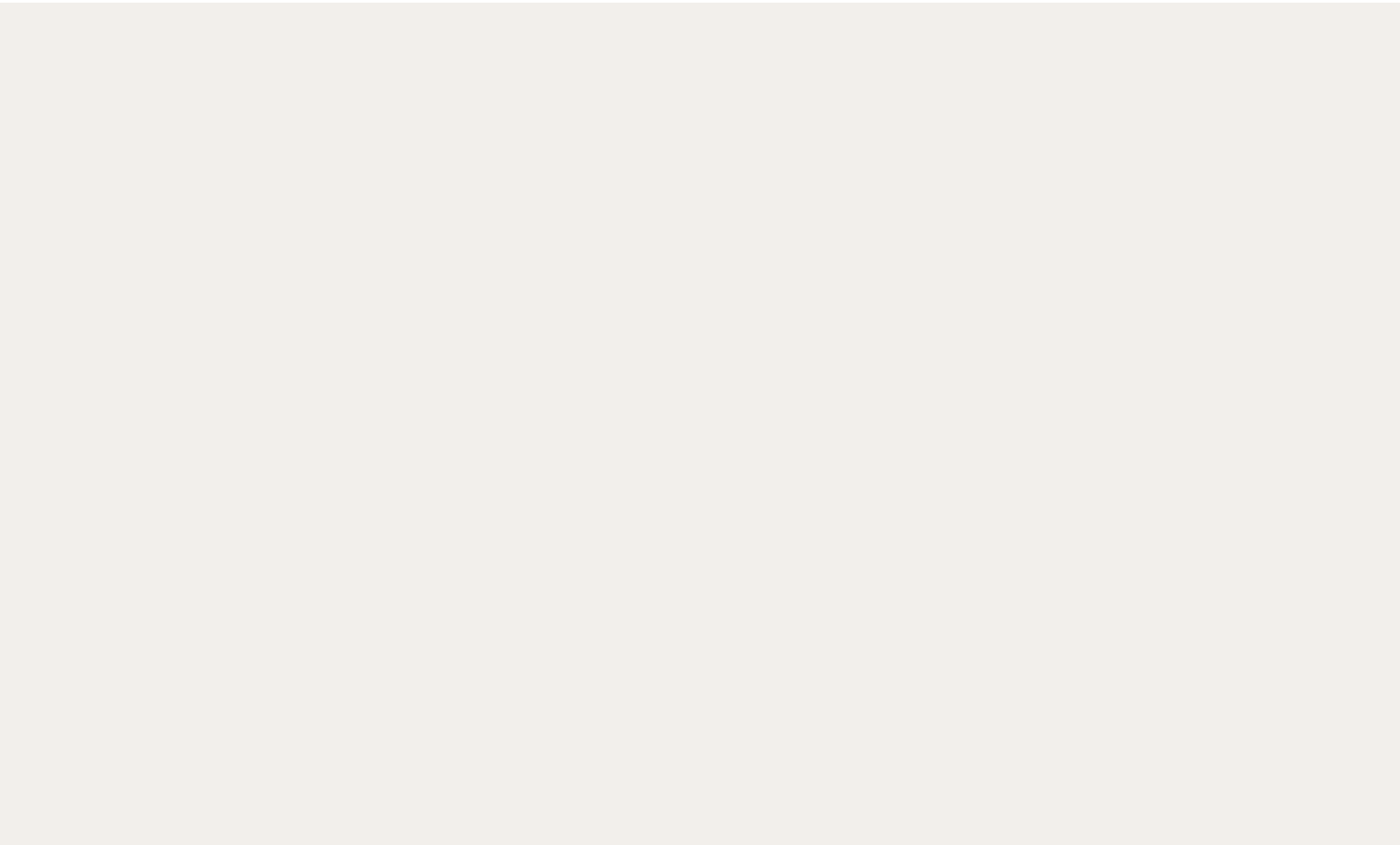
Figure 51. Oxidative DNA damage in Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$). 63

Figure 52. PCA on normalised data for combined biomarkers across study sites for goldband snapper. Top: Phase I, Middle: Phase II, Bottom Phase III. Insert histogram represents cumulative contribution of PCs used in the analysis. Insert matrix shows significant differences between sites as identified by analysis of similarity (ANOSIM) Euclidean distance. 65

Figure 53. PCA on normalised data for combined biomarkers across study sites for red emperor. Top: Phase I, Middle: Phase II, Bottom Phase III. Insert histogram represents cumulative contribution of PCs used in the analysis. Insert matrix shows significant differences between sites as identified by analysis of similarity (ANOSIM) Euclidean distance. 66

List of Tables

Table 1. Classification of physiological indices and biomarkers included in this study into indicators of exposure or effect.....	3
Table 2. Details of S4A Phase II sampling sites.	15
Table 3. Numbers of demersal fish from which stomach/intestine content and muscle for TPH analyses were collected during Phase I of the study.....	20
Table 4. Numbers of pelagic fish from which stomach/intestine content and muscle for TPH analyses were collected during Phase I of the study.....	20
Table 5. Numbers of demersal fish from which stomach/intestine content and muscle for TPH analyses and taint assessment were collected during Phase II of the study.....	21
Table 6. Numbers of pelagic fish from which stomach/intestine content and muscle for TPH analyses and taint assessment were collected during Phase II of the study.....	21
Table 7. Numbers of demersal fish from which stomach/intestine content and muscle for TPH analyses and taint assessment were collected during Phase III of the study.....	21
Table 8. Numbers of pelagic fish from which stomach/intestine content and muscle for TPH analyses and taint assessment were collected during Phase III of the study.....	22
Table 9. Diversity and number of each pelagic and demersal taxon captured during the November 2009 Phase I sampling effort.....	28
Table 10. Diversity and number of each taxon captured in demersal traps during the March 2010 sampling effort.	28
Table 11. Numbers of demersal fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase I of the study.	29
Table 12. Numbers of pelagic fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase I of the study.	29
Table 13. Numbers of demersal fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase II of the study.....	29
Table 14. Numbers of pelagic fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase II of the study.	30
Table 15. Numbers of demersal fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase III of the study.....	30
Table 16. Numbers of pelagic fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase III of the study.	30





Background

The seventy-four day long Montara well release that occurred in the Timor Sea discharged an estimated 23,000 barrels of oil and gas condensate in a high biodiversity marine environment. The release occurred in an area utilised for commercial fishing activities exploiting demersal and pelagic fisheries. The area is an ecologically important habitat to a variety of marine life including fish, reptiles, cetaceans and aquatic birds. It is crucial that any short and long-term impacts of the well release on the health of the resident fish populations be assessed and hence any commercial impact on the fishery be understood.

The presence of petroleum hydrocarbons in the receiving marine environment or in the food chain does not, by itself, indicate injurious effects. In addition, deleterious effects on populations are often difficult to detect in feral organisms as many biologically relevant effects tend to manifest themselves only after chronic, sustained exposure to contaminants (van der Oost *et al.*, 2003). Because contaminant effects initially occur at the molecular level, it is relevant to use molecular, biochemical and histological markers of exposure and effects as early warning indicators of environmental harm. With sustained exposure, effects at biochemical and cellular levels will translate in effects at higher levels of biological organisation. It is therefore pertinent to complement biochemical and cellular markers with physiological indices that will vary with chronic, sustained exposure. No single biomarker can provide a comprehensive assessment of the health status of a fish population however, a suite of biomarkers will inform on the exposure and effects induced by the sustained assimilation of contaminants by marine organisms.

Biomarkers of exposure and effect are currently the most relevant and reliable tools for the assessment of fish health. Biomarkers are measurements in body fluids, cells or tissues indicating biochemical or cellular modifications due to the presence of toxicants. These include measuring the metabolites of polycyclic aromatic hydrocarbons (PAHs) in the bile of the fish (since PAHs are generally not accumulated in fish muscle: van der Oost *et al.*, 2003; Meador *et al.*, 1995), estimating the amount of DNA damage that has occurred, measuring the activity of liver detoxification enzymes, and measuring liver integrity and cellular damage. In addition, histological examination of the gonads can give important information on whether the reproductive state of the animal has been altered as a result of exposure to the contaminants. Finally, physiological parameters such as condition factor, liver somatic index, and gonado-somatic index will inform on the potential alteration of organs following chronic exposure to petroleum hydrocarbons.

The measurement of a suite of biomarkers of fish health is necessary as no single biomarker can provide comprehensive information on the health status of an organism. Amongst the biomarkers listed above, it has been demonstrated that the measurement of liver detoxification enzymes, of PAH biliary metabolites, and of genotoxic parameters such as DNA damage are currently the most valuable and informative tools to in the evaluation of environmental impacts (van der Oost *et al.*, 2003).

Toxicological Concepts

As higher organisms, fish integrate the direct and indirect effects of contaminant exposure and environmental variations. Their long life span, relative to most invertebrates, allows the integration of various natural and induced pressures, which reflects the integrated biological response and compensation mechanisms.

As no one single biological parameter can inform on the overall health status of an organism, a suite of biological parameters have to be measured. In this study on the health of fish potentially impacted by the Montara well release, a suite of physiological and biochemical parameters have been determined at different levels of biological organisation. Physiological parameters such as liver enlargement are usually associated with chronic exposure to bioavailable contaminants, while biochemical responses originating at the subcellular levels are used as sensitive early-warning indicators of toxicity.

Physiological Parameters

At the organism level, sustained (i.e. chronic) exposure can potentially results in modification of physiology, such as the loss or accumulation of lipid reserves, or hypo-/hypertrophy of specific organs. Simple physiological measurement of weight as related to the length of a fish provides a Condition Factor (CF), and can inform of the 'fattiness' of the fish. A low CF has been related in many laboratory and field studies to chronic chemical stress imposed on a fish.

Liver is a very plastic tissue and when an organism is assimilating and metabolising contaminants, the liver becomes enlarged in response to the increased metabolic demand imposed by the contaminants to metabolise. Consequently, the weight of the liver as a proportion of the total weight of the fish provides a Liver Somatic Index (LSI). A high LSI is often observed in relation to chronic exposure of fish to bioavailable contaminants.

Exposure to contaminants often results in a reduced ability of an individual to successfully reproduce, by inhibiting gonadal development, by affecting behaviour or via other mechanisms. A simple measure signifying normal or impaired gonadal development is the gonado-somatic index (GSI) calculated using the weight of the gonad as a proportion of the total weight of the fish. Similar GSI in fish exposed to contaminants, relative to fish collected in reference zones, is a first indication that gonad development is proceeding normally. It is prudent however, to complement this physiological index with histology of the gonads.

Histological examination of gonadal tissue can provide important information on the reproductive status of a fish. Where contamination occurs it is possible that malformations can be observed histologically, particularly where there is disruption to the normal steroidal hormone function. Possible malformations include the presence of female tissue in a male gonad (or vice versa), halted gamete production or delayed reproduction as energy resources might be redirected to contaminant metabolism in the stressed animal.

Biochemical Markers (Biomarkers)

The use of biomonitoring methods in the management of chemical pollution has several advantages over chemical monitoring, as chemical determination alone is not indicative of possible adverse effects on an organism. To measure biologically significant alterations related to bioavailable chemicals, biochemical markers (biomarkers) are measured in various organs.

Biomarkers measured in fish represent the best tool available today to measure environmental impacts related to industrial discharges to the aquatic environment (van der Oost *et al.*, 2003), leading to the incorporation of biomarker measurements in several large-scale monitoring programmes such as NOAA's National Status and Trends Program and the North Sea Task Force Monitoring Plan (Collier *et al.*, 1995; Lam and Gray, 2003; van der Oost *et al.*, 2003).

Biomarkers of exposure indicate that the organism has been exposed to contaminants, but detection of exposure is not necessarily related to adverse biological consequences. Biomarkers of effect are, however, indicators of biological responses translating into pathological conditions for the organisms.

The suite of biomarkers selected for the present study were selected for their relevance to petroleum exposure and are recognised internationally as the most valuable fish biomarkers for environmental risk assessment (van der Oost *et al.*, 2003). These biomarkers are:

Liver Detoxification Enzymes

Liver detoxification enzymes are part of a detoxification system found in most animals. This enzymatic detoxification system has evolved because organisms have had to deal with foreign compounds since life began on Earth. Liver detoxification enzymes are present with relatively low activity in all animals; however, if the organism is exposed to bioavailable xenobiotics, the enzymatic activity increases significantly to enhance the degradation through oxidation and clearance of the offending chemicals. The enzymatic activity is highly induced within days of exposure, and usually returns to baseline levels within days (4 to 7 days) following the disappearance of the contaminant source (Gagnon and Holdway, 2000). In fish, detoxification enzymatic activity is measured in the liver. Of the large family of enzymes, ethoxyresorufin-O-deethylase (EROD) activity is the most commonly used enzyme to measure detoxification by the liver. EROD activity is a biomarker of exposure to xenobiotics.

Biliary Metabolites

Since the elimination of PAHs is generally very efficient in fish, no long-term bioaccumulation of these compounds has generally been demonstrated. PAH tissue levels are, therefore, not indicative of the levels to which the animals were exposed and cannot be used as bioaccumulation markers for exposure assessment (van der Oost *et al.*, 2003).

Table 1. Classification of physiological indices and biomarkers included in this study into indicators of exposure or effect.

	Parameter	Organ	Indication	
			Exposure	Effect
Physiological Indices	Condition Factor (CF)	Body		X
	Liver Somatic Index (LSI)	Liver		X
	Gonado-Somatic Index (GSI)	Gonad		X
	Gonad Histology	Gonad		X
Biomarkers	EROD activity	Liver	X	
	Biliary Metabolites	Bile	X	
	Sorbitol Dehydrogenase (SDH) activity	Liver		X
	Oxidative DNA damage	Body ¹		X

¹ Although oxidative DNA damage was measured in the bloodstream it can reflect damage that occurred in any organ of the body.

In order to assess the exposure of fish to petroleum hydrocarbons, it is more appropriate to determine PAH metabolite levels in the biliary secretions, as PAH metabolites can accumulate in the bile at levels 1000-fold more concentrated than in the surrounding waters (Hellou and Payne, 1987; Meador *et al.*, 1995).

PAH bile metabolites result from the metabolism of multi-ring compounds by the liver enzymes. By metabolising PAHs to make them water-soluble e.g. blood soluble, PAH metabolites are re-directed to the biliary secretions for eventual elimination out of the body via the intestinal route, avoiding accumulation of these compounds in the body. The appearance of metabolites in the bile is slightly delayed relative to EROD activity induction; therefore, the presence of biliary metabolites reveals exposure of fish which occurred in the past weeks (1-2 weeks; Gagnon and Holdway, 2000). By collecting the biliary secretions, biliary PAH metabolites can be quantified and their presence can confirm exposure to petroleum hydrocarbons. This biomarker is considered as an extremely sensitive marker of exposure to petroleum compounds.

Sorbitol Dehydrogenase (SDH)

In the liver, sorbitol dehydrogenase (SDH) is involved in the conversion of fructose to sorbitol. Normally, SDH concentration is negligible in the bloodstream, but its detection in the serum indicates that hepatocytes have experienced chemical injury causing cellular death and consequently, the cytoplasmic content of liver cells has been released to the bloodstream.

The detection of SDH activity in the bloodstream is, therefore, a biomarker of effect. Fish livers with cellular injuries related to xenobiotic exposure are less capable of performing metabolic activities such as contaminant processing (Holdway *et al.*, 1994). SDH activity is not affected by conditions such as reproductive status which often are confounding factors in the interpretation of other biomarkers. Under xenobiotic stress an increase in SDH can be identified in 3-7 days and can be maintained at an increased level for a prolonged period.

Oxidative DNA Damage

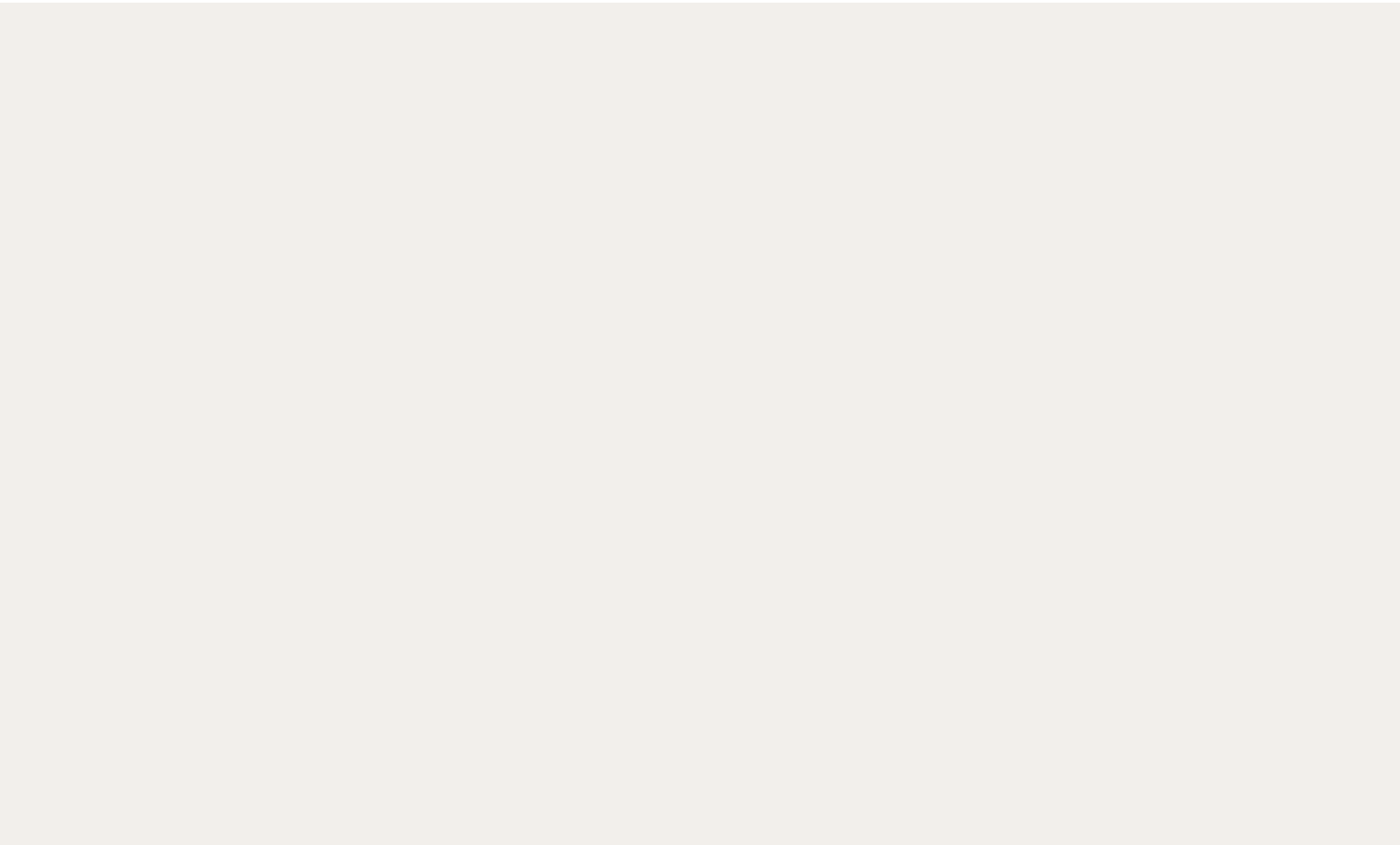
Oxidative DNA damage is the result of the formation of reactive oxygen species (ROS) in the body of an animal which can be increased under exposure to certain contaminants including metals and PAHs (Valavanidis *et al.*, 2009). These ROS are able to react with particular DNA bases forming adducts which can be removed by natural repair mechanisms. However, where the amount of oxidative damage done to the DNA exceeds the capacity of natural repair mechanisms damaged DNA molecules may lose the ability to trigger the synthesis of functional enzymes, proteins and hormones, affecting the entire homeostasis of the organism and potentially challenging reproduction and long term survival of an animal (Cooke *et al.*, 2003). Where an oxidative adduct is formed at a particular base (guanine) the concentration of the excretion of the excised adduct (8-oxo-deoxyguanosine or 8-oxo-dG) can be measured as a representation of the oxidative damage to DNA which has occurred. Oxidative DNA damage is classified as a biomarker of effect as it represents a biologically relevant adverse effect on the organism.

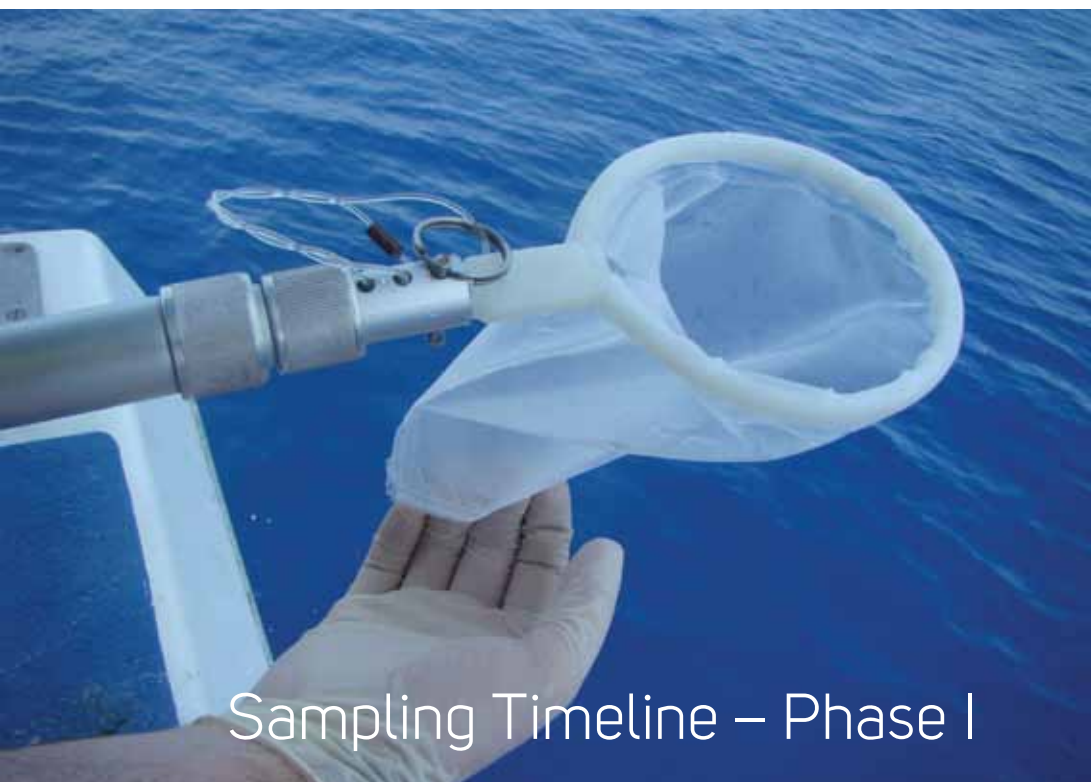


Project Aims



- To describe any exposure of the commercially important Timor Sea demersal and pelagic fishes to petroleum hydrocarbons.
- To evaluate if fish health, including reproductive health, is affected by exposure to petroleum hydrocarbons.
- To investigate temporal variations in the biological responses of fish to petroleum hydrocarbons following the control of the Montara well release.





Sampling Timeline – Phase I

- | | |
|--------------------------------|---|
| 5 th November 2009 | 1400hrs: Monique Gagnon (MG) and Christopher Rawson (CR) (Curtin University) arrived in Darwin. Delays were experienced with the transport of field material, making departure not possible on this day. Visited the FV Megan M and met the crew. |
| 6 th November 2009 | 1800hrs: MG and CR departed Darwin Harbour aboard the FV Megan M with 3 crew (Grant Barker, Shane Ross, Beau Pieterman). |
| 7 th November 2009 | Steam west toward sampling sites south of the West Atlas drilling rig. |
| 8 th November 2009 | 2350hrs: Arrive at Site 1 (20 NM south of West Atlas drilling rig) in late evening and set traps for morning collection. |
| 9 th November 2009 | 0600hrs: Lift traps and collect biopsies on the demersal species goldband snapper, red emperor and saddletail seaperch (28 fish total).
1700hrs: Steam to Site 2 (2 NM south of West Atlas drilling rig). |
| 10 th November 2009 | 0600hrs: Lift traps and collect biopsies on demersal species (39 fish total). Hydrocarbon sheen present on water surface. Biopsies taken on trawled Spanish mackerel (1 fish) |
| 11 th November 2009 | 0600hrs: Steam west to Vulcan Shoal (Site 3 – 20 NM from West Atlas drilling platform) and trawl for pelagic species. Biopsies collected on, dogtooth tuna, bigeye trevally, rainbow runner sampled (3 fish total). Hydrocarbon sheen on water at this site.
1100hrs: Lift traps for red emperor only (increase catch for this species only) (4 fish total). Steam to south to Heywood Shoal (Site 4 – 40 NM from West Atlas drilling rig). Biopsies collected on trawled rainbow runner and dogtooth tuna (4 fish total). |

- 12th November 2009 0600hrs: Trawl for pelagic species at Heywood Shoal (Site 4). Biopsies collected on rainbow runner, Spanish mackerel and dogtooth tuna (13 fish total).
1200hrs: Traps lifted near Heywood Shoal (Site 4.1) and water samples taken. Biopsies collected on Saddletail seaperch and red emperor (21 fish total).
Observations around Heywood Shoal of suspended brown particles and white to yellow floating wax. Steam south to reference area around Browse Island.
Trawl for pelagics. Biopsies collected on rainbow runner (2 fish total).
- 13th November 2009 0630hrs: Traps set 6NM south-west of Browse Island and demersal samples taken on goldband snapper and red emperor only (39 fish total).
1600hrs: Trawl for pelagic around Browse Island. Biopsies collected on rainbow runner (1 fish total).
- 14th November 2009 0700hrs: Traps set for red emperor at 6 NM south-west of Browse Island. Biopsies collected on red emperor (11 fish total).
Stream north to Echuca Shoal.
- 15th November 2009 0600hrs: Trawl for pelagics at Echuca Shoal (Site 6). Biopsies collected on rainbow runner, narrow-barred Spanish mackerel, dogtooth tuna (11 fish total).
1300hrs: Traps set for additional red emperor. Biopsies collected on 2 red emperor.
- 16th November 2009 0630hrs: Trawl for pelagic at Echuca Shoal. Biopsies collected on rainbow runner and Spanish mackerel (2 fish total).
0730hrs: Trap set for additional red emperor (1 fish total).
0930hrs: Steam north toward Site 1 and Site 2 for additional sampling.
2230hrs: Steam toward Darwin.
- 17th November 2009 Steaming toward Darwin.
- 18th November 2009 Steaming toward Darwin.
- 19th November 2009 0500hrs: Arrive Darwin Harbour.
1800hrs: Disembark FV Megan M.



Sampling Timeline – Phase II

5th March 2010	1400hrs: MG and CR arrived at fishermans harbour (Duckpond), Darwin and boarded the FV Megan M. Timing of tides made refuelling and departure impractical until following morning. 1800hrs exit Duckpond through tide lock and moored alongside Fishermans Wharf for refuelling overnight.
6th March 2010	0530hrs: Megan M departs from Fishermans Wharf, Darwin. Aboard: Scientific team: MG and CR (Curtin University); FV Megan M skipper Shane Ross; FV Megan M crew Matt Batty, Beau Pieterman. Steam due west.
7th March 2009	Arrival at Heywood Shoal. Set and lift traps for demersal fishes and trawl for pelagic. Water samples taken. Continue steaming west toward Scott Reef.
8th March 2009	1100hrs: Arrive at Scott Reef. Set and lift traps for demersal fishes and trawled for pelagic in the evening. 2200hrs: Anchor at Scott Reef.
9th March 2010	0630hrs: Scott Reef: Trawl for pelagic fishes and set traps for demersal fishes. 1400hrs: Depart Scott Reef. Steam east toward Browse Island.
10th March 2010	0720hrs: Arrive Browse Island and set traps for demersal fishes. Trawl for pelagic fishes around island. 1900hrs: Steam NE toward Echuca Shoal.
11th March 2010	0630hrs: Trawl for pelagic fishes. Steam toward Heywood Shoal.
12th March 2010	0700hrs: Trawl for pelagic species at Heywood Shoal. 1200hrs: Traps set for demersal fishes and lifted near Heywood Shoal. Steam to location 20 NM south of the West Atlas drilling rig.

13th March 2010	0630hrs: Traps set for demersal fishes 20 NM south of the West Atlas drilling platform. 1930hrs: Anchor 2NM from West Atlas rig.
14th March 2010	0700hrs: Traps set for demersal fishes at 0.5 – 2 NM from the West Atlas drilling rig.
15th March 2010	0600hrs: Traps set for demersal fishes and lifted. Completion of scientific collection. 1700hrs: Steam east toward Darwin.
16th March 2010	Steaming toward Darwin.
17th March 2010	Steaming toward Darwin. 2200hrs: Arrive Darwin.
18th March 2010	Enter Duckpond through tide lock and disembark FV Megan M.



Sampling Timeline – Phase III

- 7th November 2010 1300hrs: MG and CR (Curtin University) arrive in Darwin from Perth. 1350hrs: L. Cooper arrives Darwin from Melbourne via Alice Springs. Travel to Duckpond to meet crew (Shane Ross, Matt Badart, Mitch Seelander, Leif Cooper) of FV Megan M. Due to mechanical alterations to Megan M departure is delayed.
- 8th November 2010 Due to mechanical alterations to Megan M departure is further delayed.
- 9th November 2010 1930hrs: Departure from Darwin harbour aboard Megan M.
- 10th November 2010 Steam west toward study sites.
- 11th November 2010 0200hrs: Arrive Site 8. 0700hrs begin trapping for fish sampling at Site 8. This site is in the same location as a site sampled by WA fisheries in Jan 2010 (Study S4B). This site was designated as Site 1 in study S4B. Steamed to next sampling site.
2200hrs: Arrived at Site 9.
- 12th November 2010 0700hrs: begin trapping for fish sampling at Site 9. This site is in the same location as a site sampled by WA fisheries in Jan 2010 (Study S4B). This site was designated as Site 2 in study S4B.
1800hrs biopsy work completed and steam to next sampling site. Arrived 2300hrs.
- 13th November 2010 0700hrs – 1800 hrs: Grid sediment sampling around West Atlas drilling rig by Leeder Consulting.
- 14th November 2010 0700hrs – 1800 hrs: Grid sediment sampling around West Atlas drilling rig by Leeder Consulting.

15th November 2010 0700hrs – 1100 hrs: Grid sediment sampling around West Atlas drilling rig by Leeder Consulting. :
0900hrs: set traps for fish sampling at site 2. Fish biopsy collection (Site 2). Steamed to Site 1. 1800hrs: arrive Site 1.

16th November 2010 0700hrs: set traps for fish sampling (Site 1). Biopsy collection for demersal fish at this site. 1700hrs: fish biopsy collection completed. Steam south toward Heywood Shoal.

17th November 2010 0200hrs: arrive Heywood Shoal. 0600hrs: commence trawling for pelagic fishes (Site 4). 1800hrs: anchor and line fish for demersals.

18th November 2010 0700hrs: set traps for demersal fish sampling at site 4 (Heywood Shoal). Biopsy collection from demersal fish (Site 4). Steam toward Echuca Shoal.

19th November 2010 0100hrs: Arrive at Echuca Shoal. 0600hrs commence trawling for pelagic fishes. Biopsy collection (Site 6) 1900hrs: steam toward Broome for refuel and crew changeover.

20th November 2010 Steam toward Broome.

21st November 2010 0930hrs: Arrive Broome and take on fuel and stores. Crew change Grant Barker embarks, Mitchell Seelander, Leif Cooper disembark.
2030hrs: depart Broome. Steam toward Scott Reef.

22th November 2010 Steam toward Scott Reef.

23th November 2010 0300hrs: Arrive Scott Reef (Site 7). 0600hrs: Traps set for demersal fishes. 0700hrs: Commence trawling for pelagic fish. Biopsy collection Site 7. 1900hrs anchor at Scott Reef.

24th November 2010 0600hrs traps set for demersal fishes.
0700hrs Commence trawling for pelagic fishes. Collection of biopsies throughout the day as fish are captured.
1800 depart Scott Reef and steam toward Browse Island.

25th November 2010 0400hrs: Arrive Browse Island (Site 5).
0700hrs set traps for demersal fishes. Biopsy collection from demersal fish (Site 5).
Steam toward Echuca Shoal.

26th November 2010 0100hrs: arrive at Echuca shoal (Site 6). 0600hrs commence trawling for pelagic fishes. Biopsy collection (Site 6). Steam toward Heywood Shoal.
2300hrs: arrive Heywood Shoal.

27th November 2010 0600hrs: commence trawling for pelagic fishes. Biopsy collection (Site 4). Steam toward the West Atlas rig (Site 2).

28th November 2010 0400hrs: arrive Site 2. 0600hrs set traps for demersal fish collection. Biopsy collection (Site 2). 1200hrs: Steam toward Site 1 (20 NM from rig).
1400hrs set traps for demersal fish collection. Simultaneous biopsy collection at Site 1.
2340hrs. Completion of biopsy collection. Steam toward Darwin.

29th November 2010 Steam toward Darwin.

30th November 2010 Steam toward Darwin. 2300 hrs: arrive Darwin.

1st December 2010 Disembark FV Megan M.



Fish sampling sites were designated as “Impacted” or “Reference”. Based on information available at the time of the November 2009 sampling, reference sites were selected not less than 80 NM south west of the West Atlas drilling rig. The locations of the sampling sites were originally selected based on the extent, location and direction of hydrocarbon sheen and/or surface (wax) residue (based on AMSA satellite imagery and ship and aircraft observations) provided by DEWHA and PTTEP Australasia. Information provided subsequent to the Phase I sampling suggested that the well release had affected areas outside of the 80 NM radius making the selection of a reference area an increased challenge for Phase II sampling. However, by re-sampling the same sites and an additional reference site at Scott Reef it was expected that any temporal changes in impact will be observed.

Between the Phase I and II sampling events a collection of red emperor and goldband snapper was made by WA Fisheries in a parallel study as part of the Montara Monitoring Program. This study utilised two of the impacted sites sampled during Phases I and II of Study S4A (Sites 1 and 2) and selected 2 reference sites 75 and 54 NM to the north-east of the West Atlas drilling rig (Sites 8 and 9 respectively). These additional sites were therefore added to the sampling program for Study S4A Phase III. These sites were within 80 NM of the rig but had been previously designated as reference by the Department of Fisheries Western Australia so remained “reference” sites for these studies. Site 3 was not re-sampled during Phase II and Phase III due to the lack of habitat for the target species and consequently, the absence of fish at this site.

Further, sites were selected based on water depth (shallow reef <30m, or deep sea > 70m) and sea floor structure (hard or soft bottom) which allowed for the reasonable expectation of consistency of catch diversity and number of fish. Sites 1, 2, 3 and 4 were located within the impacted area, Sites 5 and 6 were located within the reference area and Site 7 was located at Scott Reef (also a reference site). Sites 1, 2 were deep sea sites where demersal fishes were targeted and Sites 3, 4, and 6 were shallow reef sites where pelagic fishes were targeted. Site 5 incorporated shallow reef around Browse Island and a nearby deep sea site.

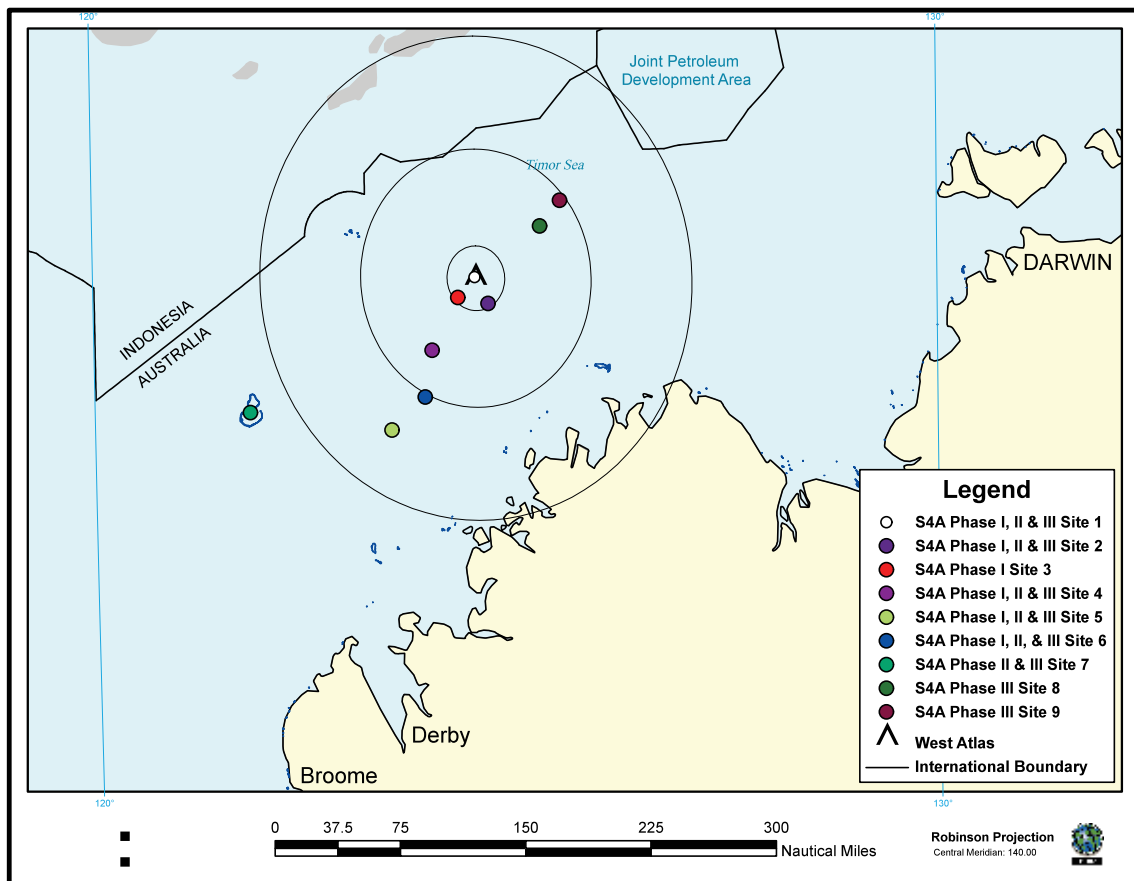
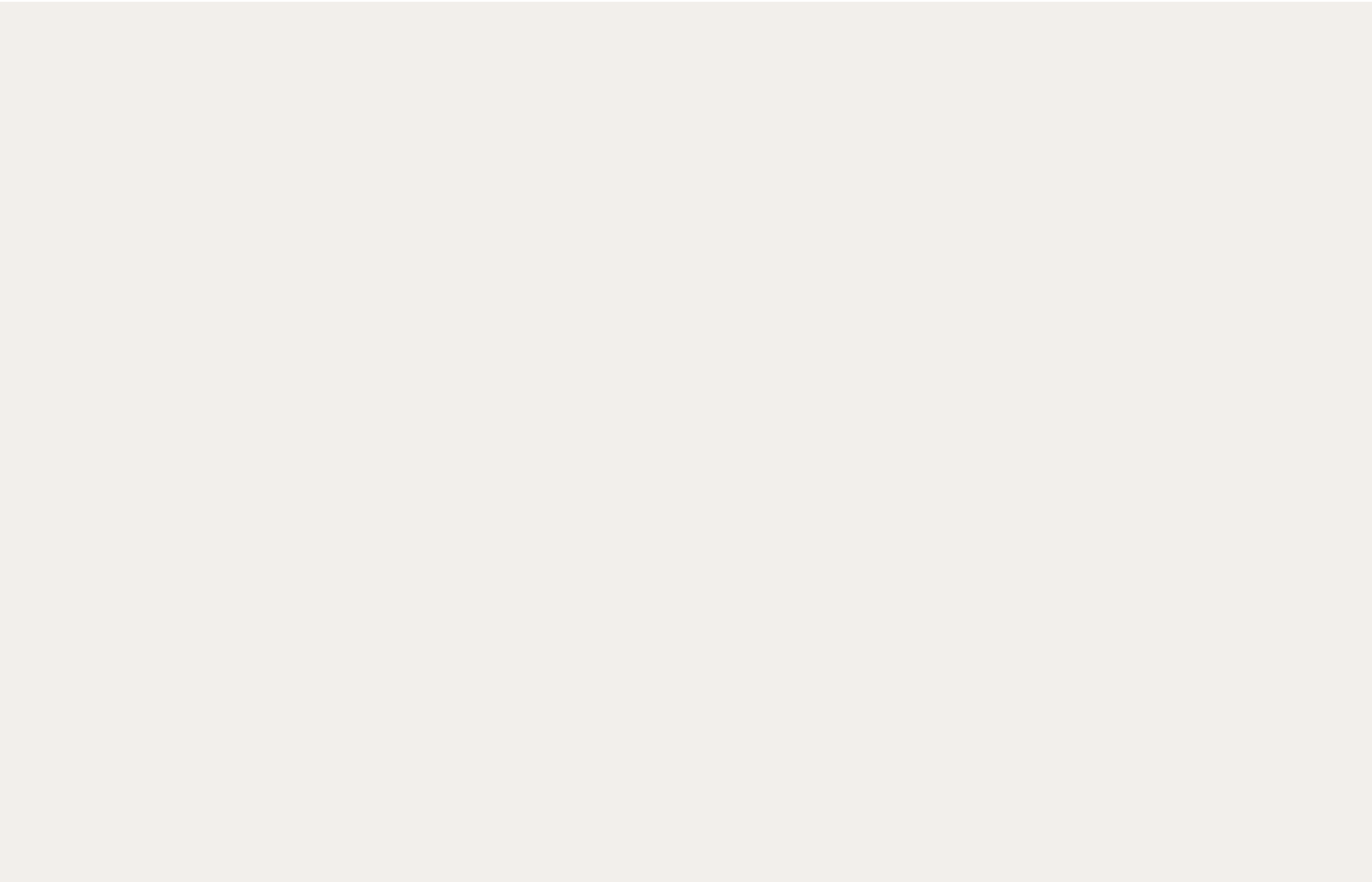


Figure 1. Map showing the location of the West Atlas drilling rig and the sampling sites. Concentric rings represent 20, 80 and 150 NM from the platform. Sites 1, 2, 3 and 4 were designated as impacted sites and Sites 5 and 6 were designated as reference sites in Phase I sampling. Site 7 is located at Scott Reef. Surface samples are plankton net trawls collecting observed floating material.

Table 2. Details of S4A Phase II sampling sites.

		Location		Distance from West Atlas rig (NM)	Approx Depth (m)
Site		Lat (°)	Long (°)		
1	Site 2	12.9450 S	124.6900 E	< 1	70
2	Site 1	12.6697 S	124.5345 E	20	75
4	Heywood Shoal	13.4305 S	124.0173 E	53	25 – 100
5	Browse Island	14.2563 S	123.5255 E	82	15 – 30
6	Echuca Shoal	13.9152 S	123.9278 E	110	0 – 120
7	Scott Reef	14.0763 S	121.8507 E	177	0 – 400
8	WAF Site 1	11.8725 S	125.5530 E	75	80
9	WAF Site 2	12.1402 S	125.3135 E	54	75





Field Methodology

Fish Sampling

The methods of fish capture were similar in all phases of the investigation. Demersal fishes were collected from 70-120m depth using baited stainless steel fish traps (sampling at Scott Reef was at around 40m since the reef has no suitable fishing grounds at the above preferred depth). The traps were dropped at the sampling locations and left for between 1 and 12 hours. Two demersal species were targeted: goldband snapper (*Pristipomoides multidens*) and red emperor (*Lutjanus sebae*). The depth and bottom characteristics at Scott Reef were not suitable habitat for these species and none were collected in the traps set at this site. The predominant species captured at this site was the longnose emperor (*Lethrinus olivaceus*). Biopsies were therefore collected on this species at this site but not analysed as this species has not been captured at other sites. Pelagic fishes were captured by trawling 3 single lure lines behind the fishing vessel travelling at low speed across shallow (< 20 m) shoals. It was hoped that Spanish mackerel (*Scomberomorus commerson*) could be used as a bioindicator but, given the low number of individuals collected, it was decided to focus on the rainbow runner (*Elegatis bipinnulata*) which was captured in larger numbers. Despite the low number of individuals captured during the various sampling phases, results for both Spanish mackerel and rainbow runner are presented in this report.



Figure 2. Method of demersal fish capture. Baited steel fish trap prior to deployment. In picture: Matthew Badart (left) and Beau Pieterman (right).



Figure 3. Left: flow-through live tank used to preserve the fish in good condition following capture; right: fish were initially measured and weighed prior to sacrifice.

Morphology

The fish captured at each sites were kept alive in live tanks equipped with flow through, as biopsies need to be collected on freshly scarified animals. Fish were initially measured for length (using a mm scale ruler) and for weight using an electronic spring balance. Each fish was examined for external abnormalities including lesions or excessive fin damage.

Biopsies Collected

Fish captured for biopsy collection (approximately 20 of each target species from each site) were sacrificed by *iki jimi* (spike through the brain) and a vacuutainer and needle were used to collect blood from the caudal vein. These blood samples were allowed to coagulate at 4°C for up to 20 minutes and then centrifuged at 2400 x *g* for 10 mins and the serum supernatant divided into two samples: one of which was frozen at – 20°C and the other placed in liquid nitrogen. The fish was then dissected along the ventral line and inspected internally. Many of the fish were significantly infested with parasites in the body cavity. In red emperor there were mainly nematodes while in goldband snapper most of the parasites were encapsulated and adhesive to body organs (liver, gonads, digestive tract).

The following biopsies were collected:

1. Serum samples (see above);
2. Bile was collected from the gall bladder using a 1 mL syringe and frozen at – 20°C;
3. The liver was removed, weighed and subsamples frozen in liquid nitrogen for analysis;

4. The gonads were removed and weighed.
Where available the gonads of 10 male and 10 female fish of each species from the impacted area and reference area were preserved in glutaraldehyde for histology;
5. Carcass (body less viscera) was weighed using the electronic spring balance.

Additional Sampling

Muscle Samples

Ten of the following samples were collected on each target species from both the impacted and reference areas:

1. Small muscle sample (approx. 200 g) for analysis of total petroleum hydrocarbons;
2. Larger muscle sample (approx. 400 g) for olfactory testing for taint (edible species only).

In Phase I of the study, muscle tissue was collected for total petroleum hydrocarbon (TPH) analysis and olfactory assessment on 69 demersal fish and 31 pelagic fish. In Phase II of the investigation, a total 130 muscle samples were collected for TPH analysis and olfactory assessment, with 87 demersal fish and 43 pelagic fish sampled. Finally, in Phase III 121 muscle samples were collected from demersal fish and 23 from pelagic fish. Muscle samples collected for analysis of TPH were transferred frozen to WA Fisheries for holding/analysis.

Muscle samples collected for the sensory analysis of taint have been held frozen at Curtin University Aquatic Toxicology laboratories. A random sample of muscle samples from Phases I and II have been used in olfactory testing for taint. This study was completed in March 2011.



Figure 4. Left: blood collection from the caudal artery on a goldband snapper; right: bile collection using a 1 mL syringe.

Stomach/Intestinal Contents

Stomach and intestinal contents were collected from 10 of each species at each location where available. The stomach contents of the demersal fish were biased due to the bait placed in the cages. Consequently, the stomach contents were collected for demersals only if it was identified as 'other than bait'. The more relevant intestine contents, if any, of the demersal fish was collected when the stomach was empty. For the pelagic fish, the stomach contents as well as intestine contents were collected from all fish.

Stomach/intestinal contents were collected for 69 demersal fish and 23 pelagic fish in Phase I of the study (92 samples total). In Phase II, stomach/intestinal contents were collected for 87 demersal fish and 28 pelagic fish (115 total). The stomach/intestinal contents were transferred frozen to WA Fisheries for holding/analysis. Results will be presented in a report produced by the Department of Fisheries, Western Australia, to be submitted in 2011 to the Commonwealth Department of Sustainability, Environment, Water, Population and Communities (SEWPaC).

Table 3. Numbers of demersal fish from which stomach/intestine content and muscle for TPH analyses were collected during Phase I of the study.

		Goldband Snapper		Red Emperor		Saddletail Seaperch	
		Gut	TPH	Gut	TPH	Gut	TPH
Impacted	1	13	17	7	7	3	4
	2	8	10	8	4	3	4
	3	0	0	0	0	0	0
	4	0	0	2	1	2	2
	Total	21	27	17	12	8	10
Reference	5	10	10	13	10	0	0
	6	0	0	0	0	0	0
	Total	10	10	13	10	0	0
Grand Total		31	37	30	22	8	10

Table 4. Numbers of pelagic fish from which stomach/intestine content and muscle for TPH analyses were collected during Phase I of the study.

		Rainbow Runner		Spanish Mackerel		Dogtooth Tuna	
		Gut	TPH	Gut	TPH	Gut	TPH
Impacted	1	0	0	0	0	0	0
	2	0	0	1	1	0	0
	3	0	1	0	0	1	1
	4	6	10	1	1	3	3
	Total	6	11	2	2	4	4
Reference	5	1	1	0	0	0	0
	6	7	8	3	4	0	1
	Total	8	9	3	4	0	1
Grand Total		14	20	5	6	4	5

Table 5. Numbers of demersal fish from which stomach/intestine content and muscle for TPH analyses and taint assessment were collected during Phase II of the study.

	Site	Goldband Snapper			Red Emperor			Longnose Emperor		
		TPH	Taint	Gut	TPH	Taint	Gut	TPH	Taint	Gut
Impacted	1	11	11	10	9	8	9	0	0	0
	2	10	10	10	9	9	9	0	0	0
	3	0	0	0	0	0	0	0	0	0
	4	10	10	6	10	10	10	0	0	0
	Total	31	31	26	28	27	28	0	0	0
Reference	5	8	9	10	10	10	13	0	0	0
	6	0	0	0	0	0	0	0	0	0
	7	0	0	0	0	0	0	10	10	10
	Total	8	9	10	10	10	13	10	10	10
Grand Total		39	40	36	38	37	41	10	10	10

Table 6. Numbers of pelagic fish from which stomach/intestine content and muscle for TPH analyses and taint assessment were collected during Phase II of the study.

	Site	Rainbow Runner			Spanish Mackerel			Dogtooth Tuna		
		TPH	Taint	Gut	TPH	Taint	Gut	TPH	Taint	Gut
Impacted	1	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0
	3	0	0	0	0	0	0	0	0	0
	4	12	11	11	3	3	2	1	0	1
	Total	12	11	11	3	3	2	1	0	1
Reference	5	1	1	1	0	0	0	0	0	0
	6	10	10	9	2	2	1	0	0	0
	7	5	5	1	0	0	0	9	0	2
	Total	16	16	11	2	2	1	9	0	2
Grand Total		28	27	22	5	5	3	10	0	3

Table 7. Numbers of demersal fish from which stomach/intestine content and muscle for TPH analyses and taint assessment were collected during Phase III of the study.

	Site	Goldband Snapper			Red Emperor		
		TPH	Taint	Gut	TPH	Taint	Gut
Impacted	1	10	10	10	10	10	10
	2	10	10	10	10	10	10
	3	0	0	0	0	0	0
	4	10	10	10	9	9	9
	Total	30	30	30	29	29	29
Reference	5	10	10	10	10	10	10
	6	0	0	0	0	0	0
	7	0	0	0	0	0	0
	8	10	10	11	11	11	13
	9	11	11	10	9	9	8
	Total	31	31	31	30	30	31
Grand Total		61	61	61	61	59	60

Table 8. Numbers of pelagic fish from which stomach/intestine content and muscle for TPH analyses and taint assessment were collected during Phase III of the study.

		Rainbow Runner			Spanish Mackerel		
	Site	TPH	Taint	Gut	TPH	Taint	Gut
Impacted	1	0	0	0	0	0	0
	2	0	0	0	0	0	0
	3	0	0	0	0	0	0
	4	3	3	3	13	13	10
	Total	3	3	3	13	13	10
Reference	5	0	0	0	0	0	0
	6	0	0	0	4	4	3
	7	3	3	3	0	0	0
	8	0	0	0	0	0	0
	9	0	0	0	0	0	0
	Total	3	3	3	4	4	3
Grand Total		6	6	6	17	17	17

Floating Material Samples

The presence of floating residues possibly related to oil weathering has been reported, and concerns were raised that the fish may mistakenly confuse the residues for food. Floating material was sighted and collected only in Phase III. Floating material was collected using the preferred method of a Teflon net attached to a long sampling pole and preserved for analysis. Samples of floating material collected were frozen and transported to Leeder Consulting, Melbourne.

Traces of fatty acids found in some floating material samples, and these fatty acids have been identified as being of biogenic origin. In November 2009, two samples returned positive identification of weathered crude oil. No hydrocarbons of petroleum origin were identified in other samples. Results are presented in Appendix E.

Water Samples

Water samples were collected at each of the fish collection sites and whenever surface residues were observed during Phase I and Phase II and sent to Leeder Consulting (Melbourne). Collections were made using a stainless steel bomb sampler (a van Dorne type sampling device necessary for water analysis if the presence of petroleum hydrocarbons is suspected) at the surface and at depths of 1, 3, 5, 10, and 15 m. No petroleum hydrocarbons have been identified in any of the samples. Results are presented in Appendix E.

Sediment Samples

Sediment was collected during Phase III using standard dredge grab sampling techniques. Surface sediment samples were collected over a 2km x 2km grid covering an 8-km radius around the location of the West Atlas rig. Sediment grabs were also collected at each of the locations where fish sampling occur. A small (3 kg) dredge sampler was lowered on a rope until it hit the bottom where a spring loaded trigger mechanism closed the mouth of the dredge. The winch on board the Megan M was used to raise the dredge onto the boat where the dredge was opened and the contents removed into a rinsed glass sample jar. These jars were refrigerated prior to transport for analysis. Hydrocarbons of petroleum origin could not be found in any of the 57 sediment samples collected. Results are presented in Appendix F.



Laboratory Methods

Physiological Parameters

The condition factor (k) of the fish was calculated as:

$$k = \left[\frac{W_g}{L_f^3} \right] \times 10^6$$

where W_g is the gutted weight of the fish and L_f is the fork length. Condition factor gives an indication of the health status, or 'fattiness' of the animal.

The liver somatic index (LSI) was calculated as:

$$LSI = \left[\frac{W_L}{W_g} \right] \times 100$$

where W_L is the liver weight, and gives an indication of the size of the liver relative to the body size.

Similarly the gonado-somatic index (GSI) was calculated as:

$$GSI = \left[\frac{W_G}{W_g} \right] \times 100$$

where W_G is the weight of the gonad. GSI is a measure of the fish's reproductive investment.

Liver Detoxification Enzymes (EROD Activity)

In the laboratory, a liver homogenate is centrifuged to isolate the microsomes containing the detoxification enzymes. These microsomes are then required to metabolise a model contaminant: ethoxyresorufin. The enzyme metabolising ethoxyresorufin is therefore named 'ethoxyresorufin-O-deethylase' (EROD). The metabolism of ethoxyresorufin by EROD enzyme activity results in a fluorescent product, resorufin, which can be measured by fluorimetry at excitation wavelength 530 nm and emission wavelength 585 nm (Hodson *et al.*, 1991). A higher fluorescence indicates an increased abundance of enzyme that results from the fish assimilating and metabolising high levels of contaminants. The enzyme activity is reported by unit of protein in the isolated microsome fraction, which normalizes the data according to the protein density in the liver tissue.

Biliary Metabolites

After being processed by liver detoxification enzymes, metabolites are predominantly eliminated via the bile. However, the bile turnover and by consequence the concentration of biliary metabolites is influenced by food intake. Therefore, this biomarker is relevant for recent (weeks) exposure to xenobiotics. The bile of the fish was collected using a 1 ml syringe, and immediately frozen in liquid nitrogen until biliary metabolites determination by fixed-wavelength fluorescence (FF) measurement (Lin *et al.*, 1996). The method is semi-quantitative, and reports metabolised PAHs as 'type of metabolites'. The expression 'type of metabolites' refers to the various aromatic compounds, most occurring as conjugated metabolites of PAHs, that are measured in the bile at naphthalene-, pyrene-, or benzo(a)pyrene – (B(a)P-) specific excitation/emission wavelengths. Fluorescent readings were performed for naphthalene-type metabolites at excitation/emission 290/335 nm using 1-naphthol (Sigma) as a reference standard. For pyrene-type metabolites and B(a)P-type metabolites, readings were made using 1-hydroxy pyrene (1-OH pyrene) as a reference standard at 340/380 nm and 380/430 nm for pyrene – and B(a)P-type metabolites, respectively. Naphthalene-type metabolites are reported in mg of 1-naphthol fluorescence units equivalent per mg biliary protein, and pyrene – and B(a)P-type metabolites are reported in µg of 1-OH pyrene fluorescence units equivalent per mg biliary protein. Therefore, the biliary metabolite levels observed represent fluorescence-equivalents of PAH metabolites used as standard.

Sorbitol Dehydrogenase (SDH) Activity

A blood collection was performed via the caudal artery using a vacutainer. The blood was allowed to clot on ice for 15 minutes after which it was centrifuged in a refrigerated centrifuge at 3000 g for 10 minutes. The serum was immediately collected and frozen at – 80°C until analysis of SDH activity. The quantitation of SDH activity was determined by the reduction in light absorbance at 340 nm over time. SDH activity is reported in milli-International Units (mIU), with one U of an enzyme being defined as the amount of this enzyme which will convert 1 µmol of substrate to product, per minute.

Oxidative DNA Damage (Serum 8-oxo-dG Concentration)

The concentration of the oxidative stress biomarker 8-oxo-7,8-dihydro-2'-deoxyguanosine (8-oxo-dG) in the serum samples was measured using a commercially available enzyme linked immunosorbent assay (ELISA) kit (Trevigen). Briefly, serum samples were diluted (1:5) with the diluent supplied with the kit and a supplied 8-oxo-dG standard was diluted to create a standard curve (0.94 – 60 ng L⁻¹). These were added to a 96 well microplate which was pre-coated with 8-oxo-dG with an 8-oxo-dG monoclonal antibody. After 1 hour incubation the plate was washed 6 times (Bio-Rad plate washer) and a secondary antibody (horseradish peroxidase conjugate) was introduced. After a further 1 hour incubation the plate was again washed (6 times) and the plate developed with a trimethylbenzidine (TMB) substrate and the absorbance (450 nm) was measured (Bio-Rad plate reader) after addition of an acidic stop solution. The intensity of the colour was inversely related to the amount of 8-oxo-dG in the sample or standard. These were standardised against the protein content of each serum sample (Lowry *et al.*, 1951).

Gonad Histology

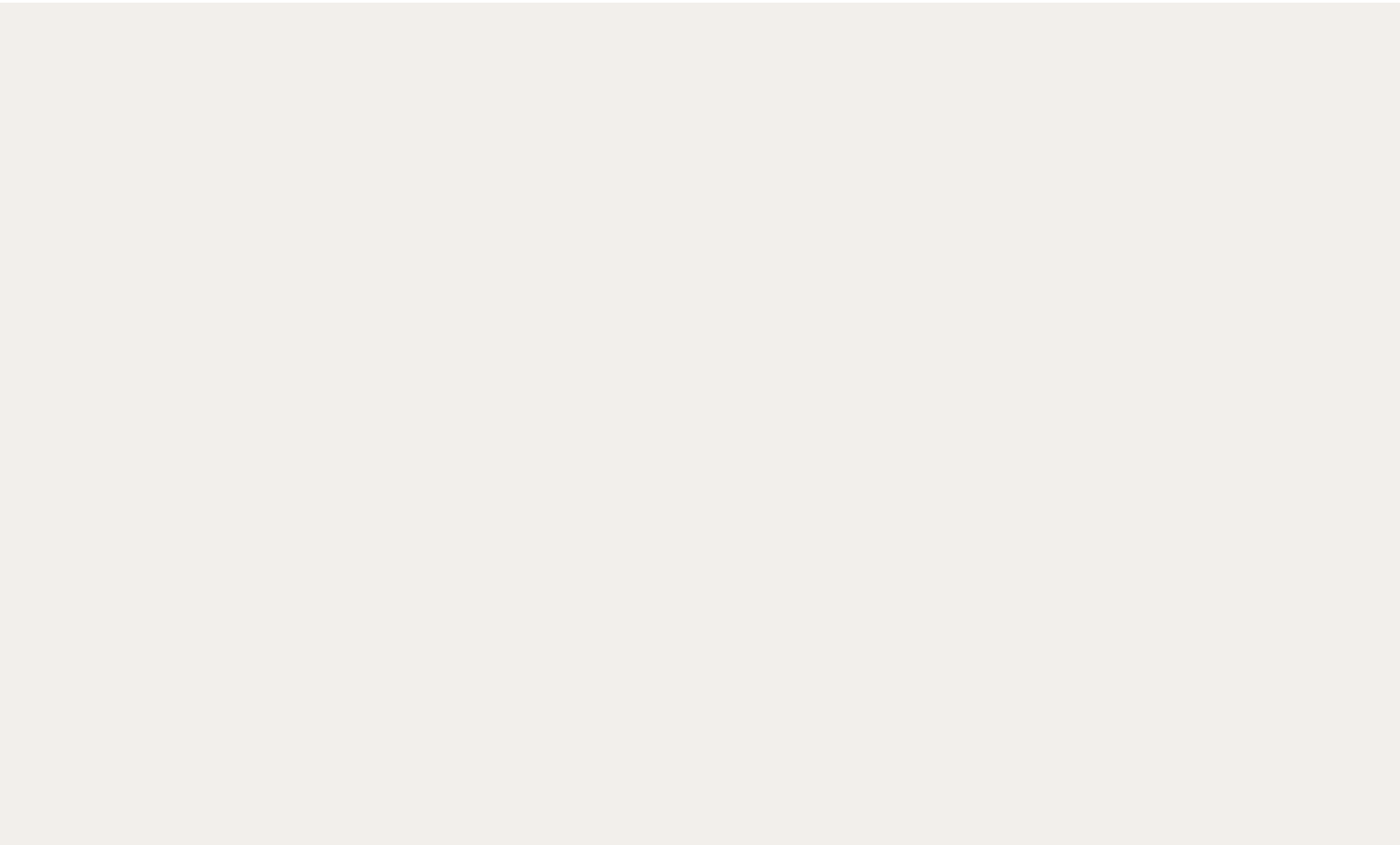
Preserved (glutaraldehyde) male and female gonads were washed and transferred to a 50% ethanol solution for longer term storage. The samples sectioned into 10 mm cross-sections which were further dehydrated by progressively increasing the ethanol concentration over four hours. They were then cleared in xylene for a further three hours before embedding in paraffin wax overnight. These samples were placed in paraffin wax blocks for sectioning (5 µm) using a microtome. These sections were fixed to glass slides and gently heated overnight prior to staining. The slides were stained (Haematoxylin – Eosin) using standard methods (Lillie, 1965). This differential staining procedure allows the identification of relevant structures in the tissues (particularly nuclei and membranes) and the identification of any abnormalities. In gonad tissue it also allows the classification of gametes at different stages of development.

Statistical Analyses

For each variable measured, descriptive statistics including average and standard errors about the mean were generated using the program SPSS ver. 17.0. Physiological parameters condition factor (CF), liver somatic index (LSI) and gonado-somatic index (GSI) as well as biomarker levels were compared between sites using a one-way analysis of variance (ANOVA) after verification of normal data distribution and homoscedasticity. Post-hoc test Tukey's-b was applied to identify specific site differences, if relevant. For all statistical tests, a significance level alpha (α) of 0.05 was applied.

In order to visualise overall trends in data across all biomarkers multivariate techniques were employed. Biomarkers selected for these analyses were SDH, oxidative DNA damage, condition factor, LSI, naphthalene metabolites, pyrene metabolites, benzo(a)pyrene metabolites and EROD activity.

GSI was not included as it is inherently bimodal (male and female modes) and therefore unsuitable. Data for each biomarker were normalised and a similarity matrix constructed using Euclidean distance. An analysis of similarity (ANOSIM) was used to detect differences between the sites and pairwise permutation tests conducted to evaluate where differences lay. Principal Components Analysis (PCA) was used to evaluate the importance of each biomarker in defining the differences between sites (maximum 5 PC) and to visualise the overall trends in the biomarker data. The analyses were split between sampling times and species as inherent, expected differences were likely to bias the results of the analysis and lead to incorrect interpretation. Multivariate analyses were conducted using Primer v6.





Results and Interpretation

Sampling Summary

Diversity

A total of 993 and 1044 demersal fish were captured in the traps during the sampling of Phases I and II respectively. Four taxa of demersal species were regularly captured in the fish traps: goldband snapper, red emperor, saddletail seaperch (*Lutjanus malabaricus*) and cod (various species) (Table 9, Table 10). Other taxa included lionfish, triggerfish and squirrelfish but were not included in the census. The pelagic fish captured were dominated by the narrow-barred Spanish mackerel and the rainbow runner, with totals of 40 and 62 individuals captured during Phase I and II, respectively (Table 9, Table 10). Diversity data were not collected during Phase III as the selection of bioindicator species had been well-determined and justified by the catches in the initial 2 Phases.

Table 9. Diversity and number of each pelagic and demersal taxon captured during the November 2009 Phase I sampling effort.

		Demersal Species					Pelagic Species					
	Site	Goldband Snapper	Red Emperor	Saddletail Seaperch	Cod	Other	Spanish Mackerel	Rainbow Runner	Dogtooth Tuna	Wahoo	Giant Trevally	Total
Impacted	1	146	6	29	0	0	0	0	0	0	0	181
	2	133	25	49	19	2	1	0	0	0	0	229
	3	0	0	0	0	0	1	1	1	0	0	3
	4	100	24	43	8	0	1	13	3	0	0	192
	Total	379	55	121	27	2	3	14	4	0	0	605
Reference	5	115	20	169	43	21	0	3	0	1	0	372
	6	0	1	0	0	0	4	8	1	1	1	16
	Total	115	21	169	43	21	4	11	1	2	1	388
Grand Total		494	76	290	70	23	7	25	5	2	1	993

Table 10. Diversity and number of each taxon captured in demersal traps during the March 2010 sampling effort.

		Goldband Snapper	Red Emperor	Saddletail Seaperch	Paddletail	Longnose Emperor	Cod	Spangled Emperor	Variegated Emperor	Coral Trout	Red Bass	Shark	Total
Impacted	1	118	48	3	0	0	44	26	0	4	0	0	243
	2	113	23	14	0	0	4	0	0	2	0	0	156
	3	0	0	0	0	0	0	0	0	0	0	0	0
	4	80	82	16	0	0	30	50	0	0	0	0	258
	Total	311	153	33	0	0	78	76	0	6	0	0	657
Reference	5	83	25	63	0	0	6	23	0	0	0	0	200
	6	0	0	0	0	0	0	0	0	0	0	0	0
	7	0	0	0	81	44	6	0	23	9	22	2	187
	Total	83	25	63	81	44	12	23	23	9	22	2	387
Grand Total		394	178	96	81	44	90	99	23	15	22	2	1044

Note: Wahoo: *Acanthocybium solandri*; Crocodile longtom: *Tylosurus crocodilus*; Yellowfin tuna: *Thunnus albacares*; Shark mackerel: *Grammatocynus bicarinayus*; Red bass: *Lutjanus bohar*.

Biopsy Collection

Biopsies for analysis at Curtin University (serum, bile, liver and gonad samples) were collected on a total of 181 fish in Phase I, and 262 fish in Phase II and 338 fish in Phase III. Attempts were made

to collect even numbers of male and female fish but this was not always possible. Not all biopsies were analysed as the study focussed on goldband snapper, red emperor, rainbow runner and Spanish mackerel only (Tables 11-16).

Table 11. Numbers of demersal fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase I of the study.

	Site	Goldband Snapper			Red Emperor			Saddletail Seaperch		
		Male	Fem	Total	Male	Fem	Total	Male	Fem	Total
Impacted	1	3	14	17	1	6	7	1	3	4
	2	13	10	23	2	10	12	0	4	4
	3	0	0	0	0	0	0	0	0	0
	4	0	0	0	4	6	10	1	10	11
	Total	24	40	7	22	29	2	17	19	7
Reference	5	15	10	25	4	21	26	0	0	0
	6	0	0	0	1	2	3	0	0	0
	Total	10	25	4	23	28	0	0	0	6
Grand Total		31	34	65	13	48	61	2	17	19

Table 12. Numbers of pelagic fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase I of the study.

		Rainbow Runner			Spanish Mackerel			Dogtooth Tuna		
		Male	Fem	Total	Male	Fem	Total	Male	Fem	Total
Impacted	1	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	1	1	0	0	0
	3	0	1	1	0	0	0	0	1	1
	4	7	6	13	0	1	1	2	1	3
	Total	7	7	14	0	2	2	2	2	4
Reference	5	3	0	3	0	0	0	0	0	0
	6	3	5	8	1	3	4	0	1	1
	Total	6	5	11	1	3	4	0	1	1
Grand Total		13	12	25	1	5	6	2	3	5

Table 13. Numbers of demersal fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase II of the study.

	Site	Goldband Snapper			Red Emperor			Longnose Emp.		
		Male	Fem	Total	Male	Fem	Total	Male	Fem	Total
Impacted	1	12	13	25	6	12	18	0	0	0
	2	6	14	20	14	12	26	0	0	0
	3	0	0	0	0	0	0	0	0	0
	4	13	18	31	9	15	24	0	0	0
	Total	31	45	76	29	39	68	0	0	0
Reference	5	3	18	21	10	13	23	0	0	0
	6	0	0	0	0	0	0	0	0	0
	7	0	0	0	0	0	0	2	12	14
	Total	3	18	21	10	13	23	2	12	14
Grand Total		34	63	97	39	52	91	2	12	14

Table 14. Numbers of pelagic fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase II of the study.

	Site	Spanish Mackerel			Rainbow Runner			Dogtooth Tuna		
		Male	Fem	Total	Male	Fem	Total	Male	Fem	Total
Impacted	1	0	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0	0
	3	0	0	0	0	0	0	0	0	0
	4	0	3	3	10	15	25	1	0	1
	Total	0	3	3	10	15	25	1	0	1
Reference	5	0	0	0	1	0	1	0	0	0
	6	1	1	2	7	6	13	0	0	0
	7	0	0	0	4	1	5	5	5	10
	Total	1	1	2	12	7	19	5	5	10
Grand Total		1	4	5	22	22	44	6	5	11

Table 15. Numbers of demersal fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase III of the study.

	Site	Red Emperor				Goldband Snapper			
		Male	Fem	Imm	Total	Male	Fem	Imm	Total
Impacted	1	13	19	3	35	15	9	0	24
	2	10	17	1	28	16	19	0	35
	4	16	22	0	38	10	13	0	23
	Total	39	58	4	101	41	41	0	82
Reference	5	7	12	1	20	14	17	0	31
	6	0	0	0	0	0	0	0	0
	7	0	0	0	0	0	0	0	0
	8	2	11	4	17	12	12	0	24
	9	8	11	1	20	13	7	0	20
	Total	17	34	6	57	39	36	0	75
Grand Total		56	92	10	158	80	77	0	157

Table 16. Numbers of pelagic fishes from which measurements and/or biopsies (serum, bile, liver and gonad samples) were collected for analysis during Phase III of the study.

	Site	Spanish Mackerel				Rainbow Runner			
		Male	Fem	Imm	Total	Male	Fem	Imm	Total
Impacted	1	0	0	0	0	0	0	0	0
	2	0	0	0	0	0	0	0	0
	4	3	10	0	13	1	2	0	3
	Total	3	10	0	13	1	2	0	3
Reference	5	0	0	0	0	0	0	0	0
	6	0	4	0	4	0	0	0	0
	7	0	0	0	0	0	3	0	3
	8	0	0	0	0	0	0	0	0
	9	0	0	0	0	0	0	0	0
	Total	0	4	0	4	0	3	0	3
Grand Total		3	14	0	17	1	5	0	6

General Fish Health

In all Phases of the investigation, no gross abnormalities or external parasites were observed on the specimen collected, and all fish appeared to be in a healthy status. Internal parasites were observed on most fish, usually cystic worms adhering to the external wall of the stomach or intestines. On a small number of fish, flat worms were noticed on the external liver tissue, but did not seem to injure the hepatic tissue. In all cases, the presence of parasites appeared 'normal' in wild-caught fish, and is not expected to influence the physiological indices or the biomarkers of fish health.



Figure 5. Parasitic cysts adhering to the stomach of a Spanish mackerel (top), and flat worm attached to the liver of a goldband snapper (bottom).

Physiological Parameters

Condition Factor

Condition factor (the weight of the fish adjusted for its length) is a useful initial screening tool providing information on the overall health of an animal.

It can be affected by environmental factors such as nutrition and disease but will also been shown to decrease upon chronic exposure to contaminants. This is the result of increased energy used to metabolise and excrete the contaminants and often with impacts on organ performing these tasks (liver, kidney). A decreased condition factor can have secondary impacts whereby an animal has decreased energy reserves (lipids) to deal with other adverse impacts (reduced food availability, disease, contaminant exposure).

For each of the four target species in Phase I the condition factor was similar in fish collected in reference and in impacted zones. This result suggests that despite the presence of petroleum hydrocarbons in their environment, the fish collected in the impacted zones had an adequate food supply to maintain a body condition similar to the fish originating from the non-impacted areas.

Similarly, goldband snapper, rainbow runner and Spanish mackerel collected from impacted areas in Phase II had condition factors which were not different to those collected from the reference areas. Red emperor had increased condition factors at the site closest to the West Atlas rig (<2 NM). It is possible that the presence of the rig provides an attractive effect for fish including red emperor prey resulting in more abundant food items than those living away from such structures.

In November 2010, the condition factor of goldband snapper and of Spanish mackerel was similar at the impacted and reference sites however, that of red emperor was slightly but significantly reduced at one reference site only (site 8), located at 75 NM from the rig. The condition factor of rainbow runner was also lower at the reference site. In both these cases where the condition factor was reduced at a reference site, it seems reasonable to hypothesise that food abundance might be responsible for the difference. The relatively small variability of this parameter ($\pm 8\%$ for red emperor and $\pm 2\%$ for rainbow runners) enables the establishment of statistical significance, however, it is not unlikely that the small differences observed would be biologically significant for the fish. It can be expected that differences in food abundance would exist between sites, and this can result in the condition factor being naturally variable between groups of fishes.

Condition factor is a general indicator of fish health and might be reduced in situations where the impacts of contaminants is extreme, reducing the availability of food or impairing the food consumption by the fish. Although condition factor is neither contaminant-specific nor very sensitive to short-term contamination events, it does provide information on the general condition of the fish and is considered a valuable initial screening tool in evaluation of pollutant impacts (Van der Oost *et al.*, 2003).

Overall, no biologically significant reduction in condition factor, in any of the fish species, was consistently observed for up to a year following the end of the Montara well release.

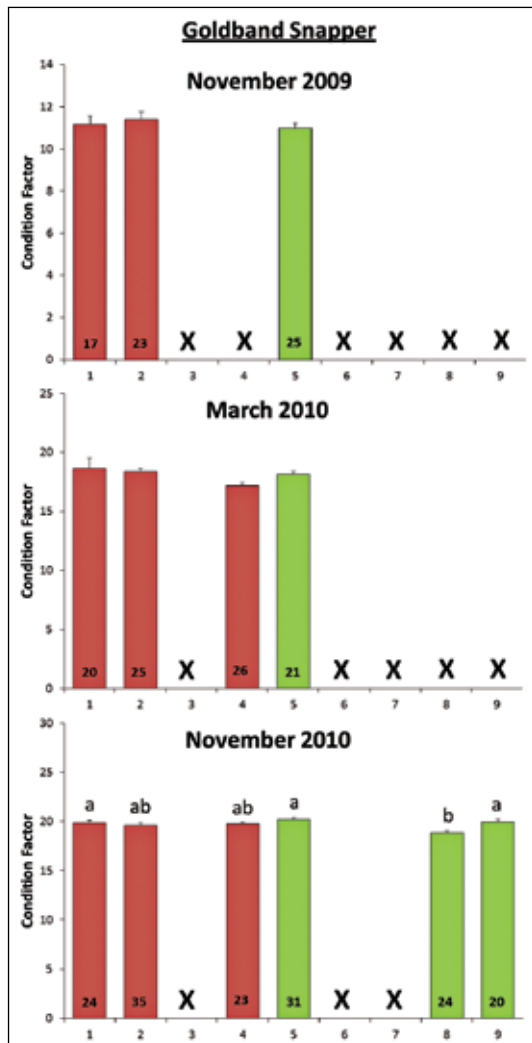


Figure 6. Condition factor of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

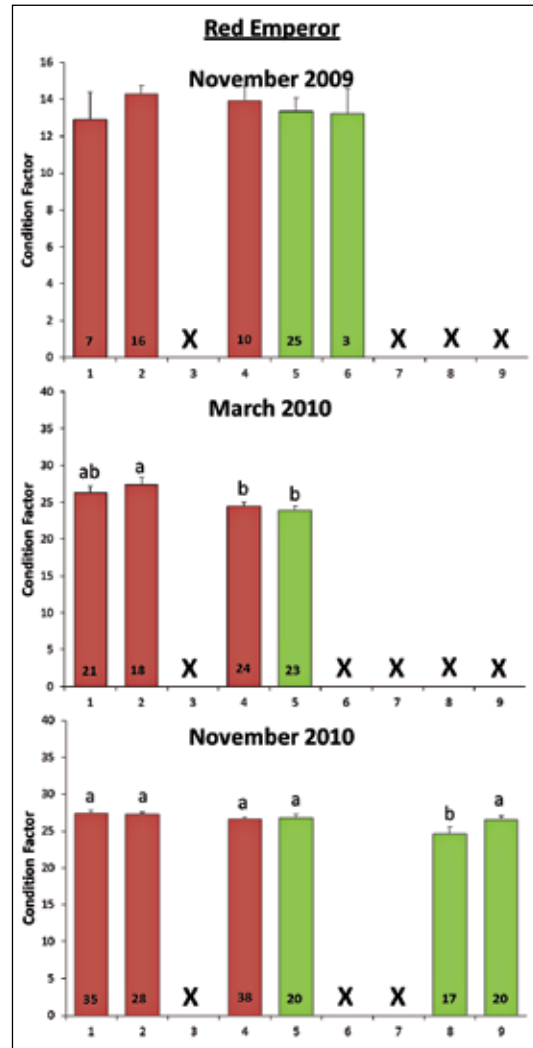


Figure 7. Condition factor of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

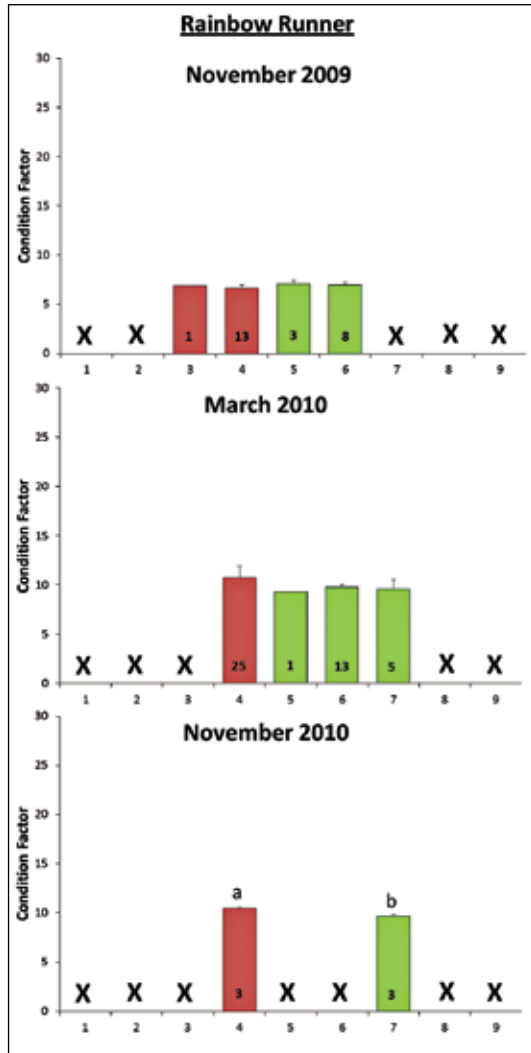


Figure 8. Condition factor of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

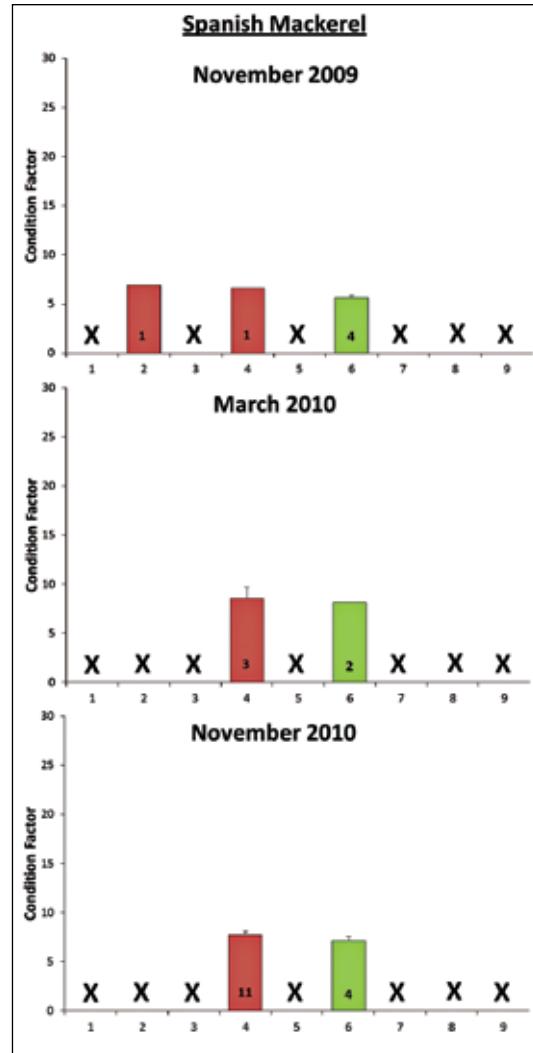


Figure 9. Condition factor of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

Liver Somatic Index (LSI)

Liver is a very plastic organ and can increase in size when continuously overworked due to a high metabolic activity (e.g., metabolism of xenobiotics). The liver somatic index (LSI) is a ratio of the liver weight relative to body weight, and is widely used as a general indicator of chronic exposure to contaminants. LSI can be affected by a number of factors other than exposure to pollutants (e.g., reproductive stage, nutritional availability) but it is a valuable initial screening tool of overall health.

In Phase I the LSI was similar at all sites for all four target species, indicating that any exposure of fish to petroleum hydrocarbons resulting from the Montara well release did not appear to be of sufficient duration, or of high concentration enough, to translate into physiological changes in LSI.

In Phase II the same pattern was measured in goldband snapper, rainbow runner and Spanish mackerel (no difference between fish captured in impacted and reference areas). However, red emperor living close to the West Atlas rig (sites 1 and 2) had larger livers relative to body weight than fish living in the reference areas. A larger liver can be the result of exposure to contaminants, in particular contaminants which are largely metabolised by the liver. Since many petrogenic compounds fall into this category the trend observed for red emperor observed in Phase III is that expected due to the chronic impact of exposure to contaminants as increased demand is placed on the liver of exposed fish to metabolise contaminants to forms which can be excreted.

Trends in LSI for Phase III were inconsistent with goldband snapper from site 2 having larger livers than those from the reference sites 5 and 9, but red emperor livers from the same site were smaller than those from the reference sites 8 and 9. The LSI of rainbow runner and Spanish mackerel were similar across sites.

Overall the observed trends which may be attributable to exposure to the products of the Montara well release occurred in March 2010 (red emperor) and November 2010 (goldband snapper). In the former of these cases a similar trend was not observed in November 2010. Therefore, any impact was transient. In the case of goldband snapper this was the first sampling period in which this trend was observed. It is unlikely that this can be related to the Montara well release.

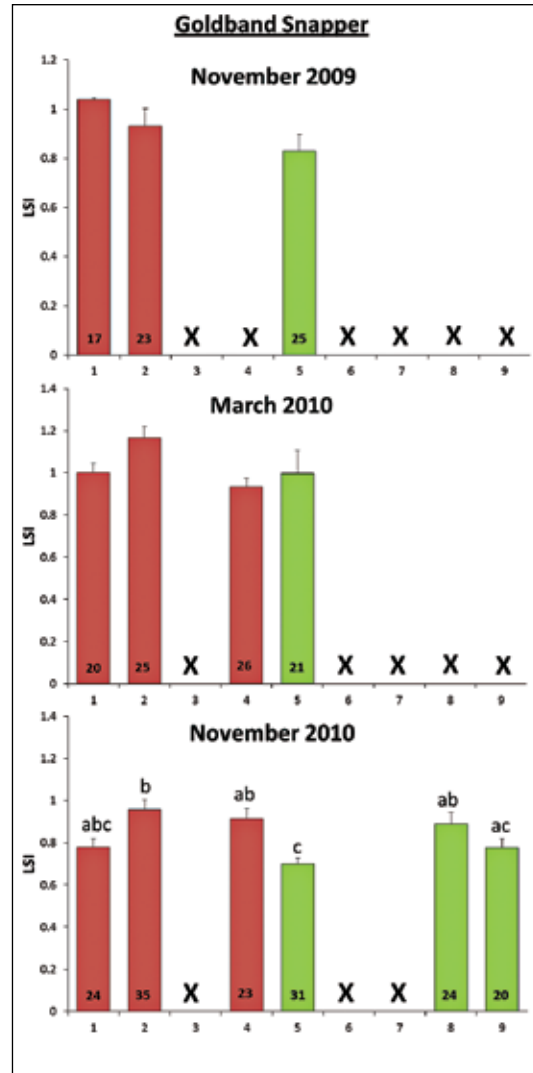


Figure 10. Liver somatic index of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

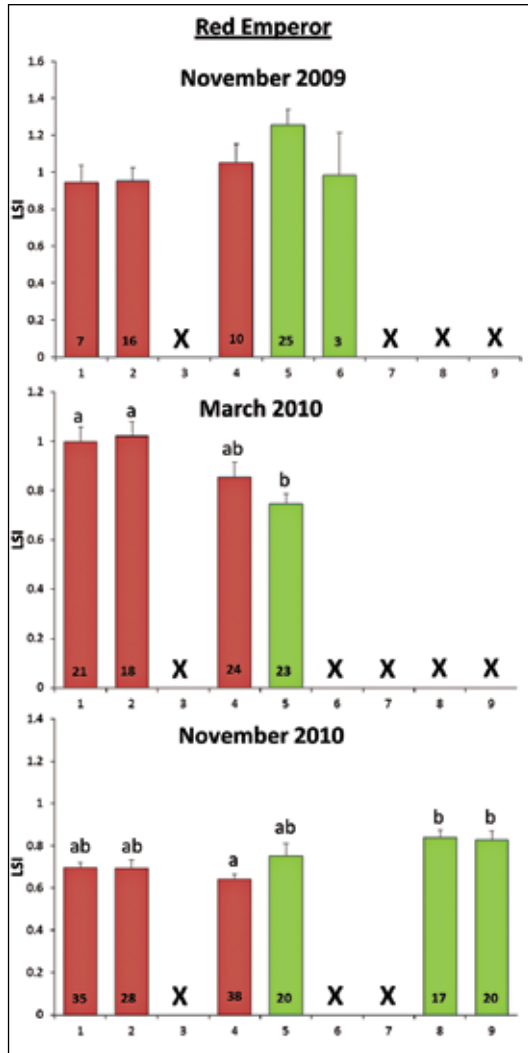


Figure 11. Liver somatic index of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

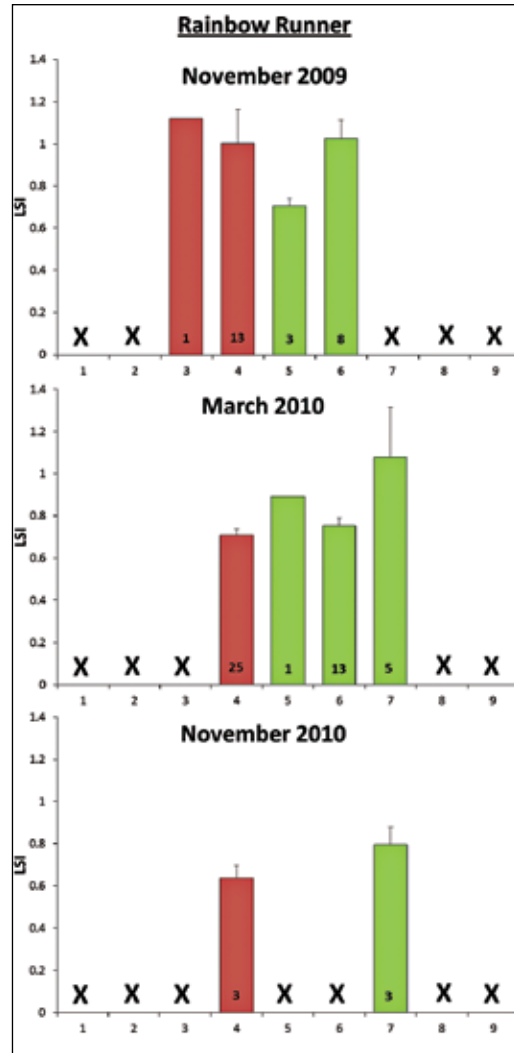


Figure 12. Liver somatic index of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

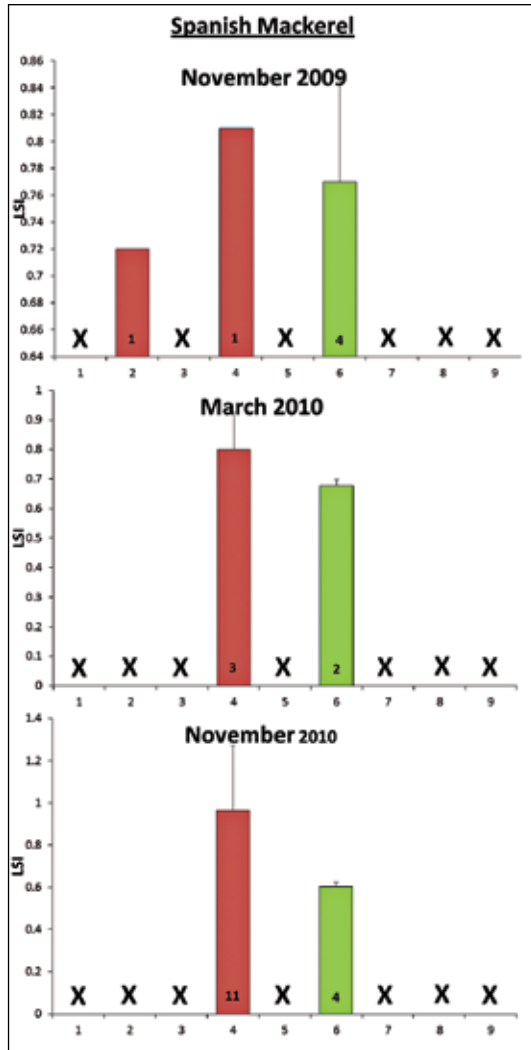


Figure 13. Liver somatic index of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2011. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

Gonado-somatic Index (GSI)

The GSI (ratio of the gonad weight to the body weight) provides an indication of the reproductive status of a fish. The GSI increases immediately prior to spawning and remains high through the reproductive season before decreasing as spawning ceases. When chronically or acutely exposed to contamination, reproduction is often one of the first parameters to be affected in adult fish.

Male

In November 2009 (Phase I) male fish of each of the four target species collected in the hydrocarbon-affected zone, had similar GSI to fish collected in the reference zone, suggesting that gonad development had proceeded at a similar rate. No gonad resorption or other abnormalities were observed in any of the specimens obtained. It was therefore concluded after Phase I that exposure to any hydrocarbons released from the Montara incident had not affected male gonadal development of the fish collected and examined.

In Phase II, there was again no statistical difference in male GSI in rainbow runner between impacted and reference areas (no statistical analysis was run on Spanish mackerel in Phase III as only one male was collected from each of the impacted and reference sites. In Phase III no males of either of these species were collected at the impacted sites so no comparison is possible.

Male goldband snapper had significantly lower GSI within 20 NM of the West Atlas rig than those fish captured in the reference area in Phase III. While the timing of reproduction varies between populations based on environmental conditions, it is possible that the difference measured here indicates a decrease in metabolic resources allocated to reproduction as a result of increase resources allocated to dealing with contamination, or simply a natural temporal variation in spawning activity between populations. Either way, the GSI measured in male goldband snapper in November 2010 (Phase III) was similar across all sites. Indicating that the effect was not on-going and, importantly, was not present during the onset of the reproductive period.

Male red emperor collected in the impacted area in November 2010 had lower GSI than those collected in at the non-impacted Site 5. This trend is similar to a non-significant trend noted in the March 2010 sampling period. It is possible that this is an impact of the Montara well release which is apparent at the beginning of the following reproductive season (Phase III).

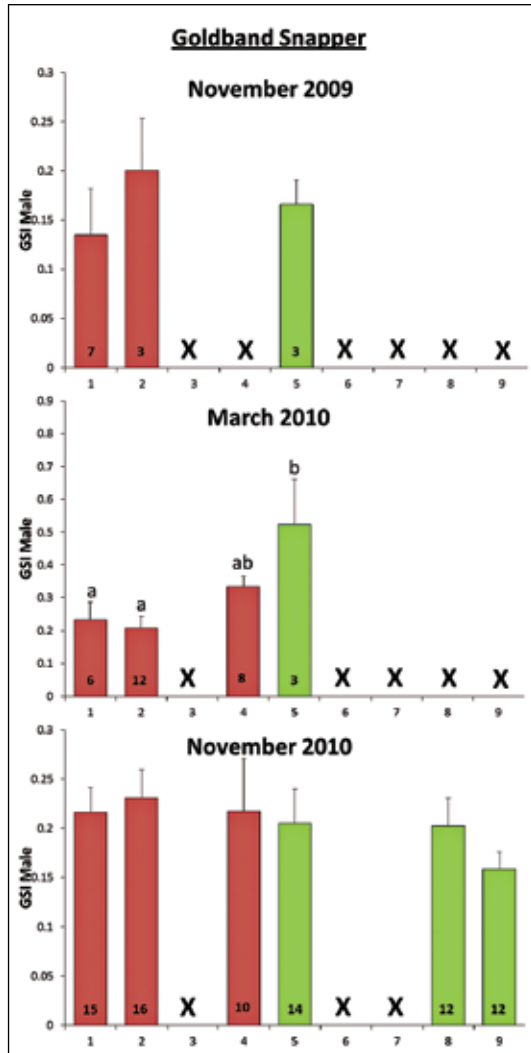


Figure 14. Gonado-somatic index of male goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

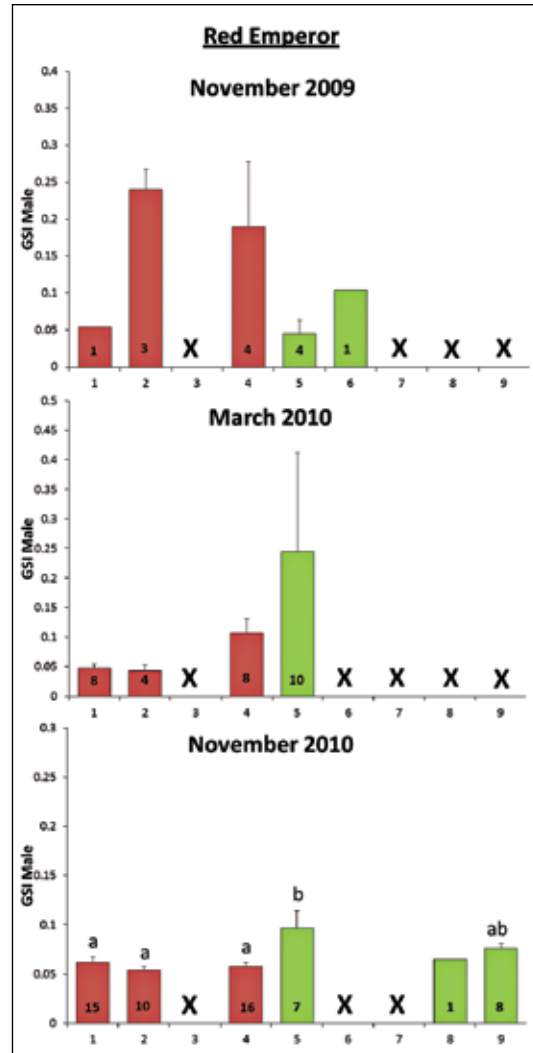


Figure 15. Gonado-somatic index of male red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

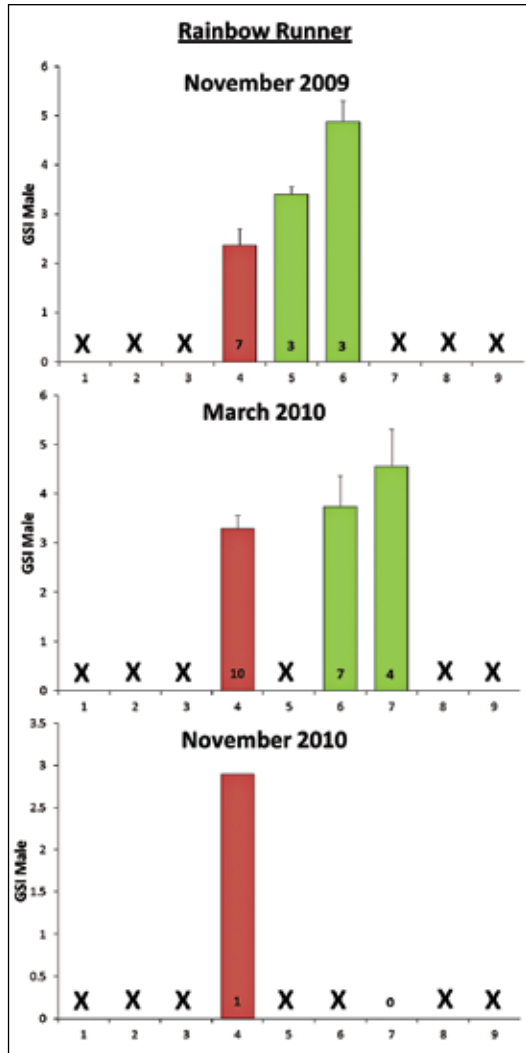


Figure 16. Gonado-somatic index of male rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

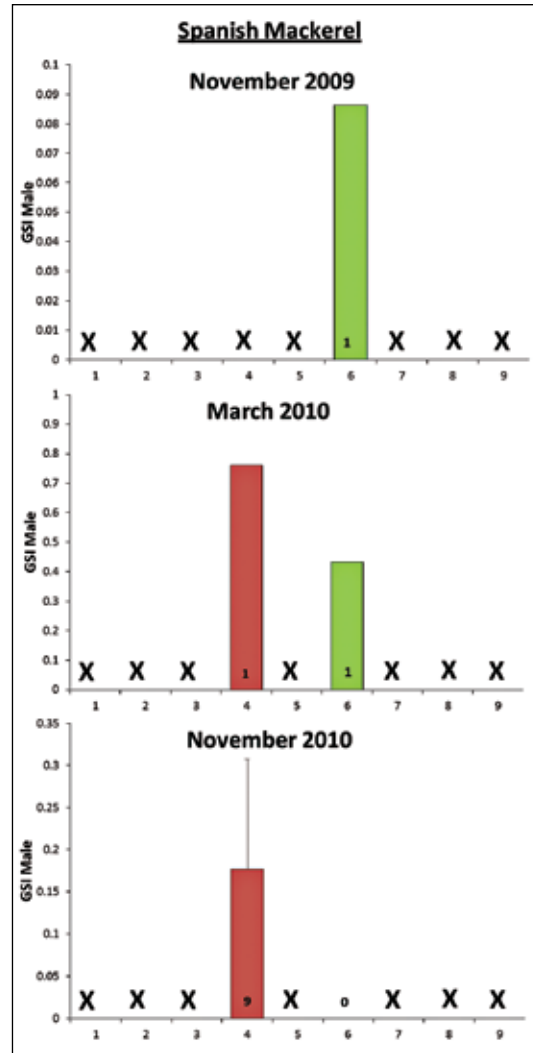


Figure 17. Gonado-somatic index of male Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

Female

In Phase I there was no difference between the GSI of females collected in impacted and reference areas for rainbow runner and Spanish mackerel. Female red emperor living closest to the West Atlas drilling had lower GSIs than those captured in reference areas. These differences were not observed in subsequent phases. Female goldband snapper captured in the site closest to the West Atlas drilling rig (Site 2) in Phase I had increased GSI compared to those captured further away from the drilling rig (Sites 1 and 5). As was the case with the trend observed for red emperor this difference was not maintained in Phases II and III. There were differences in the GSI of female goldband snapper but these did not follow any trend associated with the location of the West Atlas drilling rig.

Although there was no difference detected in the GSI of female rainbow runner collected at impacted and non-impacted sites during Phase I, those collected closest to the rig in Phase II (Site 4) had a lower GSI than those collected at the reference site (Site 6). While the same trend was observed in female rainbow runner collected during Phase III this trend was not statistically significant. As previously mentioned such a difference could be the result of differences in breeding status in the different populations, but could also be the result of a reduction in reproductive effort due to contamination. Gonad histology can provide a greater insight to gonadal development.

There were no significant differences between the GSI of female Spanish mackerel collected from impacted and non-impacted sites during any of the collection times (Phases I, II and III). Given the small number of individuals collected, this result is not surprising.

Overall

GSI as an indicator of reproductive status of fish is a biomarker with important potential population level impacts. The differences noted here require further investigation to establish if they are due to exposure to petroleum hydrocarbons from the Montara well or whether they are due to population-based differences.

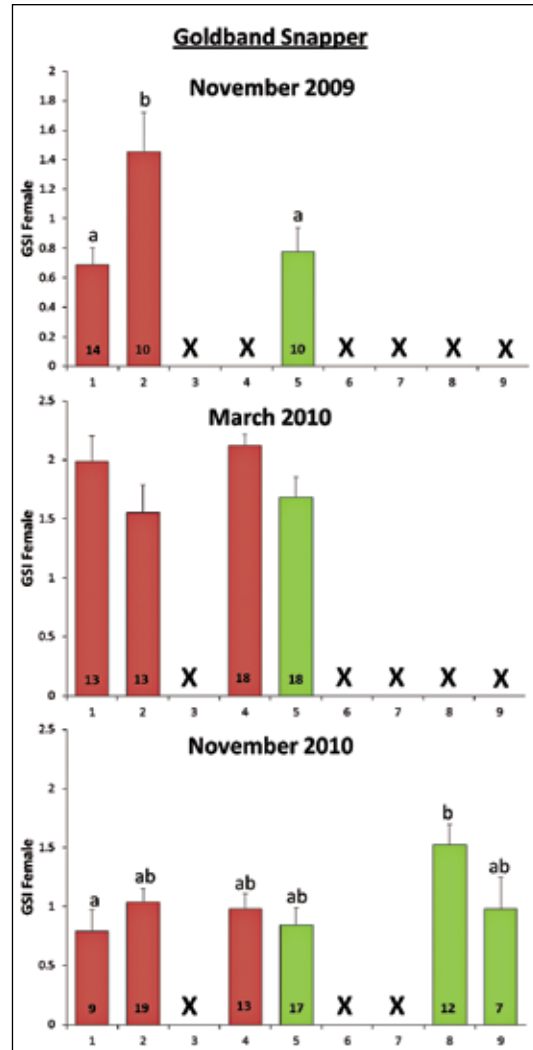


Figure 18. Gonado-somatic index of female goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

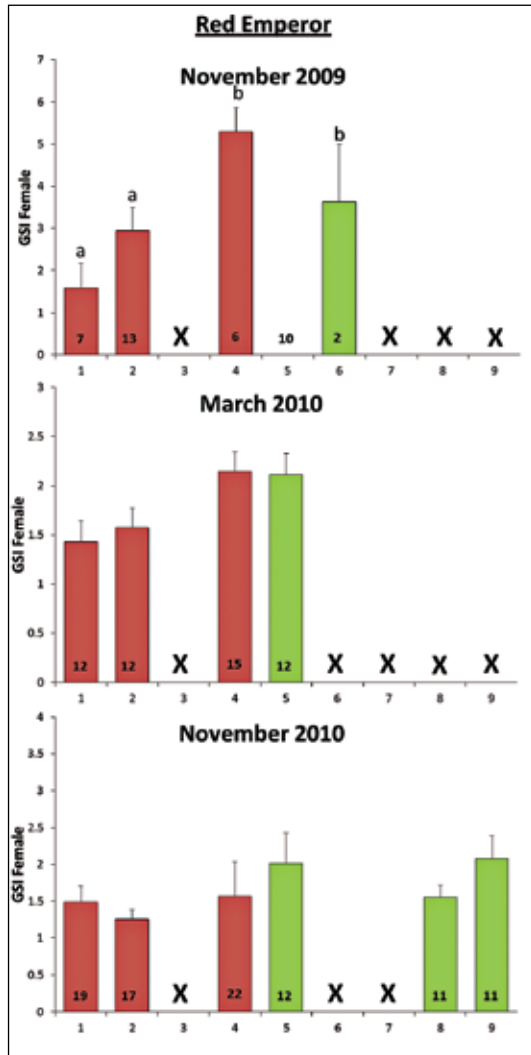


Figure 19. Gonado-somatic index of female red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

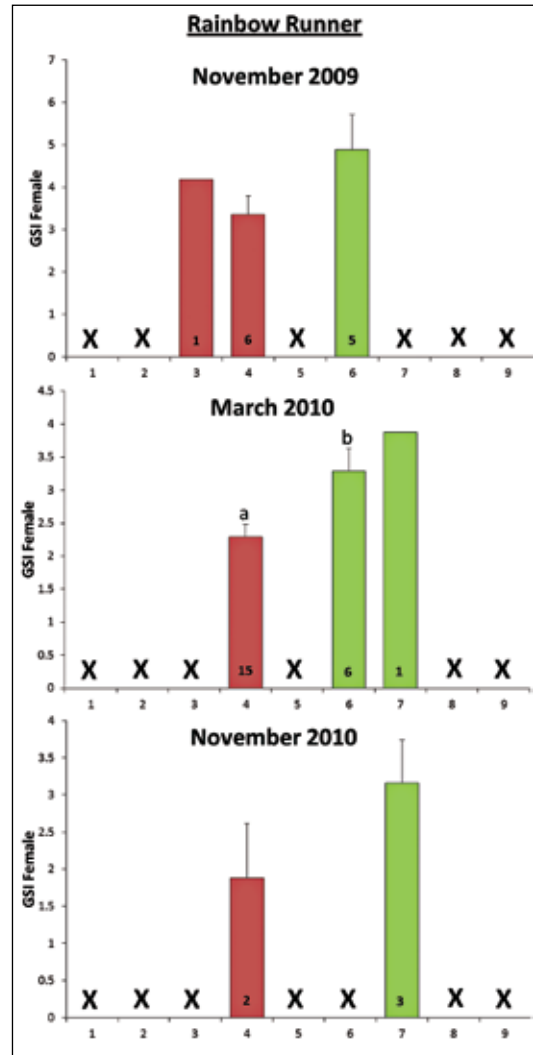


Figure 20. Gonado-somatic index of female rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

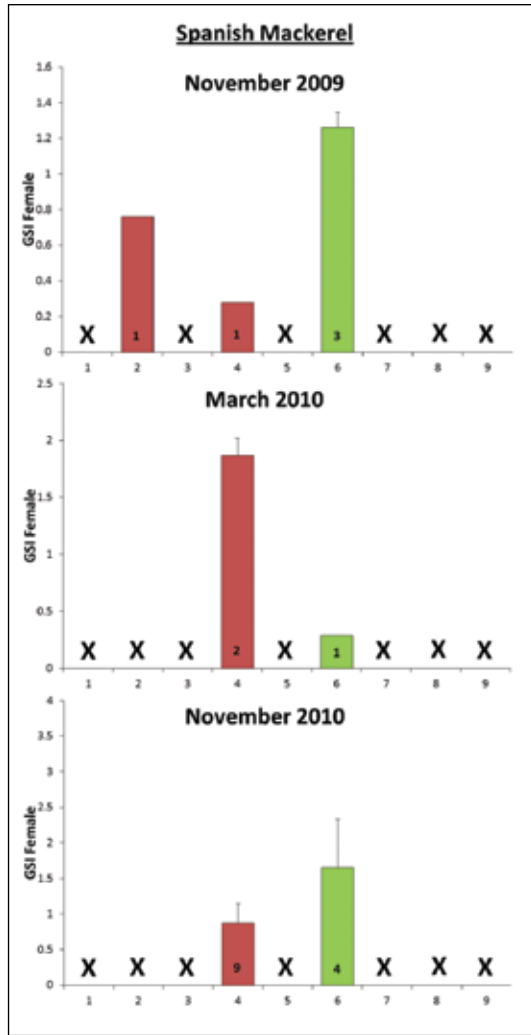


Figure 21. Gonado-somatic index of female Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

Gonad Histology

Histological examination gonadal tissue can provide information on the reproductive state of an animal and reveal pathologies which may otherwise go undetected (Bjerregaard *et al.*, 2006). Such examinations can be either quantitative (usually expressed as proportional distribution of different gamete developmental stage) or qualitative (based on visual examination only). Below is a qualitative histological analysis of gonadal tissue on collected fish in Phase I and II of this study.

Male

In Phase I it was noted that male fish of all four target species had mature spermatazoa visible in the efferent duct indicating that the fish were either spawning or that spawning was close to commencing and that the gonad also contained developing gametes of a variety of stages. In male rainbow runner and Spanish mackerel gonads mature spermatazoa within wide seminiferous tubules made up the bulk of the organ indicating a high reproductive output at the time of sampling. In red emperor and goldband snapper there was a greater proportion of primary and secondary spermatogonia evident toward the periphery suggesting that these species were probably early in their reproductive season.

In Phase II the gonad of the male rainbow runner, again had very high numbers of mature spermatazoa in the efferent duct indicating that this species was still actively spawning. Male goldband snapper had mature sperm in the efferent duct but appeared to have reduced numbers of developing spermatogonia indicating that it was toward the end of the reproductive season. Although the male red emperor had increased GSI (see above) histological analysis indicated that their gonad still contained mature spermatazoa.

Female

The ovaries of the female fish of each species in Phase I contained fully developed oocytes suggesting that the animals were reproductively fit and either preparing to spawn or in the process of spawning. In each species the majority of the ovary was made up of Stage III oocytes, with a smaller proportion of Stage IV and V oocytes present toward the periphery. In Phase II rainbow runner ovaries appeared to contain a slightly higher proportion of Stage IV and V oocytes than those in Phase I and there were no obvious differences in gonadal structure between the gonads of females from impacted and reference areas.

Female goldband snapper sampled in Phase I had a small proportion of stage IV oocytes and even fewer stage V oocytes. In Phase II both stage IV and V oocytes were much more common in goldband snapper ovaries suggesting that these fish were either actively reproducing or had recently completed their reproductive season. Based on visual analysis the ovaries collected from impacted and reference areas were histologically similar.

There was no histological difference between red emperor ovaries from impacted and reference areas in Phase I. Ovaries from fish from both locations were predominantly made up of stage III oocytes. In Phase II red emperor gonads contained higher numbers of stage IV and V oocytes. There were no visual differences between the ovaries of females red emperor collected from impacted and reference areas.

Overall

There were histological differences between both male and female gonads sampled in Phase I and Phase II. This was particularly evident in male and female goldband snapper and red emperor. In both cases female gonads contained more numerous late developmental stage oocytes. In both Phase I and Phase II there were no apparent histological differences between fish captured in impacted or reference areas indicating no impact of exposure to petroleum hydrocarbons on the normal reproductive cycle (gonadal and gamete development).

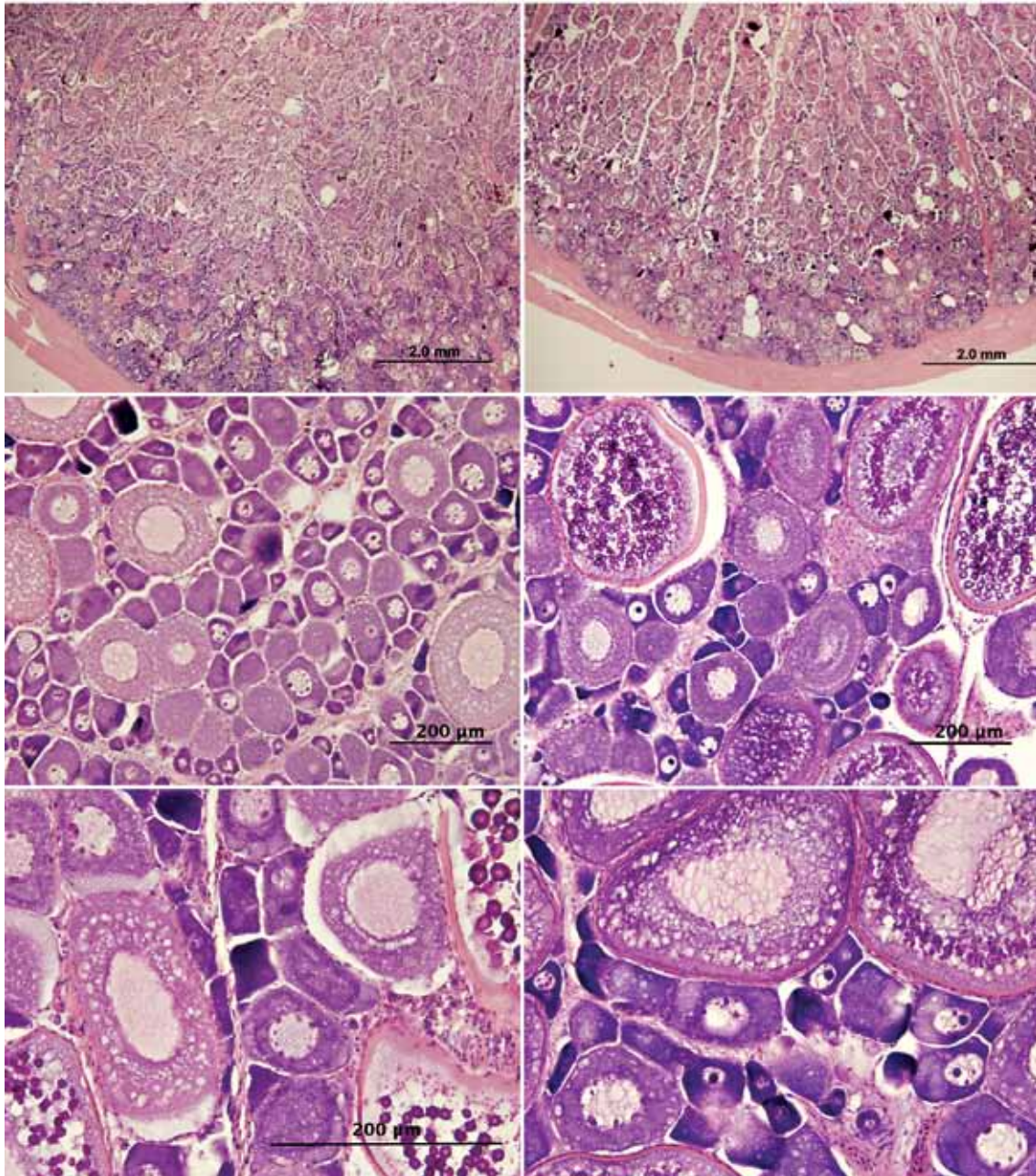


Figure 22. Stained (haematoxylin-eosin) slides of gonadal tissue of female goldband snapper collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.

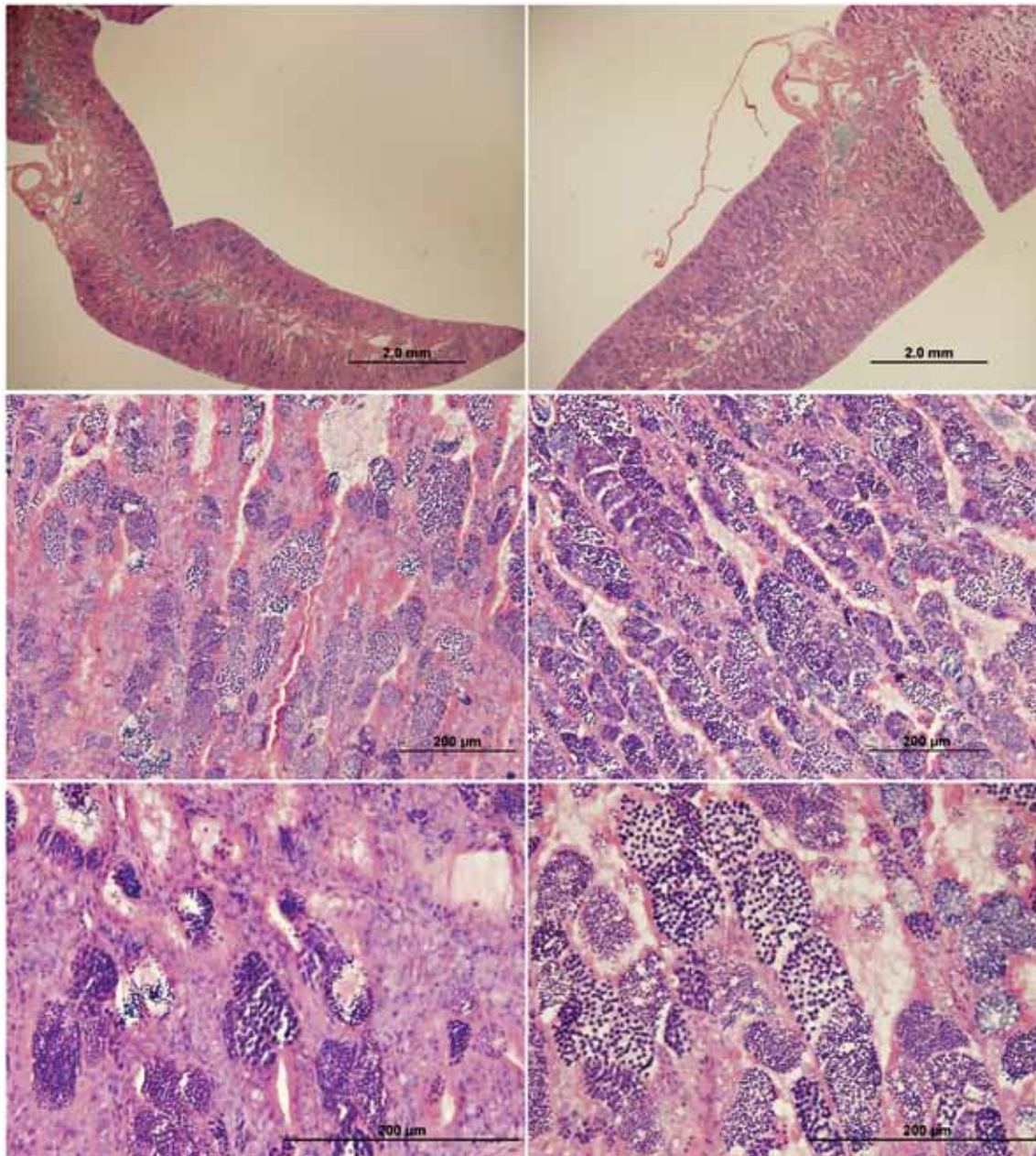


Figure 23. Stained (haematoxylin-eosin) slides of gonadal tissue of male goldband snapper collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010

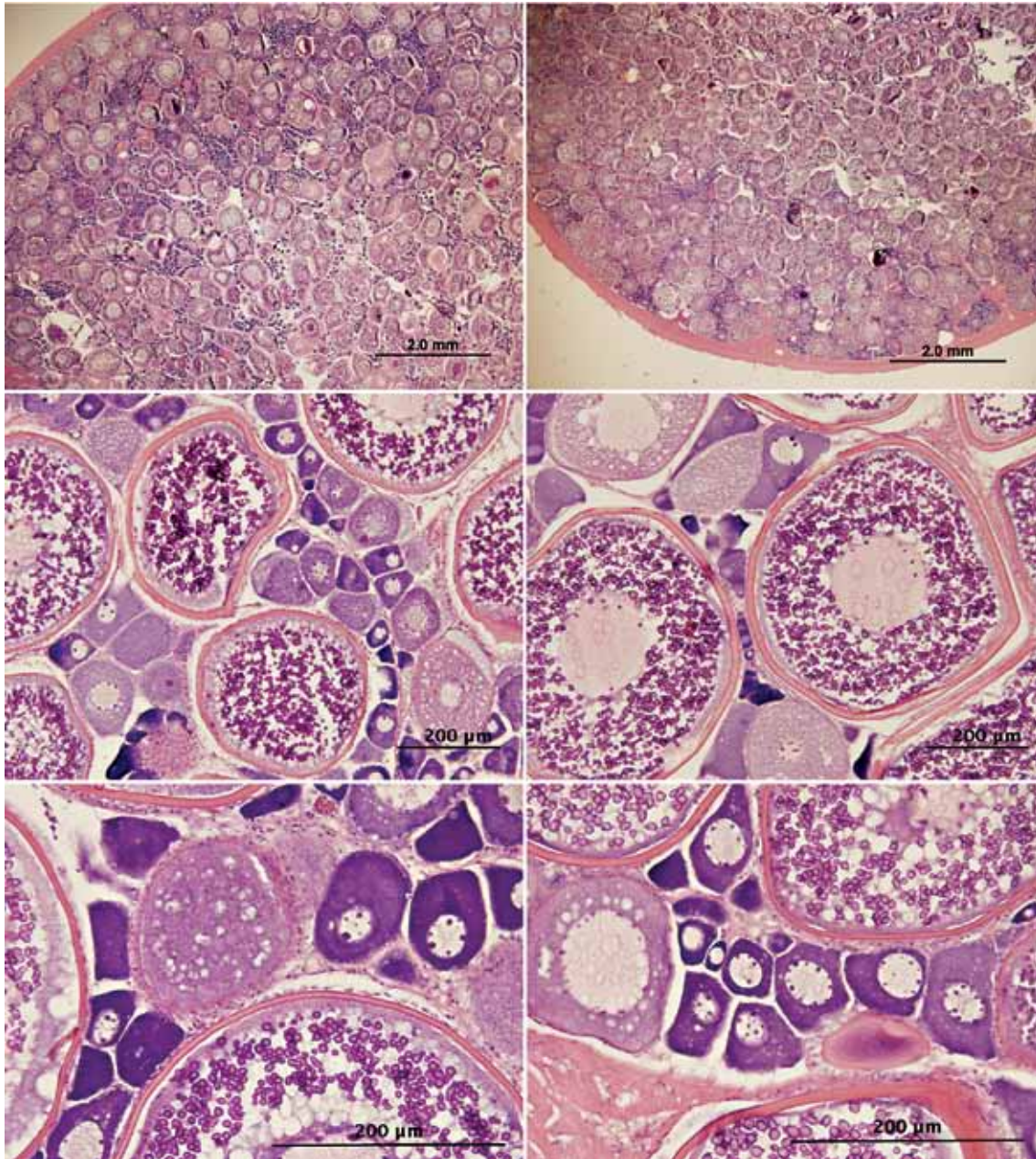


Figure 24. Stained (haematoxylin-eosin) slides of gonadal tissue of female rainbow runner collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010

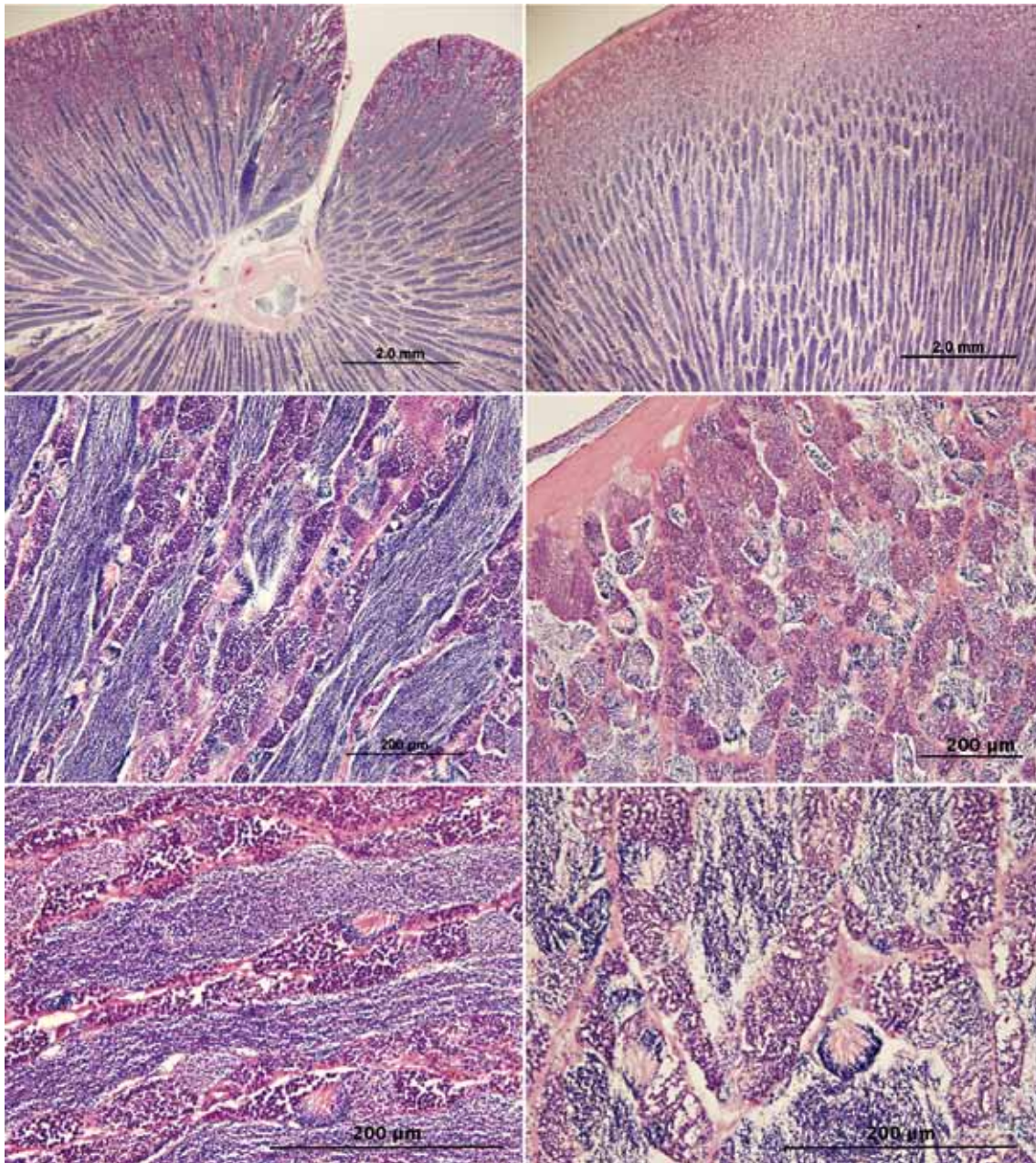


Figure 25. Stained (haematoxylin-eosin) slides of gonadal tissue of male rainbow runner collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.

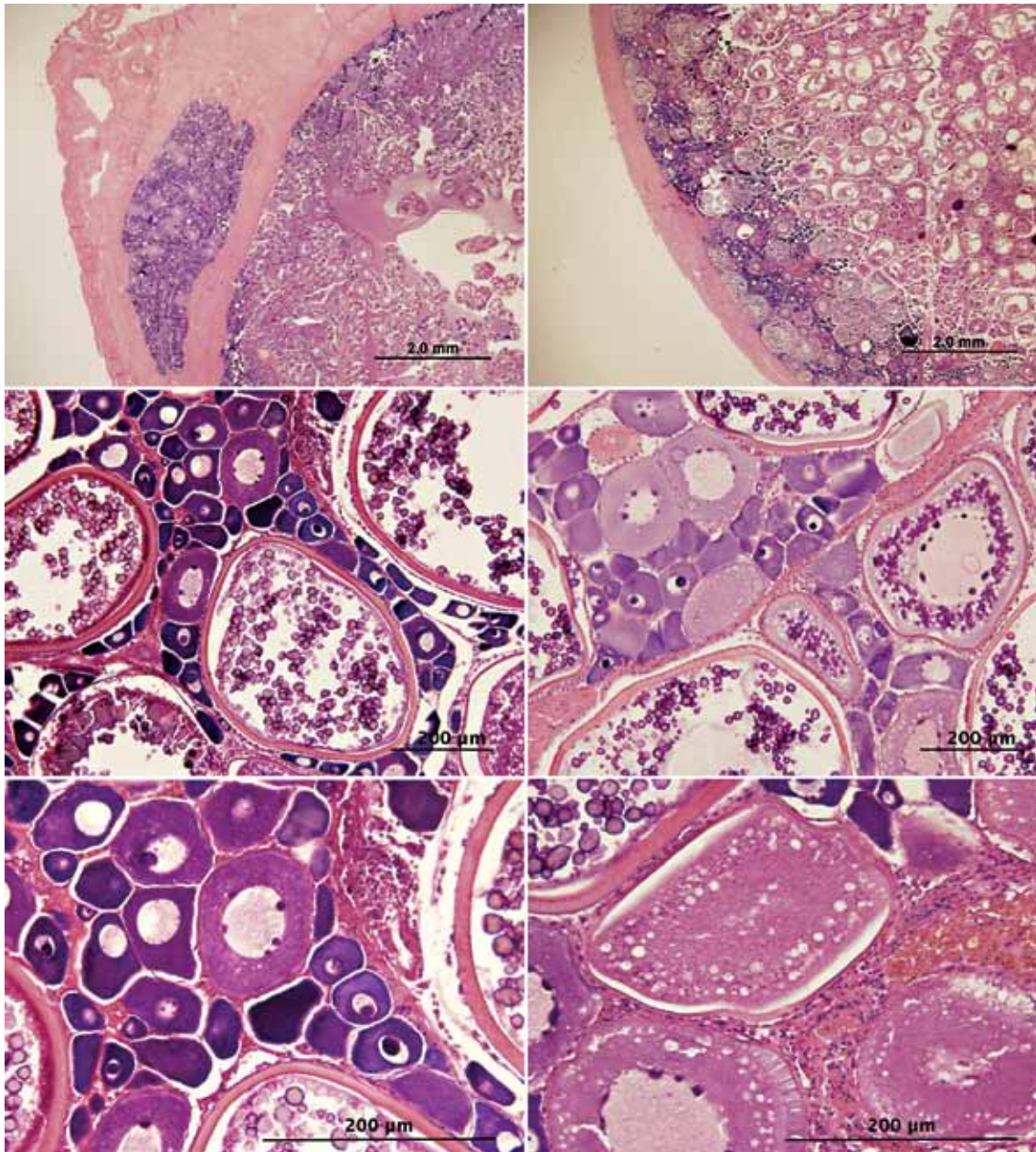


Figure 26. Stained (haematoxylin-eosin) slides of gonadal tissue of female red emperor collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.

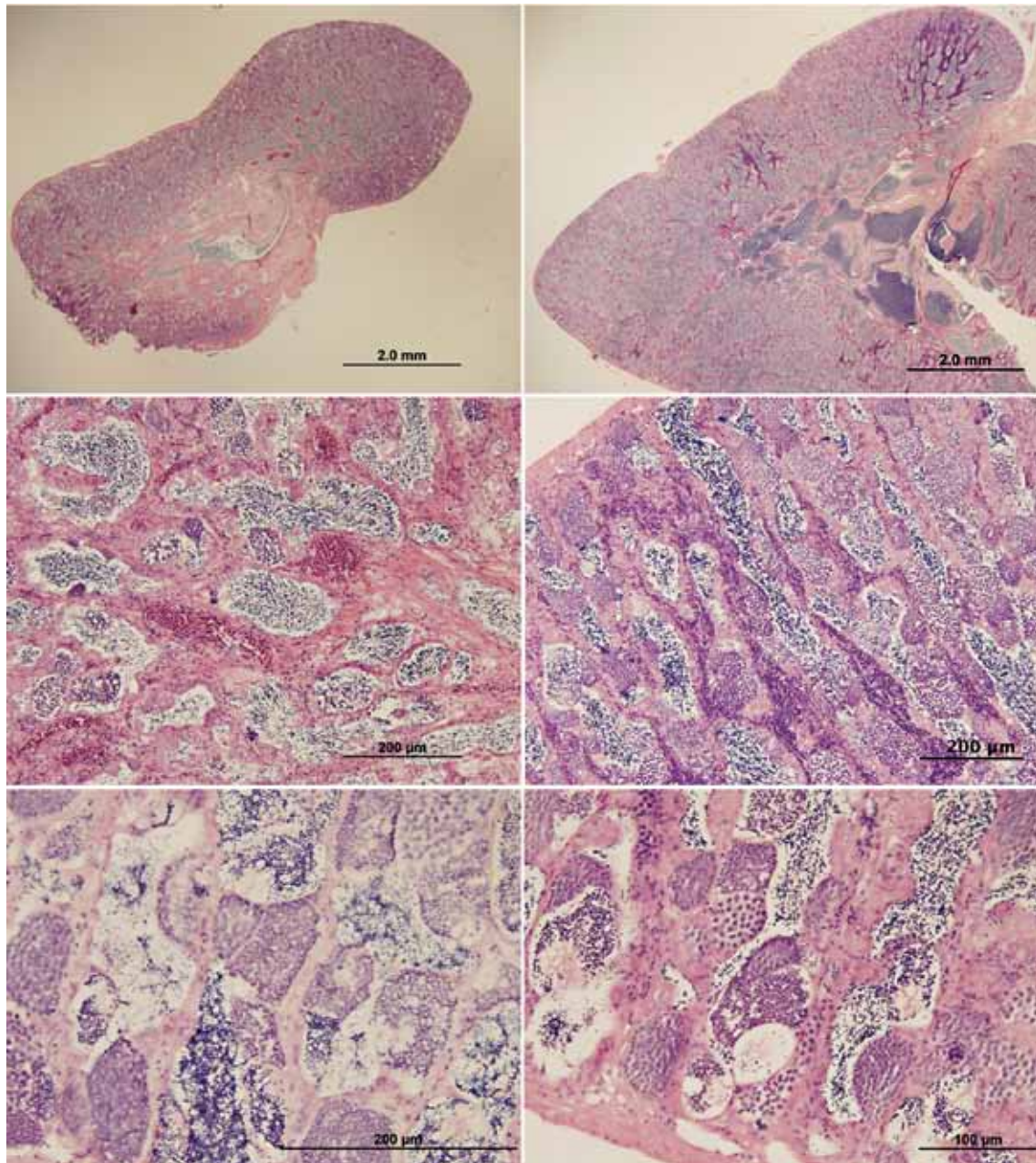


Figure 27. Stained (haematoxylin-eosin) slides of gonadal tissue of male red emperor collected from reference areas (left) and impacted areas (right) in the Timor Sea in March 2010.

Biochemical Parameters

Liver Detoxification Enzymes

Assimilated contaminants such as petroleum hydrocarbons are metabolised by liver enzymes, in order to render them more water-soluble (e.g. blood soluble). Specific enzymes, such as those of the CYP1A1 system, are measurable in fish livers even if the inducing chemical is well below detectable levels in the environment (Landis and Yu, 1995). Not all contaminants induce the activity of the liver detoxification enzymes, but it is known that petroleum hydrocarbons, especially the heavier components, are potent CYP1A1 inducers (McDonald *et al.*, 1995). Detoxification activity can be measured by the conversion of ethoxyresorufin to resorufin by CYP1A1 via ethoxyresorufin-o-deethylase (EROD). Detoxification enzymes are present in all animals, and while basal levels are species-specific, a significant increase in EROD activity between individuals of the same species collected at different locations suggests a chronic and sustained exposure to contaminants that are metabolised by the liver. Liver detoxification activity can be induced within days following exposure, and can return to basal levels within a week following the end of exposure to the inducing compounds. Surface fish were at higher risk of exposure to surface hydrocarbon residues than deep sea fish, and consequently, it could be expected that EROD activity would be induced at higher levels in surface fish relative to deep sea fish.

The activity of the liver enzymes can vary naturally with sex hormones, increasing the variability of this biomarker. For example, the presence of estrogen in the reproductively active female fish can significantly inhibit the measured EROD activity levels (van der Oost *et al.*, 2003). For every phase of the study, the EROD activity levels were been compared on a species basis between sexes at each location and found to be not significantly different between sexes. Consequently, sexes of a same species were pooled to proceed with data interpretation.

In Phase I, none of the demersal or pelagic fish species appeared to have significantly activated their liver detoxification systems. The fresh light crude oil escaping from the Montara well head had a low proportion of high-molecular weight PAHs and consequently this oil had low potential to induce this detoxification enzyme. As a result, no significantly increased levels of detoxification enzymes could be detected in the liver of four species of fish sampled during Phase I November 2009 investigation.

During Phase II March 2010 sampling, however, the demersal fish collected close to the West Atlas rig exhibited high liver detoxification activity, suggesting that residues of petroleum compounds have reached deeper waters through suspended particles or vertically migrating prey ingested by goldband snapper and red emperor.

Phase III, liver detoxification activity in goldband snapper and red emperor collected within 20 NM from the rig was still slightly elevated relative to one of the reference site however, the enzymatic activity clearly had a trend towards returning to reference levels in both species of fish.

Interestingly, goldband snapper and red emperor sampled in Phase III exhibited slightly elevated detoxification activity at Heywood shoal, a site deemed impacted located at 53 NM south-west from the West Atlas well release. The continuing liver detoxification activity observed in goldband snapper and red emperor captured in the vicinity of Heywood shoal suggests that natural seepages might be present in the area. It is unlikely that petroleum hydrocarbons from the West Atlas well release would contribute significantly to the continuing detoxification activity in these fish, as environmental conditions, e.g. warm water temperatures, favour a rapid degradation of oil in the Timor Sea (Burns *et al.*, 2010). It is therefore unlikely that spilled oil even does reach the ocean floor (Burns *et al.*, 2010).

Similarly, the rainbow runners collected at Heywood shoal impacted site 53 NM away from the rig showed elevated hepatic detoxification, while individuals of this species collected at the reference sites were exhibiting only background levels. Statistically, detoxification activities were at similar levels in Spanish mackerel from impacted and reference sites however, only two individuals of this species were collected at each sites making statistical significance difficult to achieve. The trend in Spanish mackerel was similar to that observed in rainbow runner, with liver detoxification higher in fish collected closer from the impacted area, relative to observations made in fish originating from the reference area.

The most important feature of an active liver enzyme system is its ability to metabolise large, bulky contaminants bound for elimination out of the body.

These enzymes initiate the elimination process by transforming lipophilic xenobiotics into more water-soluble compounds which are then directed to the biliary secretion via the bloodstream. Occasionally, metabolism of PAHs creates reactive metabolites that are highly toxic, mutagenic to carcinogenic (van der Oost *et al.*, 2003). The oxidative metabolism of PAHs, for instance, proceeds via highly electrophilic intermediates, some of which bind covalently to cellular macromolecules such as DNA (van der Oost *et al.*, 2003).

While a higher level of liver detoxification system confirms continuing exposure to contaminants in fish captured closer to the rig, it does not confirm that adverse effects have occurred in these individuals. Liver detoxification activity is a biomarker of exposure to contaminants and confirms uptake and processing of xenobiotics. In order to assess potential effects related to the metabolism of xenobiotics, it is relevant to measure complementary biomarkers such as DNA damage.

During acute exposure of fish to aquatic contaminants, EROD activity can be induced several fold relative to the reference levels. While EROD activity for other species such as trout is recognised to be highly inducible, e.g. at least 10-fold higher relative to control fish (Whyte *et al.*, 2000), most fish species can only be induced at much lower x-fold when subjected to continuous exposure to contaminants. In the present study, a maximum of two-fold induction was observed at any time between gold-band snapper and red emperor from reference and impacted sites. The EROD activity levels were, however, of the same order of magnitude as those observed in another study conducted in the Timor Sea. Codi-King *et al.*, (2005) measured EROD activity levels in gold-spotted trevally (*Carangoides fulvoguttatus*) and the bar-cheeked coral trout (*Plectropomus maculatus*) and found, for reference and produced formation water-exposed fish, similar EROD activity levels to those reported in the present study.

Comparatively, a three-year fish health monitoring program conducted following the 2002 Prestige oil spill that occurred off the north-west coast of Spain established that only after two to three years did the fish biomarker levels, including EROD activity, returned to baseline levels (Martinez-Gomez *et al.*, 2009). Of particular concern to both the Prestige and Montara spills is the release in the marine environment of polycyclic aromatic

hydrocarbons (PAHs), as the parent compounds as well as their metabolites can exhibit mutagenic and/or carcinogenic potential, as well as induce EROD activity. The Prestige oil involved a heavy fuel oil (M-100) of different composition to the Montara oil, however both crude oils had a low presence of high molecular weight polycyclic aromatic hydrocarbons (PAHs) (Alzaga *et al.*, 2004).

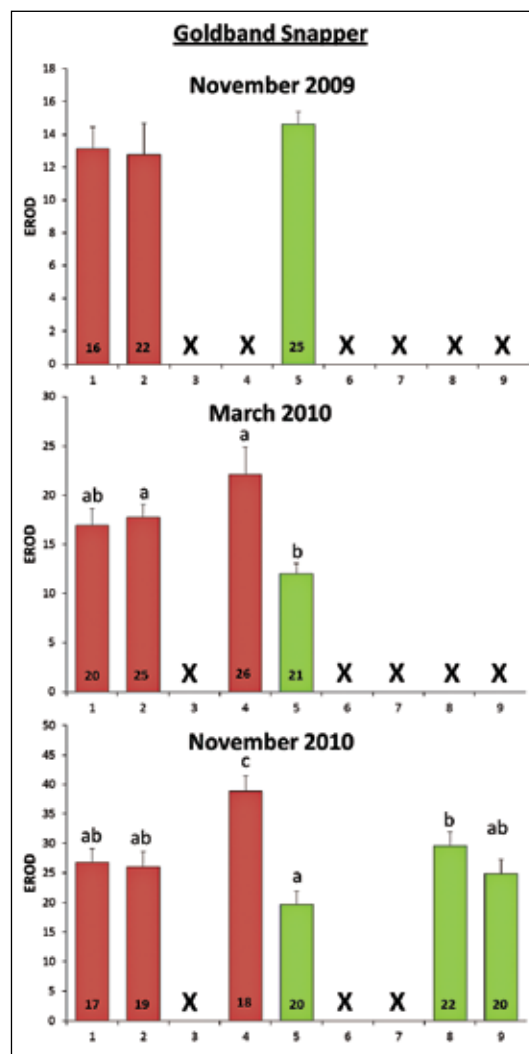


Figure 28. EROD activity (pmol/ mg protein/ min) in goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

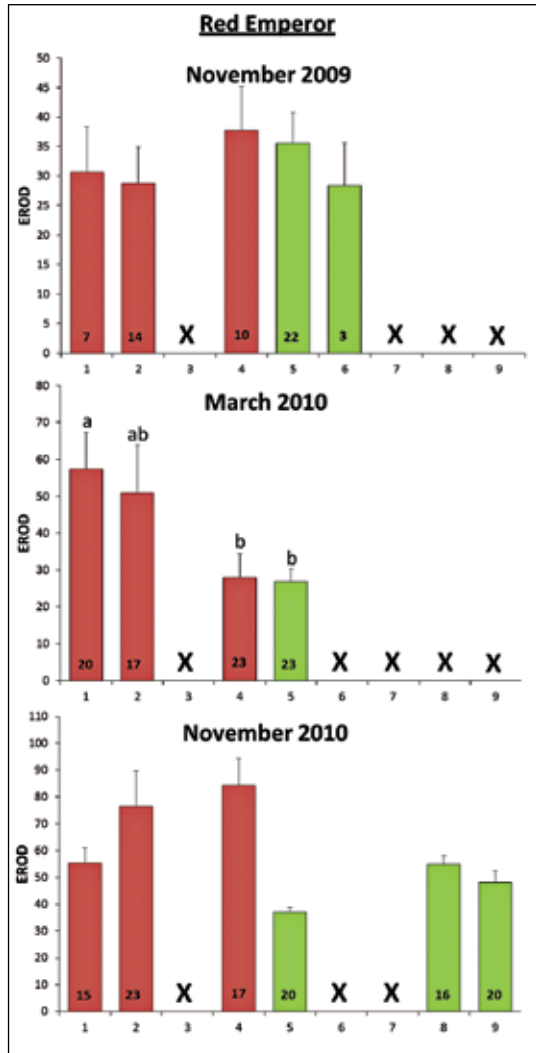


Figure 29. EROD activity (pmol/ mg protein/ min) in red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

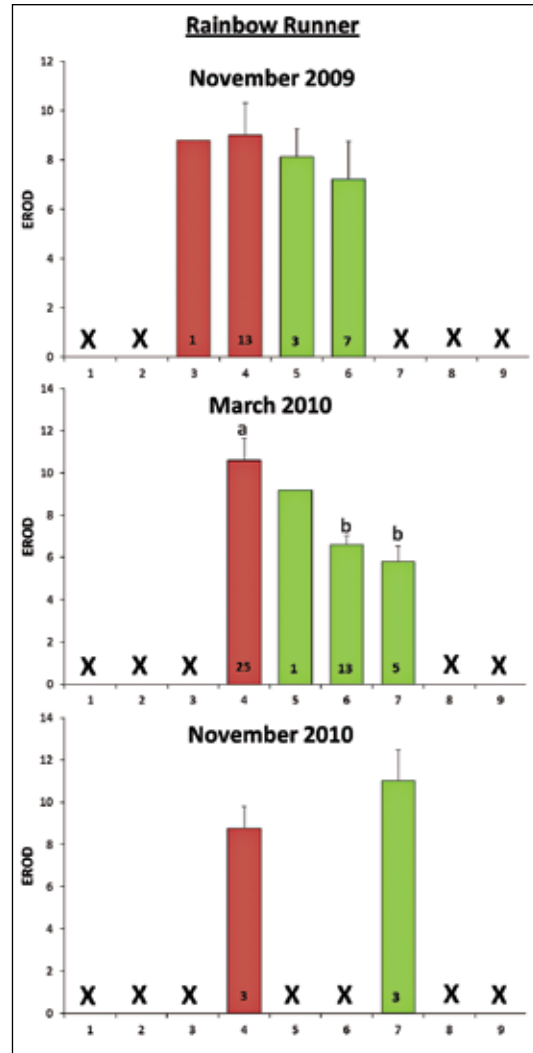


Figure 30. EROD activity (pmol/ mg protein/ min) in rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

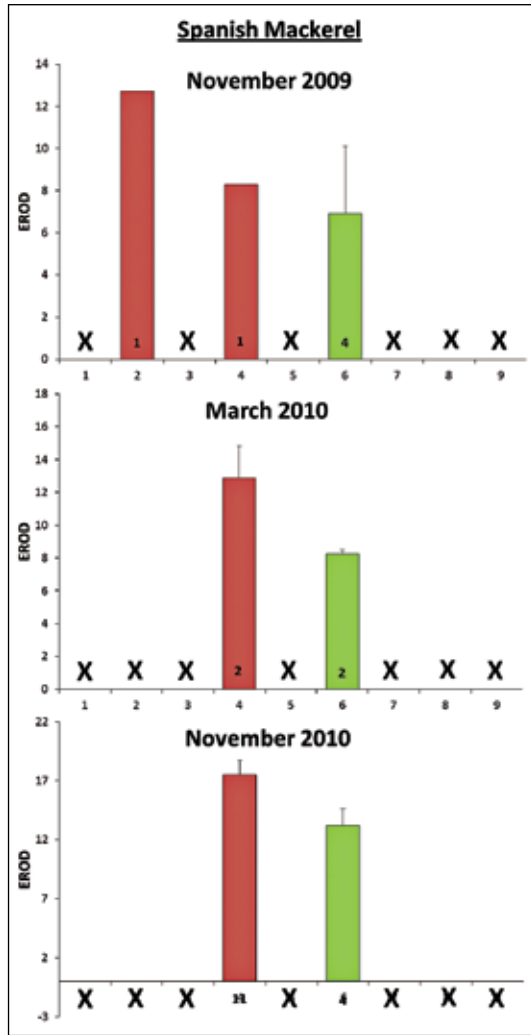


Figure 31. EROD activity (pmol/ mg protein/ min) in Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

Biliary Metabolites

PAHs processed by the liver enzymes are directed to the biliary secretions for elimination out of the body via the intestinal route. The detection of PAH metabolites in the biliary secretions indicates that fish have recently (within the past week) assimilated petroleum hydrocarbons, have metabolised them and are in the process of eliminating these compounds from the body. As is the case with most vertebrates, fish do not accumulate petroleum hydrocarbons in their flesh because they are capable of metabolising PAHs at rates that prevent significant bioaccumulation (Hartung, 1995).

The elimination of PAH metabolites is evaluated by the measurement of three types of metabolites in the biliary secretions, these being naphthalene, pyrene and benzo(a)pyrene [B(a)P] metabolites.

The presence of PAH biliary metabolites in fish is an extremely sensitive biomarker of exposure. As a result, PAHs might be found in biliary secretions at levels up to 1000 x more concentrated than in the surrounding environment (Hellou and Payne, 1987; Meador *et al.*, 1995). The presence of PAH metabolites in the biliary secretions indicates uptake of petroleum compounds by the fish, however, it is not suggestive of adverse effects on fish health.

In the initial sampling conducted in November 2009, goldband snapper, red emperor and rainbow runner collected in the impacted areas all had significant levels of naphthalene, pyrene and B(a)P metabolites in their biliary secretions, relative to their respective species collected in reference zones. Despite the fact that these PAHs were not detected in the water column (AMSA 2009) and are not in the long term accumulated in fish flesh, their presence in the biliary secretions indicates that the fish were recently (within 7 days, Gagnon and Holdway, 2000) exposed to petroleum compounds. Too few Spanish mackerel were collected during Phase I sampling to provide a reliable assessment of the biliary metabolite levels.

In Phase II of the study, the demersal fish collected at the sites closest to the West Atlas rig consistently exhibited elevated PAH biliary metabolites, a fact that supports observations of high liver detoxification activity in fish closest to the rig. This pattern of biliary metabolites further suggests that petroleum products might make their way to deeper waters via the sinking of suspended particulates, or via the vertical migration of prey eventually consumed by the demersal fish species. In pelagic species however, the biliary metabolite levels were at similar levels at all sites, which suggests that petroleum residues have disappeared from the surface.

Phase III sampling saw a levelling of most biliary metabolite levels in both demersal and pelagic fish species, with PAH bile metabolites being at comparable levels in fish from reference and impacted sites.

The low variability of this biomarker and the sizeable number of fish collected contribute to the establishment of statistical differences between reference and impacted sites for goldband snapper and red emperor, however, these differences are small compared to previous observations made in this monitoring program. Fish collected in Phases I and II exhibited bile metabolites frequently 2-fold higher in fish collected from impacted sites. In Phase III sampling, these differences in the abundance of biliary metabolite levels between fish collected at reference or impacted sites were much reduced, suggesting that exposure of fish to petroleum hydrocarbons has now reached similar levels at impacted and reference sites.

In another field study where fish were collected in a the PAH-contaminated Port Philip Bay, sand flathead (*Platycephalus bassesnsis*) collected closer to industrial areas exhibited up to 9-fold increase in PAH biliary metabolites, relative to fish from a reference area (Gagnon and Holdway, 2002). A laboratory study demonstrated that juvenile Atlantic salmon exposed to dispersed Bass Strait crude oil can have levels of biliary naphthalene metabolites up to 15 times that of control fish (Gagnon and Holdway 2000). By comparison, the 2-fold increase observed in goldband snapper and red emperor during the monitoring of fish health is modest. The levelling of biliary fluorescence levels between fish collected at reference and impacted sites further suggests that the presence of petroleum hydrocarbons is much reduced in this environment, relative to the sampling performed immediately after the end of the well release.

Bile metabolites represent an extremely sensitive biomarker of exposure to petroleum hydrocarbons, with biliary metabolite concentrations being up to 1000-fold more concentrated in fish bile relative to the surrounding water (Hellou and Payne, 1987, Meador *et al.*, 1995). The presence of PAH metabolites in the biliary secretions indicates uptake of petroleum compounds by the fish however, it is not suggestive of adverse effects on fish health.

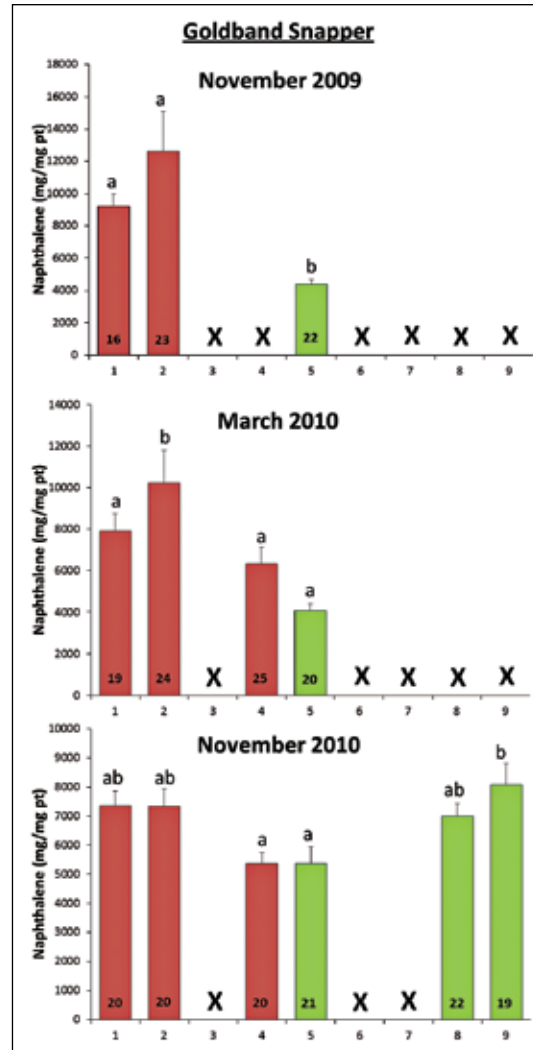


Figure 32. Naphthalene type metabolite concentrations in the bile of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

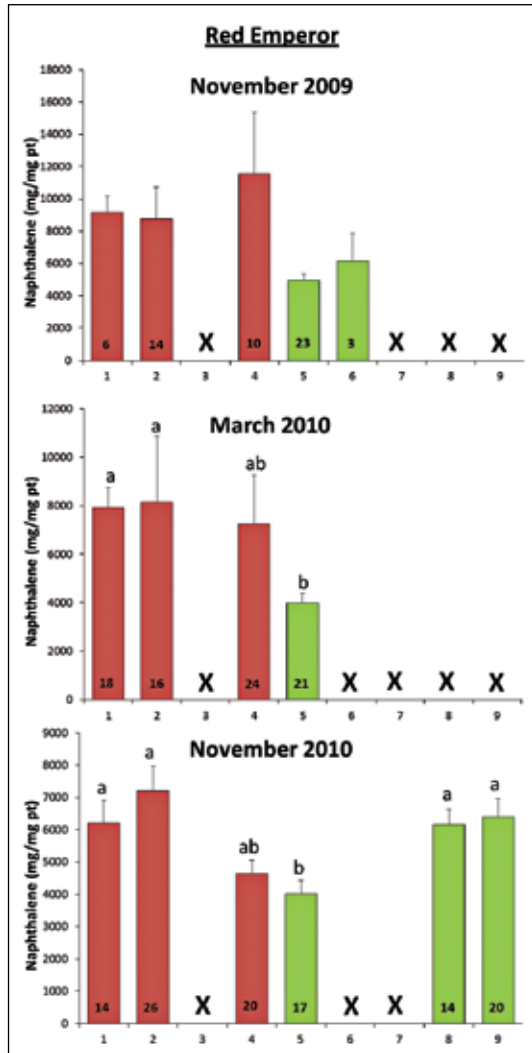


Figure 33. Naphthalene type metabolite concentrations in the bile of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

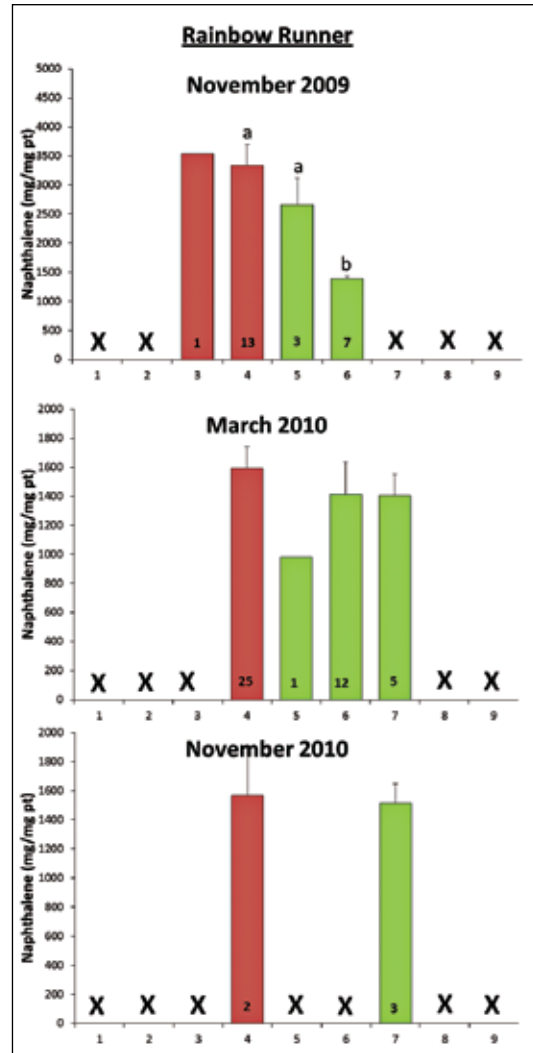


Figure 34. Naphthalene type metabolite concentrations in the bile of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

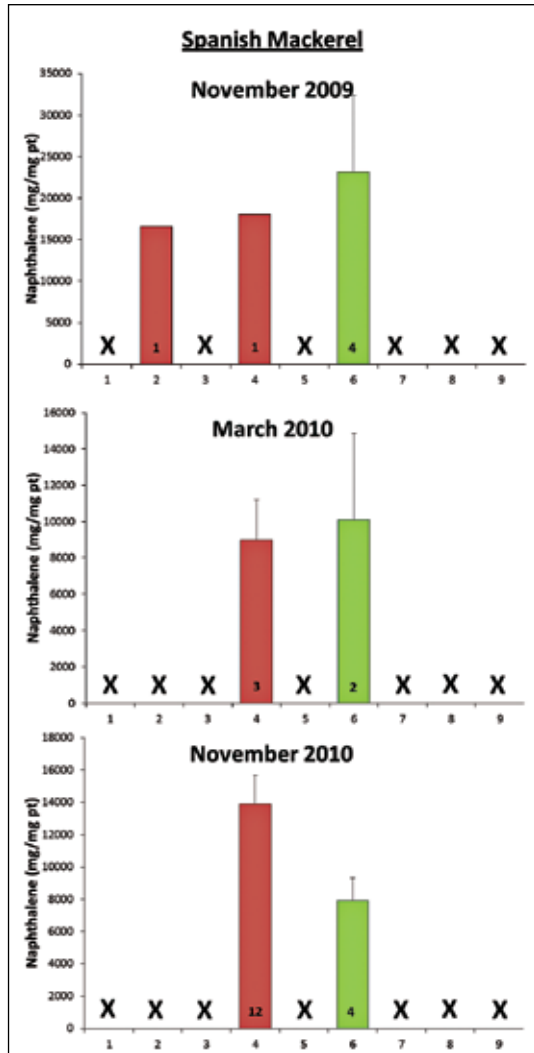


Figure 35. Naphthalene type metabolite concentrations in the bile of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

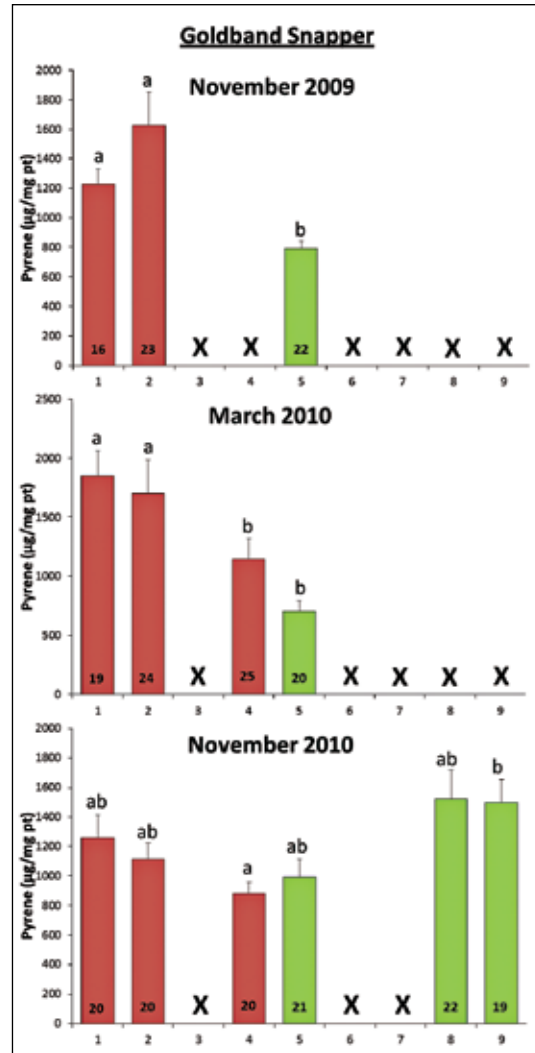


Figure 36. Pyrene type metabolite concentrations in the bile of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

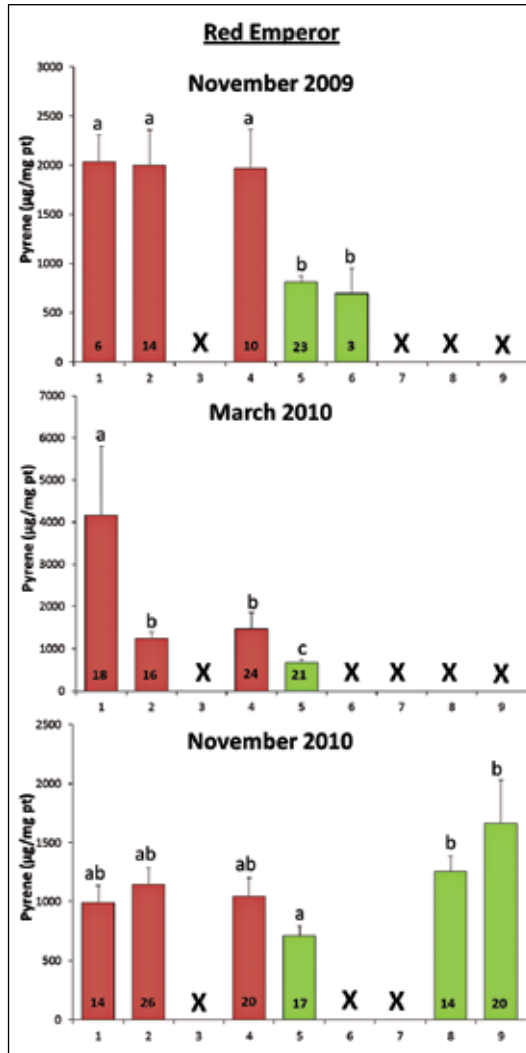


Figure 37. Pyrene type metabolite concentrations in the bile of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

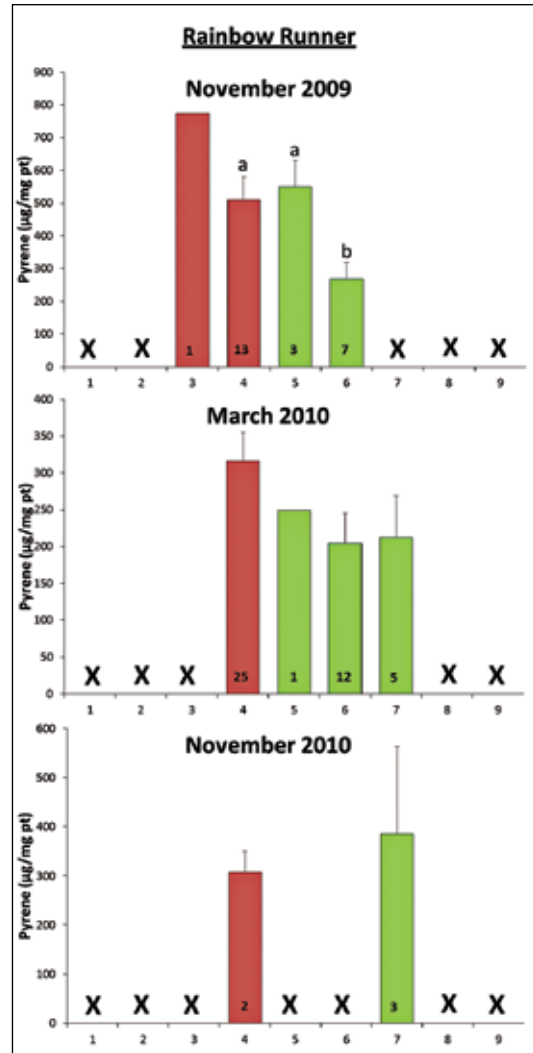


Figure 38. Pyrene type metabolite concentrations in the bile of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

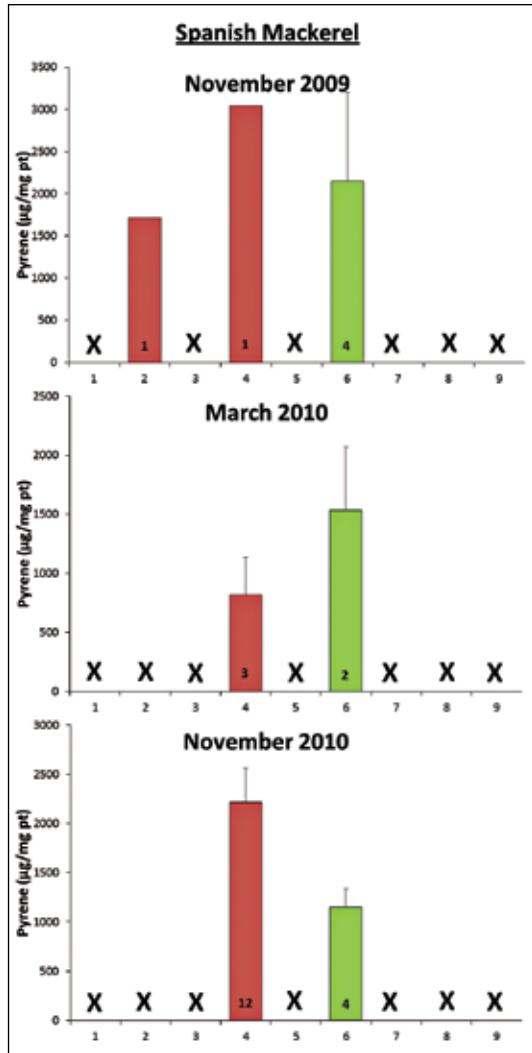


Figure 39. Pyrene type metabolite concentrations in the bile of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

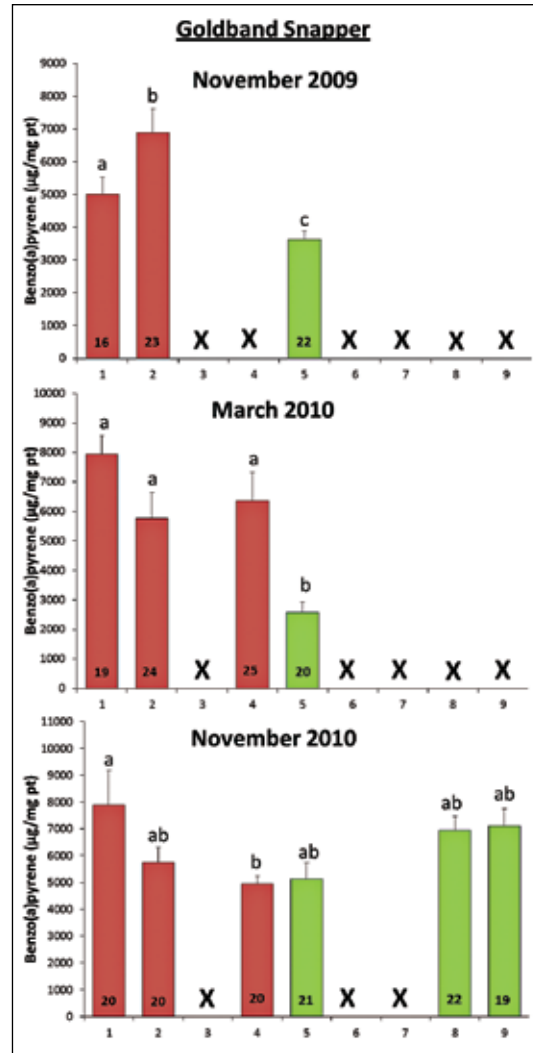


Figure 40. Benzo(a)pyrene type metabolite concentrations in the bile of goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

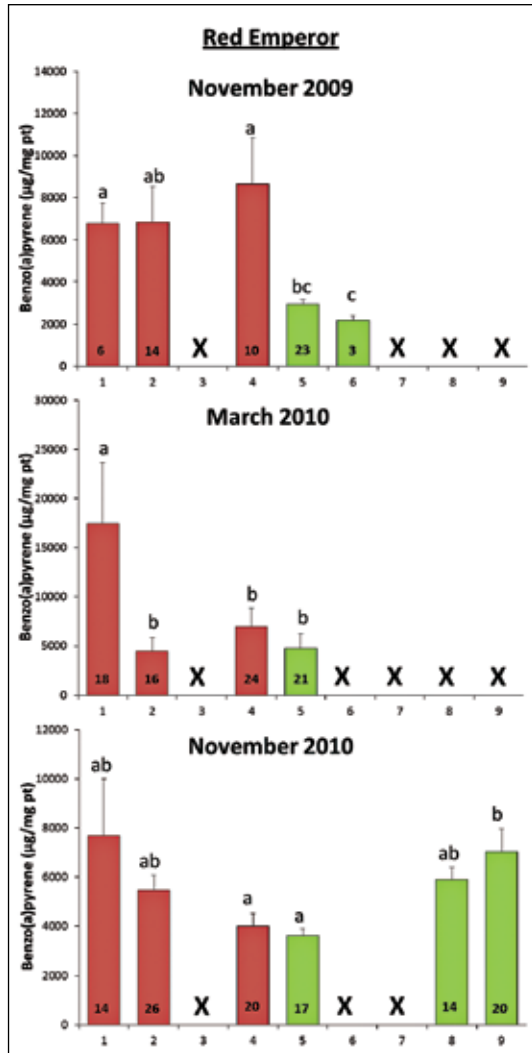


Figure 41. Benzo(a)pyrene type metabolite concentrations in the bile of red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

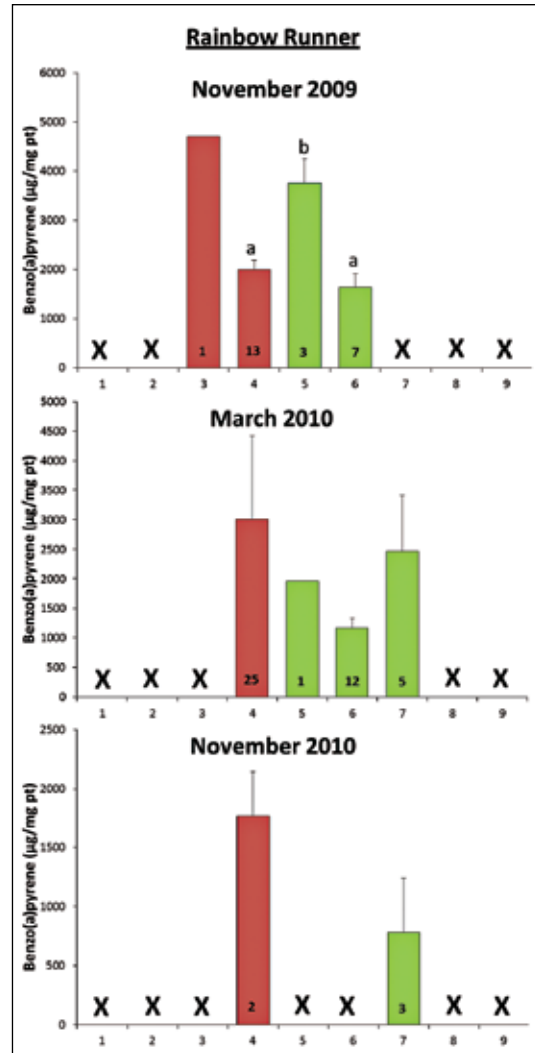


Figure 42. Benzo(a)pyrene type metabolite concentrations in the bile of rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

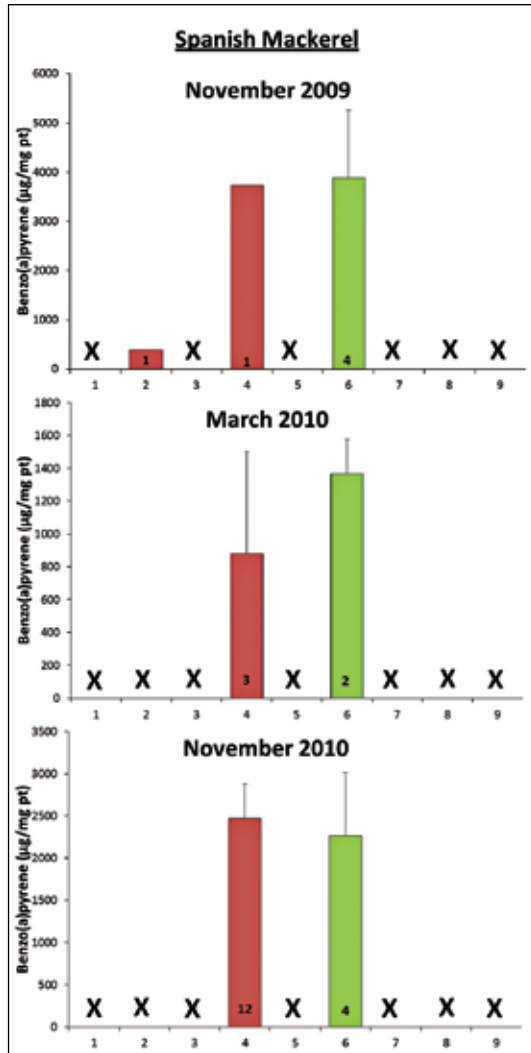


Figure 43. Benzo(a)pyrene type metabolite concentrations in the bile of Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

Sorbitol Dehydrogenase (SDH) Activity

The hepatic enzyme sorbitol dehydrogenase (SDH) is mainly involved in energy metabolism and is normally present in healthy animals including mammals and fish. Where hepatic injury has been sustained or hepatic function otherwise compromised, SDH can be released to the bloodstream and its measurement in blood is a valuable biomarker of a decreased ability of the liver to perform normal functions. Previous work has shown that such increases in SDH in the bloodstream can follow exposure to organic contaminants including petrogenic compounds (e.g., PAHs) (Ozetric and Krajnovic-Ozetric, 1993) and that SDH activity is not affected by conditions such as reproductive status which often are confounding factors in the interpretation of other biomarkers. An elevated SDH activity in the bloodstream is a biomarker of biologically significant adverse effect. Fish livers with cellular injuries related to xenobiotic exposure are less capable of performing metabolic activities (Holdway *et al.*, 1994) and in the long term, the metabolic performance of the organism may be compromised and affect growth and reproductive outputs.

In Phase I sampling (November 2009) the SDH activity in the goldband snapper, rainbow runner and the Spanish mackerel collected from the hydrocarbon-affected zones was comparable to that in fish from the reference areas, indicating that the presence of petroleum hydrocarbons in the environment did not affect liver integrity in these species. Similarly, none of the fish populations (goldband snapper, red emperor, rainbow runner, Spanish mackerel) sampled close to the rig in Phases II and III had increased SDH relative to the reference areas indicating that exposure to any petroleum hydrocarbons as a result of the Montara well release has not caused chronic hepatic damage.

In Phase I a small number of individual red emperor captured in the vicinity of the West Atlas rig had highly elevated SDH in the bloodstream indicating liver damage. However, in Phase II no such individuals were captured with SDH levels close to the rig (and elsewhere) relatively uniform.

Overall, results gathered using this biomarker indicate that the Montara well release has not resulted in any chronic hepatic damage in resident populations of the four target species in this study. Increased SDH in some individual red emperor immediately following the release was not sustained to translate in chronic liver damage.

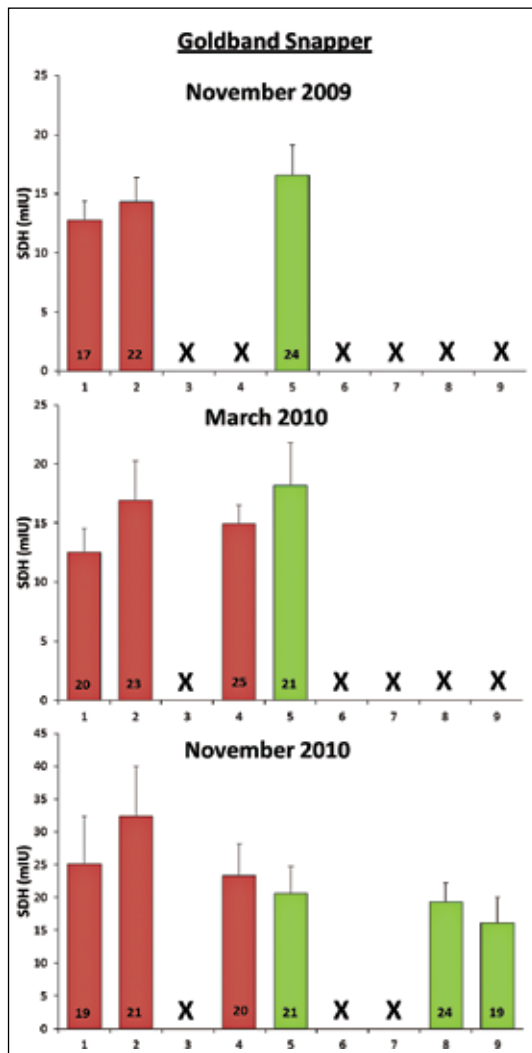


Figure 44. SDH activity in goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

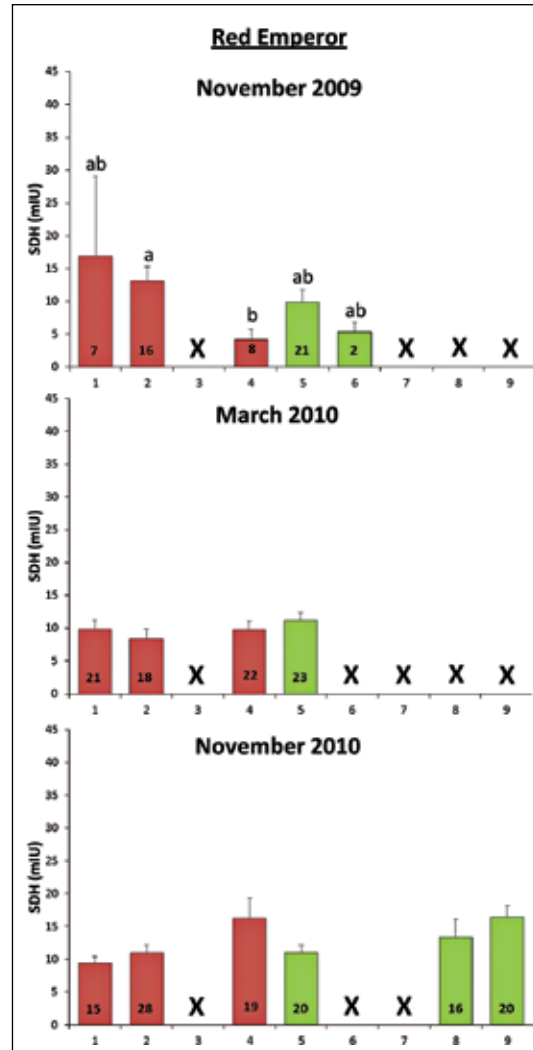


Figure 45. SDH activity in red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

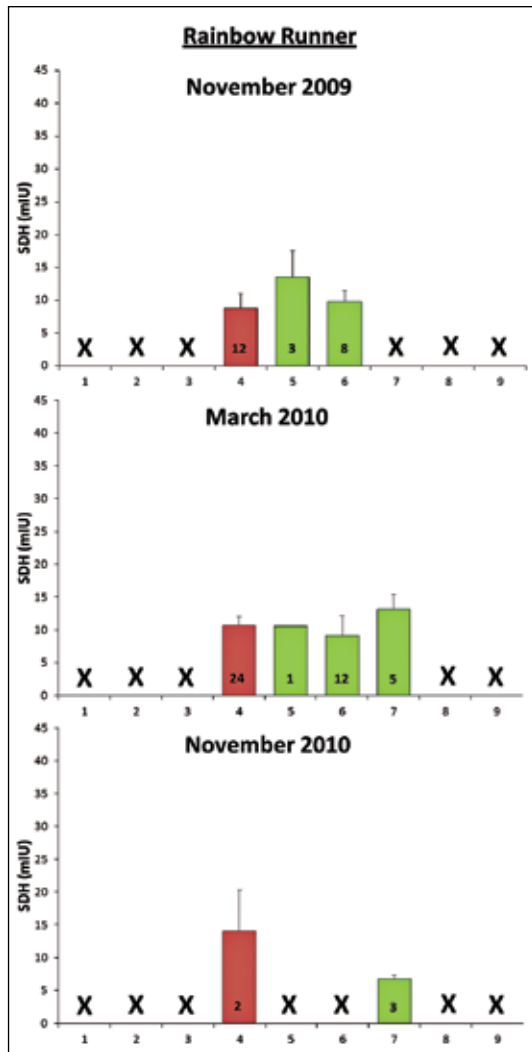


Figure 46. SDH activity in rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

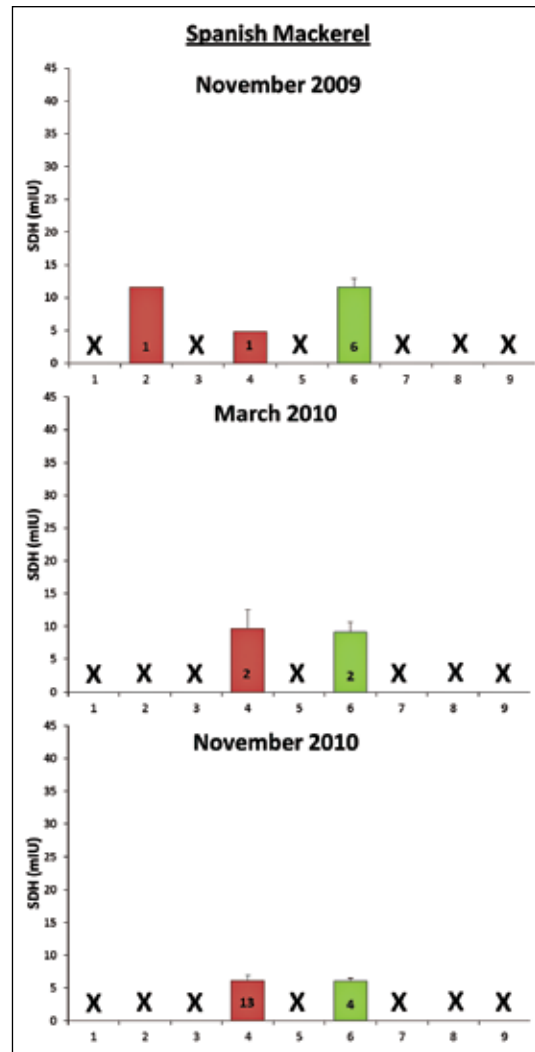


Figure 47. SDH activity in Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

Oxidative DNA Damage (8-oxo-dG Concentration)

Oxidative DNA damage is a normal event in fish (and other animals). Some causes originate from internal processes and metabolism but exposure to certain environmental factors (sunlight, free radicals) can increase the incidence of such damage. Exposure to PAHs (including petrogenic PAHs) has been a well-studied cause of the oxidative DNA damage measured in this study (reviewed by Pavanello and Clonfero, 2000). It should be noted that between species differences in the rate of oxidative DNA damage are normal and therefore no comparison can be made between species.

In the Phase I sampling period there was no difference in oxidative DNA damage between goldband snapper, rainbow runner and red emperor captured in the impacted and reference zones. The only statistically significant difference in this biomarker was in the red emperor where Site 5 (within the reference zone) was significantly higher than two of the sites within the impacted zone (Sites 2 and 4). This is the opposite of the trend expected if those fish captured close to the West Atlas drilling rig had been exposed to contaminants capable of causing oxidative DNA damage. Therefore this difference was clearly not the result of exposure to petroleum compounds from the Montara well release but some other unexamined factor.

Goldband snapper, and rainbow runner showed the same response in Phase II as in Phase I with no significant differences in oxidative DNA damage between fish captured in the impacted areas and the reference areas. While these fish may have been exposed to petroleum compounds from the Montara well release the biotransformation of these compounds to forms which can adversely interact with DNA has not occurred. The degree of replication in Spanish mackerel in Phase II does not allow the drawing of definitive conclusions on differences in oxidative DNA damage.

Red emperor captured closer to the rig had significantly higher levels of oxidative DNA damage than those captured in the reference zone.

The population of fish with the highest DNA damage were those 20 NM from the rig (site 1). Oxidative DNA damage is a biomarker of effect since the link between increased damage (including the serum concentrations of 8-oxo-dG) and carcinogenesis is well established. Immediately following the well release no increase in DNA damage could be measured in fish captured close to the rig (Phase I) but 5 months later there is evidence to indicate that red emperor living close to the rig (<20 NM) are at greater risk of carcinogenesis. It is unlikely that fish living at depth (e.g., red emperor) will be directly exposed to oil released from the Montara field since it is considered a light high wax crude oil. However secondary exposure through ingestion by prey or adhesion to large particles that can later sink to the ocean floor is possible. This exposure is likely to occur sometime after the initial release and could explain why the response measured in Phase II was not present in Phase I. Such exposure would be of short duration as it has been reported that oil degradation is a rapid process in the Timor Sea, and that it is unlikely that spilled oil even does reach the ocean floor (Burns *et al.*, 2010).

Further, this trend was not evident in red emperor captured during Phase III of the study. At this time there was a statistical difference between the DNA damage measured in fish collected from Site 8 and those collected from Site 1. This difference appears unlikely to be related to the Montara release.

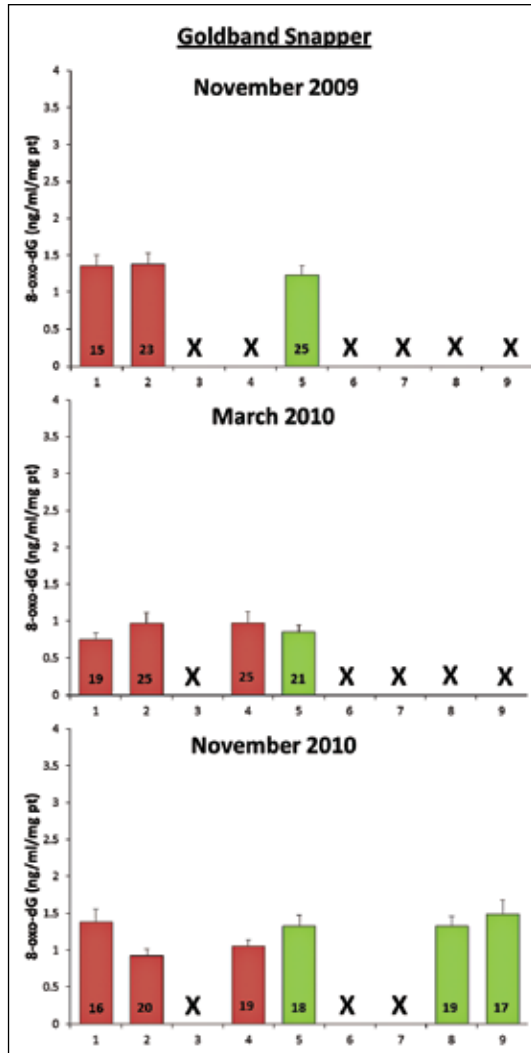


Figure 48. Oxidative DNA damage in goldband snapper captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

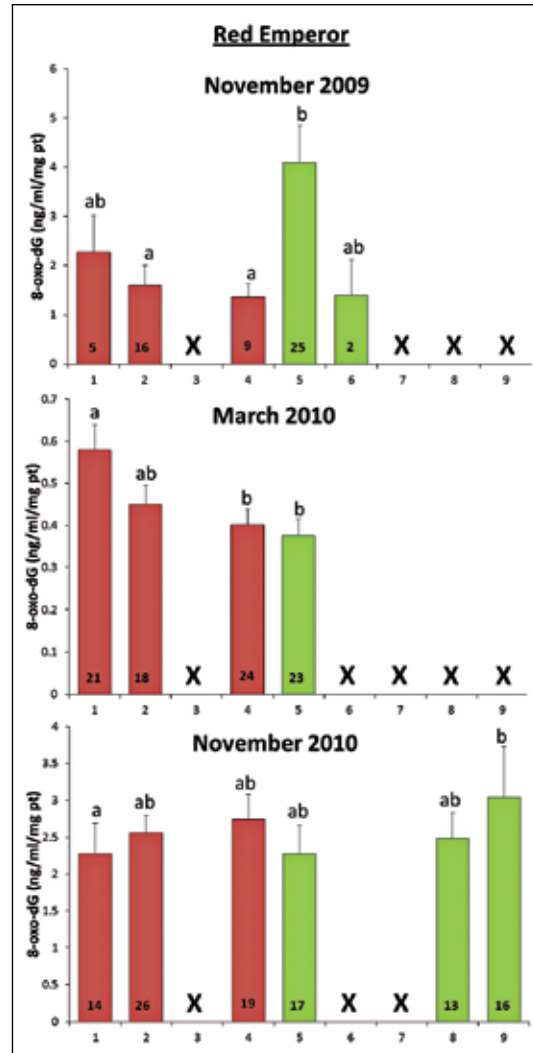


Figure 49. Oxidative DNA damage in red emperor captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

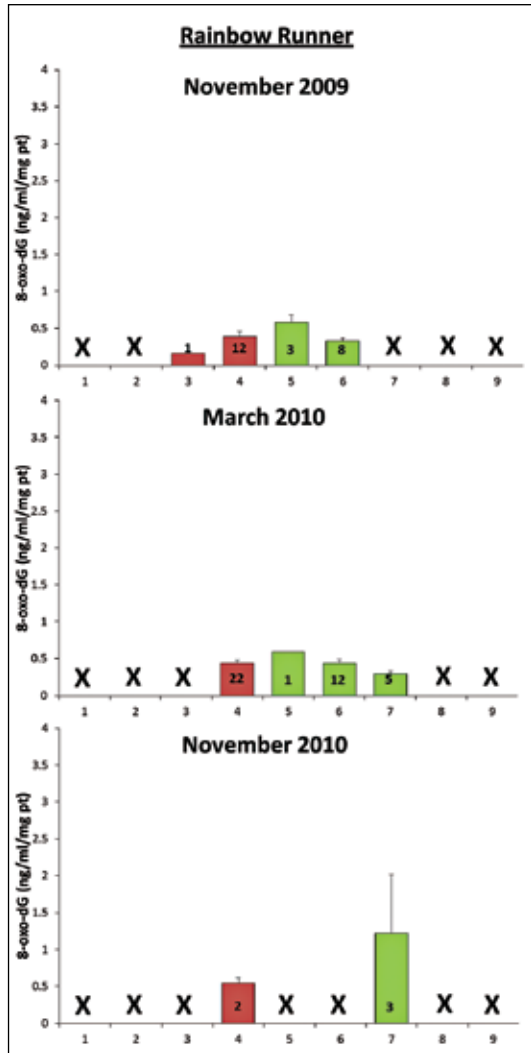


Figure 50. Oxidative DNA damage in rainbow runner captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

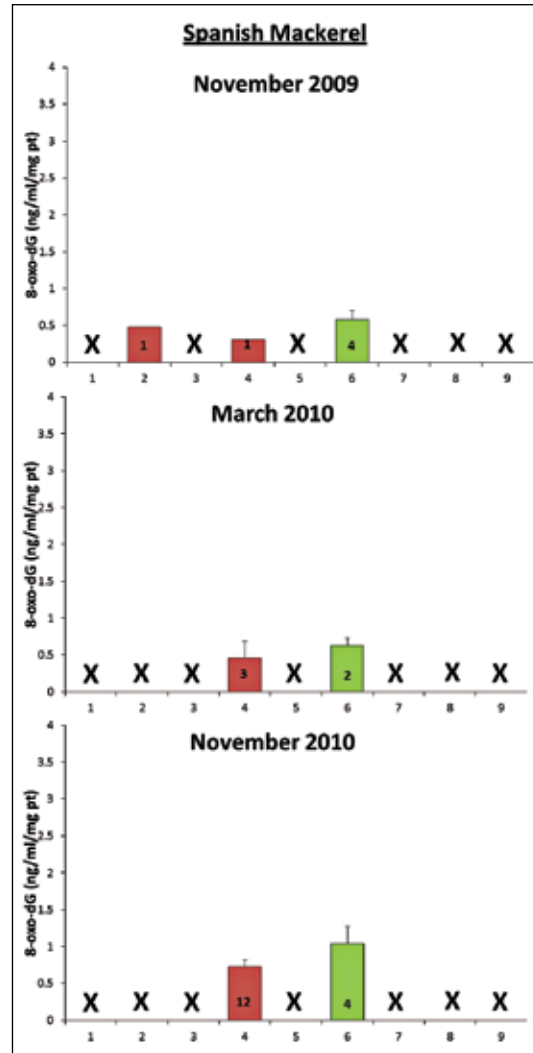


Figure 51. Oxidative DNA damage in Spanish mackerel captured at study sites in the Timor Sea in November 2009, March 2010 and November 2010. “Impacted” sites are in red and “reference” sites are in green. X denotes that no fish of this species were examined from this site. Numbers in columns denote the number of fish included in the analysis. Different letters above the columns denote significant difference ($\alpha = 0.05$).

Multivariate Analysis

Multivariate analysis is a powerful tool for visualising and explaining overall trends across a number of variables. In this case, these variables were the key measured biomarkers measured in the study (SDH, oxidative DNA damage, condition factor, LSI, naphthalene metabolites, pyrene metabolites and benzo(a)pyrene metabolites and EROD activity). Only the results for goldband snapper and red emperor are discussed as the low catch rate of pelagic species reduced the efficacy of the analysis.

In Phase I there were overall significant differences between goldband snapper captured at the impacted sites (site 1 and site 2) and the reference site (site 5) (ANOSIM). Principal Components Analysis (PCA) showed that these differences were along a component (PC1) which was strongly explained by pyrene and benzo(a)pyrene metabolites and to a lesser extent by naphthalene metabolites. The two impacted sites were separated from the reference site by increasing levels of these biomarkers. Other biomarkers contributed little to this separation between sites. In phase III there was a similar trend in separation of sites along an axis explained by all bile metabolites and there was a significant difference between each of the impacted sites and the reference site when all biomarkers were considered together (ANOSIM). In Phase III there were a greater number of statistical differences between the sites (ANOSIM) separation of sites was greater (e.g., the impacted sites 1 and 2 were considered different whereas in Phases I and II they were considered statistically similar). A PCA of this data indicates that there appeared to be a decreased separation of individual animals and collectively by collection site along an axis strongly explained by increasing levels of bile metabolites.

Similar trends were found when the results for the biomarkers measured in red emperor were analysed using these techniques. In Phase I, the reference site (site 5) was found to be statistically different to the other sites in the analysis (ANOSIM). A PCA including the same biomarkers as above indicated a separation of these sites along PC1 which was influenced by the bile metabolite measures. In Phase II the differences between the reference site (site 5) and the 2 impacted sites located nearest the West Atlas drilling rig (sites 1 and 2) remained but there was no difference between the impacted site 4 and this reference site. As with goldband snapper there was an increase in the reported differences between sites (ANOSIM) in Phase III and PCA indicated that this was mostly along PC1 which was explained mostly by differences in bile metabolite measures.

Despite, the PCA plot suggests an increasing overlap of individuals from different sites indicating a convergence of the overall biomarker signal.

These analyses indicate that the measurement of bile metabolites is the most sensitive of these biomarkers in identifying differences between fish captured at different study sites in the Timor Sea. If fish were exposed to a single pulse of hydrocarbons from the Montara well release it would be expected that the separation between sites due to increases in levels of these metabolites would decrease through time. The sequential PCA plots for both goldband snapper and red emperor indicate that this may broadly be the case with increasing overlap between individuals from the different study sites through time.

It should be noted that the observed increased in statistical difference between the sites may be due to the increased power in these tests to detect a statistical difference associated with increasing replication. Care must be taken in interpreting the results on ANOSIM with increased replication. The algorithms associated with this analysis mean that increased replication could eventually lead to biologically irrelevant statistical differences being reported.

Care should also be taken in attributing cause and effect based on the above analyses. There are number of factors which could contribute to variation between individuals and populations which have not been included in this analysis. The most important of these is location (latitude). In Phase I, both goldband snapper and red emperor showed a neat separation between the impacted sites 1 and 2 (grouped together by ANOSIM) and the reference site 5. Site 5 is located approximately 80 NM south of site 1 and 60 NM south of site 2, It is possible that the variation noted in the analysis is simply a latitudinal gradient. It is also possible that the differences could be due to continued exposure to hydrocarbons from natural sea floor seeps which are known to occur in the area (Burns *et al.*, 2010) or that if the study were to extend to sites at lower latitudes that a continued increase in measured bile metabolites would be observed beyond the site of the Montara well release. Finally, it is also possible that despite the continuing trend towards convergence observed through time, that the inter-site differences observed might be largely due to natural variations between fish populations. These results suggest that the fish at the different sites express the biomarkers differently and that the bile metabolites are the most sensitive biomarker in detecting this difference. The PCA suggests that the differences between sites are decreasing through time.

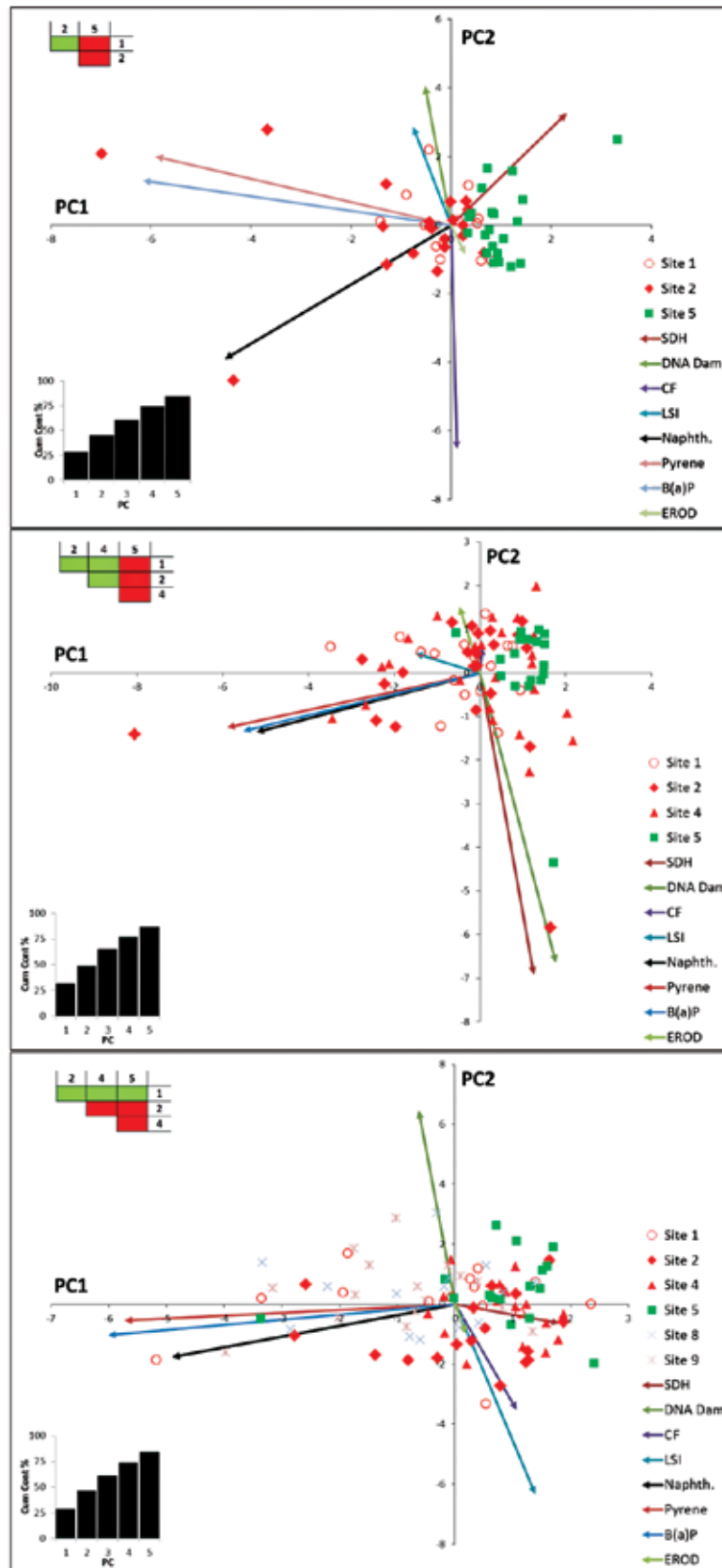


Figure 52. PCA on normalised data for combined biomarkers across study sites for goldband snapper. Top: Phase I, Middle: Phase II, Bottom Phase III. Insert histogram represents cumulative contribution of PCs used in the analysis. Insert matrix shows significant differences between sites as identified by analysis of similarity (ANOSIM) Euclidean distance.

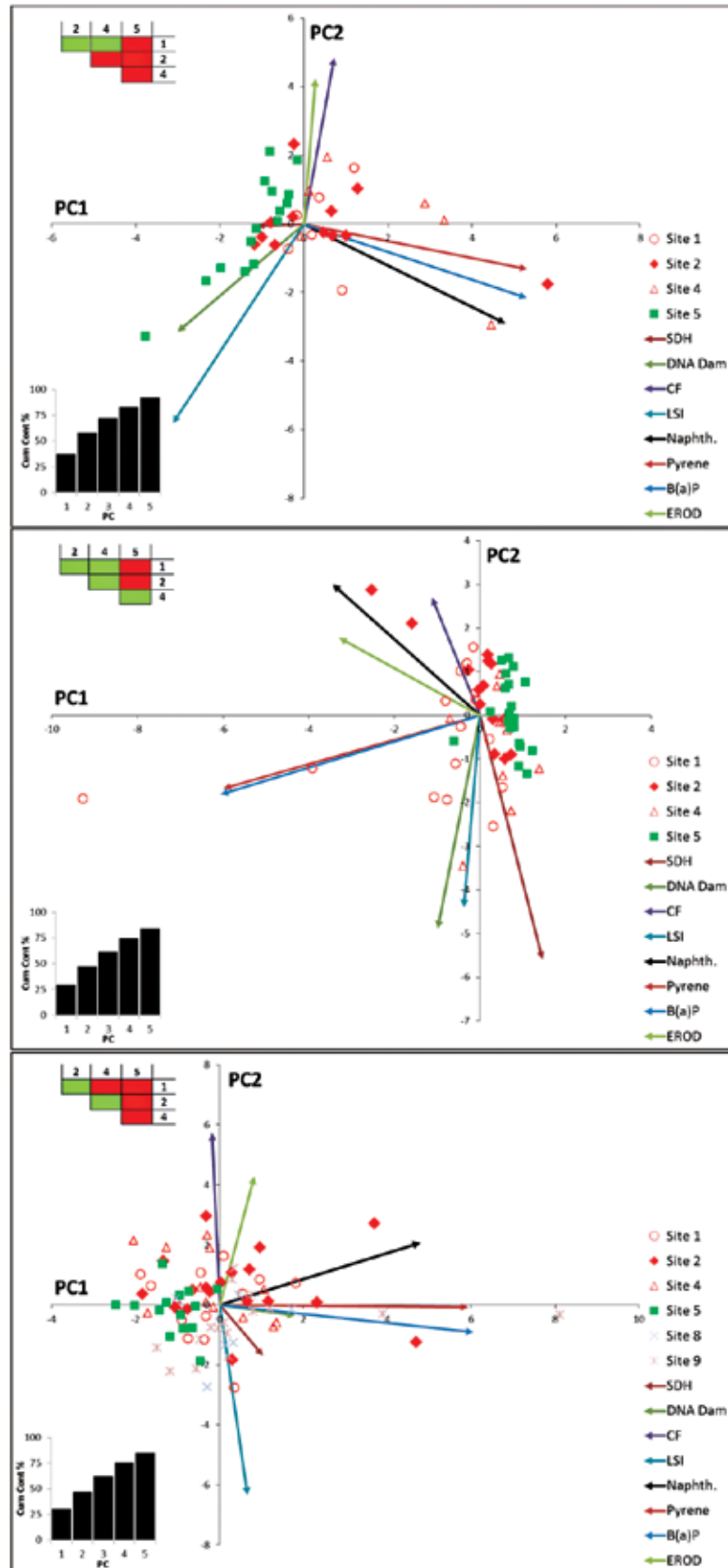


Figure 53. PCA on normalised data for combined biomarkers across study sites for red emperor. Top: Phase I, Middle: Phase II, Bottom Phase III. Insert histogram represents cumulative contribution of PCs used in the analysis. Insert matrix shows significant differences between sites as identified by analysis of similarity (ANOSIM) Euclidean distance.



Conclusions

The Montara well release that occurred in August 2009 released an estimated 23,000 barrels of oil and gas condensate in the Timor Sea over seventy-four days, and caused immediate concerns regarding the adverse impacts on commercial fisheries. To assess the potential impacts of hydrocarbon exposure on fish health, an investigation was undertaken to collect biopsies on two demersal and two pelagic fish species found in the impacted areas of the Timor Sea, and compare observations to those made in reference, non-impacted areas. The investigation evaluated fish health using physiological indices, reproductive histology, and biochemical markers on goldband snapper, red emperor, rainbow runner and Spanish mackerel. Biopsies were collected immediately after the end of the well release in November 2009 (Phase I), and again four months after the end of the spill in March 2010 (Phase II) and a year after the well release in November 2010 (Phase III) on a total of 781 fish.

The initial Phase I investigation revealed that exposure of surface and deep sea fish to petroleum hydrocarbons had occurred, but no ill effects could be identified consistently in any of the selected fish species. Phase I results showed that although uptake of petroleum hydrocarbons by the fish was confirmed by high biliary PAH metabolite contents fish were in good physical condition; physiological indices CF, LSI and GSI were unchanged following exposure to petroleum hydrocarbons; no alteration of gonadal development could be detected by histological examination; liver detoxification and liver integrity were unchanged; and DNA integrity was not compromised in hydrocarbon fish of all species. Overall, Phase I confirmed exposure to petroleum hydrocarbons, but limited ill effects could be detected in a small number of individual fish only, and no consistent adverse effects of exposure on fish health could be detected within two weeks following the end of the well release.

Phase II of the study investigated possible chronic effects related to the release of petroleum hydrocarbons in the marine environment. Four months following the end of the well release, evidences of continuing uptake of petroleum hydrocarbons was still apparent in fish collected within 20 NM from the West Atlas rig. Results from Phase II indicated that, relative to reference areas, fish collected in the hydrocarbon-affected zones remained in good physical condition, with red emperor showing a higher CF in fish captured closest to the rig; red emperor collected closest to the rig also had larger livers, while other species had unchanged liver size; male goldband snapper and female rainbow runner captured close to the rig had smaller gonad size relative to body weight while other species showed no inter-site difference; no alteration of gonadal development could be detected by histological examination in all species; red emperor, goldband snapper and rainbow runner collected in the impacted zones had elevated liver detoxification activity; liver integrity was unchanged in all fish species; Oxidative DNA damage was unchanged in goldband snapper and rainbow runner, but was increased in red emperor collected within 20 NM from the rig. Overall, Phase II confirmed continuing exposure to, and metabolism of, petroleum hydrocarbons, especially in goldband snapper and red emperor collected closest to the rig. Biologically relevant effects persisted four months following the end of the well release, with higher than expected condition factor, liver size and oxidative DNA damage levels in red emperor collected closest to the rig.

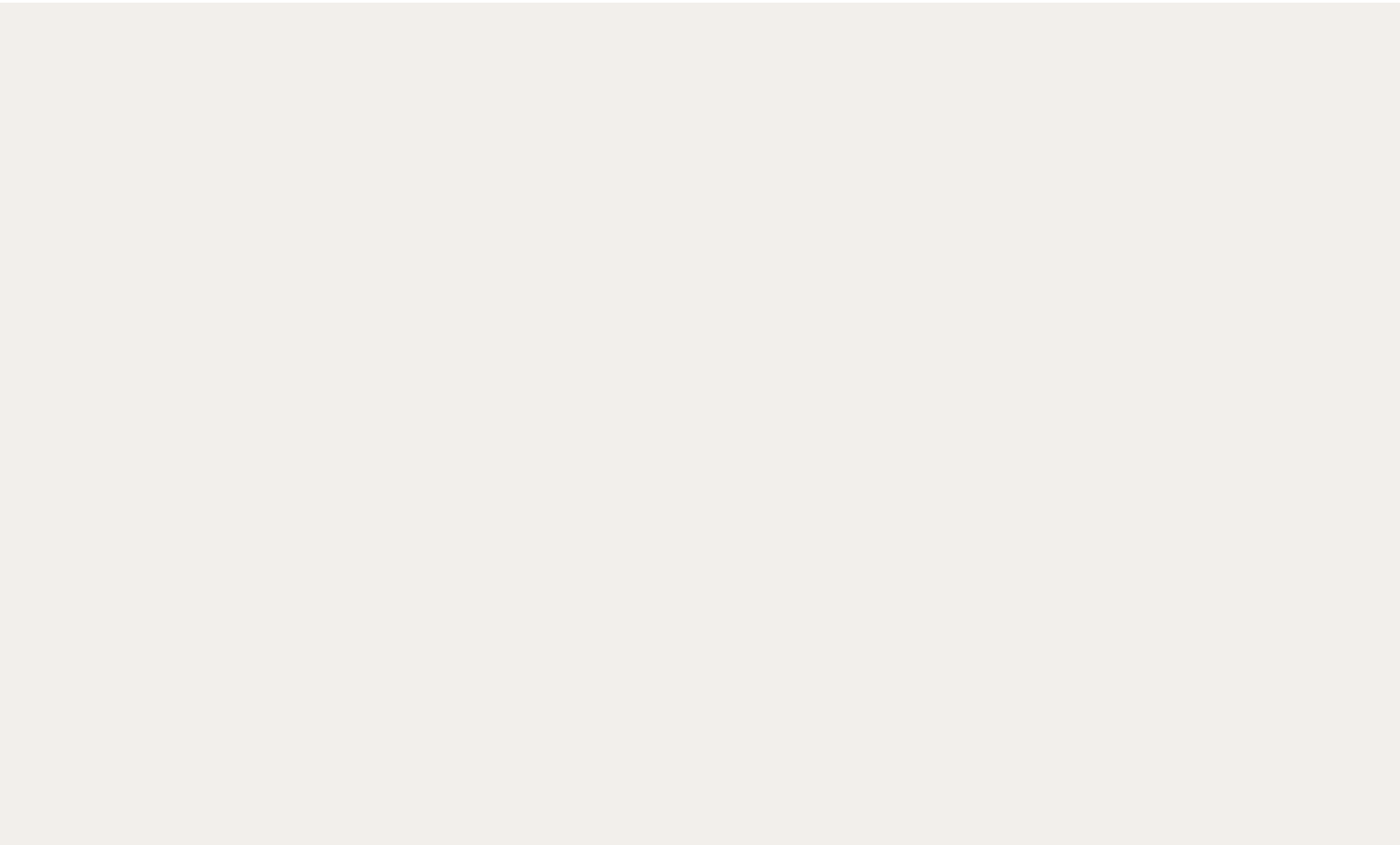
The biomarker results from Phase III indicate that the effects which may be related to exposure to petroleum hydrocarbons were reduced compared to the previous sampling periods. There were variations in the physiological indices condition factor, LSI and GSI, particularly in goldband snapper and red emperor from the different study sites, but in general the trends did not suggest any link with the location of the West Atlas drilling rig. The only results which could potentially be related to exposure to petroleum hydrocarbons was the GSI of male red emperor which was reduced at the impacted sites compared to the reference site. Similarly, there were no trends in the levels of serum SDH, oxidative DNA damage or EROD activity which could be related an exposure to petroleum hydrocarbons originating from the Montara well head. Phases I and II of this study noted higher levels of PAH metabolites in the bile of fish captured at sites close to the West Atlas drilling rig than in those captured at reference sites. Overall, in Phase III this trend was not apparent with fish from these impacted sites exhibiting similar levels of PAH metabolites as those from reference sites. This suggests a reduction in the exposure of these fish to petroleum hydrocarbons relative to those captured in previous phases of this study.

The results of a multivariate analysis which included data for all biomarkers measured in goldband snapper and red emperor (except GSI) indicates that during Phases I and II there were significant differences between each impacted site (Sites 1 and 2) and the reference site (Site 5) and that this difference was mostly due to the levels of PAH metabolites in the bile of the fish. In Phase III dissimilarities between the sites were still present and the contribution of PAH metabolites to these differences was still strong. However, the differences between impacted and non-impacted sites were not as well defined, with the impacted and non-impacted areas tending to converge towards a similar overall biomarker signal. This finding points to a reduction in the differences between the sites and, given the importance of PAH metabolites in these differences, to a reduction in exposure to petroleum hydrocarbons at the impacted sites.

The oil which was released from the Montara well head was very light and had a low content of high molecular weight PAHs. The high wax content of the oil released from the Montara well head possibly formed a buoyant waxy residue upon weathering, which could be consumed by animals and result in the entry of petroleum hydrocarbons into the food chain. While vertebrates can metabolise petroleum hydrocarbons quite efficiently, invertebrates have very limited metabolism potential and could be the link between surface waxy residues and deep sea fish. It is also likely that suspended particulates with adhering petroleum compounds would eventually settle to the sea floor and represent a temporary source of petroleum compounds for fish captured closest to the rig. It has been suggested that environmental conditions in the Timor Sea, e.g. warm water temperatures, favour a rapid degradation of oil making it unlikely that spilled oil does reach the ocean floor (Burns *et al.*, 2010). Corroborating this postulate is the intensive sediment sampling conducted around the rig in 2010, which failed to find any petroleum hydrocarbons in this environment (Appendix F).

Phase II of the investigation demonstrated that chronic effects can be seen more clearly in red emperor than in other species, which might relate to the long lifespan and biology of this species. It is not known if the effects observed in individual red emperor have the potential to translate into population-level effects but since no gross pathologies and no reproductive impairments were observed in adult fish, population-level effects appear unlikely. When considering the long term effects of the Montara well release on fish populations, it is important to consider that the Timor Sea goldband snapper and red emperor populations are commercially harvested, which can create significant population level effects non-related to the well release. In this context where multiple stakeholders operate in a common environment, potential effects on fish populations can be additive, interactive or synergistic. It is therefore difficult to isolate the effect of individual activities on fish populations.

Eventually, the petroleum hydrocarbons released during the Montara incident will be dissipated and degraded in the marine environment and will no longer be detectable in the fish living close to the rig, at which time biomarkers of exposure and biomarkers of biologically relevant effects will be similar in impacted and reference zones. From previous similar situations worldwide, no links have been established to relate directly the observed effects on individuals to population level impacts. Future monitoring in the surrounding of the Montara oil fields will confirm whether the trend towards convergence of the biomarker signal of impacted and non-impacted sites continues. In addition, future sampling of demersal fish collected in the area will provide a baseline for evaluating the environmental impacts, if any, once petroleum production has commenced at this location.



References

- AMSA [Australian Maritime Safety Authority], 2009. Montara Well Release Timor Sea: Operational Monitoring Study O2 Monitoring of Oil Character, Fate and Effects Report O2 Water Quality Report 2. 14th December 2009. Available at: http://www.amsa.gov.au/marine_environment_protection/national_plan/Reports-Fact_Sheets-Brochures/
- Bjerregaard L.B., Madsen A.H., Korsgaard B., Bjerregaard P., 2006. Gonad histology and vitellogenin concentrations in brown trout (*Salmo trutta*) from Danish streams impacted by sewage effluent. *Earth Environ. Sci.* 5: 315-327.
- Burns K. A., Brinkman D. L., Brunskill G. J., Volk H., Wasmund K., Zagorskis I., 2010. Fluxes and fate of petroleum hydrocarbons in the Timor Sea ecosystem with special reference to active hydrocarbon seepage. *Mar. Chem.* 118: 140-155.
- Codi-King S., Johnson J. E., Haasch M. L., Ryan D. A. J., Ahokas, J. T., Burns, K. A., 2005. Summary results from a pilot study conducted around an oil production platform on the northwest Shelf of Australia. *Mar. Poll. Bull.* 50: 1163-1172.
- Collier T.K., Anulacion B.F., Stein J.E., Goksoyr A., Varanasi U., 1995. A field evaluation of cytochrome P4501A as a biomarker of contaminant exposure in three species of flatfish. *Environ. Toxicol. Chem.* 14:143-152.
- Cooke S.S., Evans M.D., Dizdaroglu M., Lunec J., 2003. Oxidative DNA damage: mechanisms, mutation, and disease. *The FASEB Journal* 17, 1195-1214.
- French B.L., Reichert W.L., Hom T., Nishimoto M., Sanborn H.R., Stein J.E., 1996. Accumulation and dose-response of hepatic DNA adducts in English sole (*Pleuronectes vetulus*) exposed to a gradient of contaminated sediments. *Aquat. Toxicol.* 36: 1-16.

- Gagnon M.M., Holdway D.A., 2000. EROD Induction and Biliary Metabolite Excretion Following Exposure to the Water Accommodated Fraction of Crude Oil and to Chemically Dispersed Crude Oil. *Arch. Environ. Contam. Toxicol.* 38: 70-77.
- Gagnon M.M., Holdway D.A., 2002. EROD induction, serum SDH and PAH biliary metabolites in sand flathead (*Platycephalus bassensis*) collected in Port Philip Bay, Australia. *Mar. Poll. Bull.* 44: 230-237.
- Hartung R., 1995. Assessment of the potential for long-term toxicological effects of the Exxon Valdez oil spill on birds and animals. IN: Exxon Valdez oil spill: Fate and Effects in Alaskan waters, ASTM, pp. 693-725.
- Hellou, J., Payne, J.F., 1987. Assessment of contamination of fish by water-soluble fractions of petroleum: a role for bile metabolites. *Environ. Toxicol. Chem.* 6: 857-862.
- Hodson P.V., Kleopfer-Sams P.J., Munkittrick K.R., Lockhart W.L., Metner D.A., Luxon P.L., Smith I.R., Gagnon M.M., Servos M, Payne J.F., 1991. Protocols for measuring mixed function oxygenases of fish liver. *Can. Tech. Rep. Fish. Aquat. Sci.* No 1829, 51 pages.
- Holdway D.A., Brennan S.E., Ahokas J.T., 1994. Use of hepatic MFO and blood enzyme biomarkers in sand flathead (*Platycephalus bassensis*) as indicators of pollution in Port Phillip Bay, Australia. *Mar. Pollut. Bull.* 26: 683-695.
- Lam P.K.S., Gray J.S., 2003. The use of biomarkers in environmental monitoring programmes. *Mar. Pollut. Bull.* 46: 182-186.
- Landis G., Yu M.H., 1995. Introduction to environmental toxicology: impacts of chemicals upon ecological systems. Lewis, Boca Raton, Florida.
- Lillie R.D., 1965. Histopathologic Techniques and Practical Histochemistry, 3rd edition, McGraw-Hill Book Co., New York
- Lin E.L.C., Cormier S.M., Torsella J.A., 1996. Fish biliary polycyclic aromatic hydrocarbon metabolites estimated by fixed-wavelength fluorescence: comparison with HPLC-fluorescent detection. *Ecotoxicol. Environ. Saf.* 35: 16-23.
- Lowry O.H., Rosebrough N.J., Farr A.L., Randall R.J., 1951. Protein measurement with the Folin reagent. *J. Biol. Chem.* 193: 265-275.
- Maccubbin AE. 1994. DNA adduct analysis in fish: laboratory and field studies. IN: Malins DC, Ostrander GK (Eds). Aquatic Toxicology; molecular, Biochemical and Cellular Perspectives. Lewis Publishers, CRC Press, pp. 267-294.
- Martinez-Gomez C., Fernandez B., Valdes J., Campillo J. A., Benedicto J., Sanchez F., Vethaak A. D., 2009. Evaluation of three-year monitoring with biomarkers in fish following the Prestige oil spill (N Spain). *Chemosphere.* 74: 613-620.
- Meador JP, Stein JE, Reichert WL, Varanasi U. 1995. Bioaccumulation of polycyclic aromatic hydrocarbons by marine organisms. *Rev. Environ. Contam. Toxicol.* 143:79-165.
- McDonald S. J., Kennicutt II, M. C., Liu H., Safe S. H., 1995. Assessing aromatic hydrocarbon exposure in Antarctic fish captured near Palmer and McMurdo Stations, Antarctica. *Arch. Environ. Contam. Toxicol.* 29: 232-240.
- Ozetric B. and Krajcnovic-Ozetric M., 1993. Plasma sorbitol dehydrogenase, glutamate dehydrogenase, and alkaline phosphatase as potential indicators of liver intoxication in grey mullet (*Mugil auratus* Risso). *Bull. Environ. Toxicol.* 50: 586-592.
- Pavanello S., Clonfero E., 2000. Biological indicators of genotoxic risk and metabolic polymorphisms. *Mut. Res.* 463: 285-308.
- Stein J.E., Reichert W.L., French B., Varanasi U., 1993. ³²P-Postlabeling analysis of DNA adduct formation and persistence in English sole exposed to benzo[a]pyrene and 7H-dibenzo[c,g]carbazole. *Chem.-Biol. Interact.* 88: 55-69.
- Valavanidis A., Vlachogianni T., Fiotakis C., 2009. 8-hydroxy-2'-deoxyguanosine (8-OHdG): a critical biomarker of oxidative stress and carcinogenesis. *J. Environ. Sci. Health C* 27: 120-139.
- Van der Oost R., Beyer J., Vermeulen N.P.E., 2003. Fish bioaccumulation and biomarker in environmental risk assessment: a review. *Environ. Toxicol. Pharmacol.* 13: 57-149.

Appendix A – Statistical Results Phase I

Table A1 – 1. Summary results for goldband snapper captured in Phase I. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
Condition Factor	1	17	11.17	0.39	log	2	0.25	0.780				
	2	23	11.40	0.38								
	5	25	10.99	0.25								
LSI	1	17	1.04	0.01	log	2	2.38	0.101				
	2	23	0.93	0.07								
	5	25	0.83	0.07								
GSI (male)	1	3	0.135	0.047		2	0.343	0.713				
	2	13	0.200	0.053								
	5	15	0.166	0.025								
GSI (female)	1	14	0.689	0.113		2	5.305	0.010	1	0.6895		
	2	10	1.454	0.267					5	0.7764		
	5	10	0.776	0.1612					2		1.4539	
EROD (pmol/min/mg pr)	1	16	13.1	1.3	log	2	2.36	0.103				
	2	22	12.8	1.9								
	5	25	13.8	0.7								
Naphthalene-type metabolites (mg/mg pr)	1	16	9235	763	log	2	32.4	<0.001	5	3.6260		
	2	23	12613	2448					1		3.9283	
	5	22	4040	276					2		4.0235	
Pyrene-type metabolites (µg/mg pr)	1	16	1228	104	log	2	14.5	<0.001	5	2.8733		
	2	23	1626	226					1		3.0714	
	5	22	769	50					2		3.1549	
Benzo(a) Pyrene-type metabolites (µg/mg pr)	1	16	5004	517	log	2	13.4	<0.000	5	3.5389		
	2	23	6880	735					1		3.6691	
	5	22	3745	241					2			3.7966
SDH (mIU)	1	17	13.00	2		2	1.30	0.280				
	2	22	14.34	2.039								
	5	24	16.57	2.577								
DNA damage (ng/ml/mg pr)	1	15	1.359	0.140		2	0.35	0.710				
	2	23	1.381	0.154								
	5	25	1.235	0.122								

Table A1 – 2. Summary results for red emperor captured in Phase I. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
Condition Factor	3	7	12.90	1.49	log	4	0.064	0.64				
	2	16	14.29	0.46								
	4	10	13.94	0.77								
	5	25	13.36	0.70								
	6	3	13.23	1.35								
LSI	1	7	0.94	0.09	log	4	2.164	0.085				
	2	16	0.95	0.07								
	4	10	1.05	0.10								
	5	25	1.26	0.09								
	6	3	0.98	0.23								
GSI (male)	1	1	0.054	–		4	2.144	0.166				
	2	3	0.240	0.028								
	4	4	0.190	0.088								
	5	4	0.045	0.019								
	6	1	0.104	–								
GSI (female)	1	7	1.595	0.573	log	4	2.547	0.053				
	2	13	2.946	0.549								
	4	6	5.291	0.565								
	5	–	–	–								
	6	2	3.633	1.357								
EROD (pmol/min/mg pr)	1	7	30.6	7.7	log	4	0.370	0.83				
	2	14	28.7	6.2								
	4	10	34.0	7.4								
	5	22	35.6	5.1								
	6	3	28.4	7.2								
Naphthalene-type metabolites (mg/mg pr)	1	6	9158	1004	log	4	2.800	0.03				
	2	14	8762	1944								
	4	10	9356	3805								
	5	23	4962	368								
	6	3	6155	1701								
Pyrene-type metabolites (µg/mg pr)	1	6	2033	278	log	4	8.860	<0.001	6	2.7788		
	2	14	1998	358					5	2.8714		
	4	10	1860	395					4		3.2144	
	5	23	812	64					2		3.2240	
	6	3	698	254					1		3.2837	

		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
Benzo(a) Pyrene-type metabolites (µg/mg pr)	1	6	6765	974	log	4	6.530	<0.001	6	3.3342		
	2	14	6847	1675					5	3.4396	3.4396	
	4	10	7876	2206					2		3.7245	3.7245
	5	23	2941	204					4			3.7948
	6	3	2182	223					1			3.8101
SDH (mIU)*	1	7	16.88	12.122	log	4	3.672	0.011	4	0.4198		
	2	16	13.14	2.111					1	0.7810	0.7810	
	4	8	4.18	1.588					6	0.7880	0.7880	
	5	21	9.85	1.929					5	0.8161	0.8161	
	6	2	5.31	1.447					2		1.0636	
DNA damage (ng/ml/mg pr)	1	5	2.27	0.751								
	2	16	1.60	0.408								
	4	9	1.37	0.264				0.190†				
	5	25	4.09	0.757								
	6	2	1.39	0.725								

* Tukeys post-hoc tests did not detect significant differences so the less conservative Bonferonni post-hoc tests used

† Non-parametric (Kruskal-Wallis) tests run due to variance heteroscedascity.

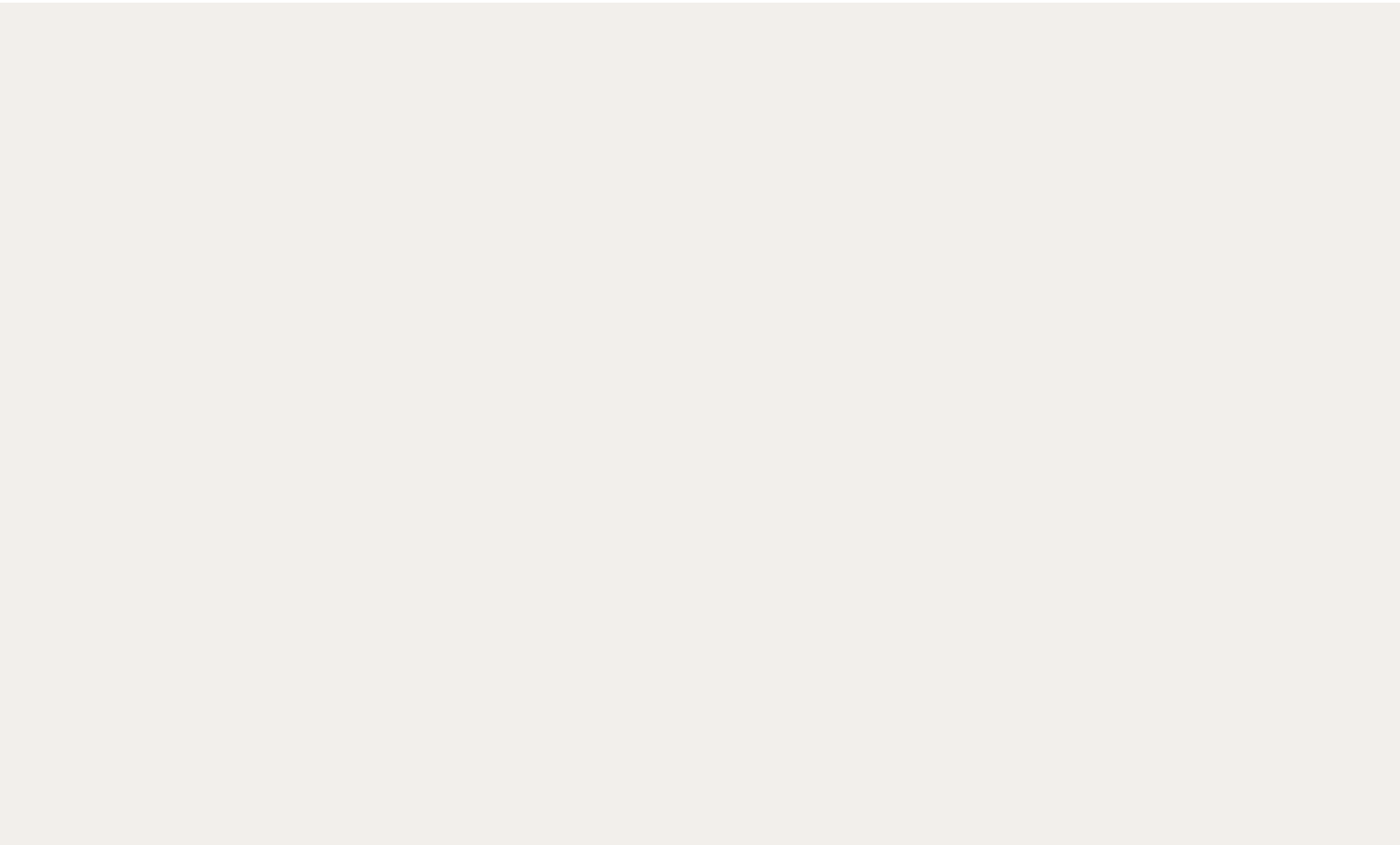
Table A1 – 3. Summary results for rainbow runner captured in Phase I. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets		
						df	F	sig	Site	1	2
Condition Factor	3	1	6.90		log	3	0.31	0.816			
	4	13	6.60	0.38							
	5	3	7.08	0.34							
	6	8	6.95	0.28							
LSI	3	1	1.12	–	log	3	0.61	0.614			
	4	13	1.00	0.16							
	5	3	0.71	0.04							
	6	8	1.02	0.09							
GSI (male)	3	–	–	–		2	2.041	0.181			
	4	7	2.356	0.342							
	5	3	3.412	0.139							
	6	3	4.878	0.419							
GSI (female)	3	1	4.176	–		2	1.501	0.274			
	4	6	3.350	0.436							
	5	–	–	–							
	6	5	4.878	0.819							
EROD (pmol/min/mg pr)	3	1	8.8	–	log	2	0.005	0.95			
	4		9.0	1.3							
	5	3	8.1	1.2							
	6	7	7.2	1.6							
Naphthalene–type metabolites (mg/mg pr)	3	1	3542	–	log	2	13.2	<0.001	6	3.1422	
	4	13	3333	361					5		3.4127
	5	3	2659	463					4		3.4904
	6	7	1391	39							
Pyrene–type metabolites (µg/mg pr)	3	1	774	–	log	2	6.3	0.008	6	2.381	
	4	13	509	71					4		2.6664
	5	3	550	80					5		2.7315
	6	7	267	360							
Benzo(a) Pyrene–type metabolites (µg/mg pr)	3	1	4705	–	log	2	7.7	0.003	6	3.1829	
	4	13	2005	185					4	3.2829	
	5	3	3751	496					5		3.5659
	6	7	1637	276							
SDH (mIU)	3	–	–	–		2	0.745	0.487			
	4	12	8.762	2.248							
	5	3	13.504	4.016							
	6	8	9.767	1.665							

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets		
						df	F	sig	Site	1	2
DNA damage (ng/ml/mg pr)	3	1	0.163	–		3	2.195	0.120			
	4	12	0.392	0.058							
	5	3	0.585	0.091							
	6	8	0.332	0.039							

Table A1 – 4. Summary results for Spanish mackerel captured in Phase I. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptives				Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	N	1	2
Condition Factor	3	1	6.90	–								
	4	1	6.60	–								
	6	4	5.63	0.24								
LSI	3	1	0.72	–								
	5	1	0.81	–								
	6	4	0.77	0.08								
GSI (male)	3	–	–	–								
	5	–	–	–								
	6	1	0.086	–								
GSI (female)	3	1	0.761	–								
	4	1	0.280	–								
	6	3	1.262	0.083								
EROD (pmol/min/mg pr)	3	1	12.7	–								
	4	1	8.3	–								
	6	4	6.9	3.2								
Naphthalene-type metabolites (mg/mg pr)	3	1	16548	–								
	4	1	18066	–								
	6	4	23165	9224								
Pyrene-type metabolites (µg/mg pr)	3	1	1716	–								
	4	1	3042	–								
	6	4	2151	1049								
Benzo(a) Pyrene-type metabolites (µg/mg pr)	3	1	394	–								
	4	1	3739	–								
	6	4	3889	1361								
SDH (mIU)	3	1	11.576	–								
	4	1	4.823	–								
	6	6	11.576	1.420								
DNA damage (ng/ml/mg pr)	3	1	0.475	–								
	4	1	0.301	–								
	6	4	0.580	0.122								



Appendix B – Statistical Results Phase II

Table A2 – 1. Summary results for goldband snapper captured in Phase II. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets		
						df	F	sig	Site	1	2
Condition Factor	1	20	18.62	0.903		3	2.054	0.112			
	2	25	18.38	0.263							
	4	26	17.18	0.258							
	5	21	18.15	0.218							
LSI	1	20	1.000	0.045		3	2.588	0.058			
	2	25	1.166	0.053							
	4	26	0.935	0.040							
	5	21	1.000	0.108							
GSI (male)	1	6	0.234	0.054		3	5.400	0.005	2	0.2081	
	2	12	0.208	0.036					1	0.2336	
	4	8	0.335	0.031					4	0.3346	0.3346
	5	3	0.523	0.137					5		0.5232
GSI (female)	1 2 4 5	13	1.983	0.220		3	1.820	0.153			
	2	13	1.553	0.234							
	4	18	2.120	0.093							
	5	18	1.684	0.170							
EROD (pmol/min/mg pr)	1	20	16.92	1.71	log	3	3.779	0.013	5	1.0390	
	2	25	17.74	1.35					1	1.1851	1.1851
	4	26	22.15	2.79					2		1.2192
	5	21	12.03	0.81					4		1.2568
Naphthalene-type metabolites (mg/mg pr)	1	19	7921	820	log	3	8.616	<0.001	1	3.5241	
	2	24	10209	1589					5	3.5879	
	4	25	6312	4105					4	3.7014	
	5	20	4087	1434					2		3.9326
Pyrene-type metabolites (µg/mg pr)	1	19	1850	209	log	3	10.002	<0.001	5	2.8239	
	2	24	1701	282					4	2.9385	
	4	25	1144	176					2		3.1491
	5	20	704	88					1		3.2231
Benzo(a) Pyrene-type metabolites (µg/mg pr)	1	19	7934	630	log	3	9.209	<0.001	5	3.3387	
	2	24	5768	862					4		3.6601
	4	25	6365	967					2		3.6832
	5	20	2574	354					1		3.8778

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets		
						df	F	sig	Site	1	2
SDH (mIU)	1	20	12.492	2.002		3	0.762	0.519			
	2	23	16.902	3.342							
	4	25	14.933	1.514							
	5	21	18.144	3.634							
DNA damage (ng/ml/mg pr)	1	19	0.755	0.084	log	3	2.393	0.074			
	2	25	0.968	0.143							
	4	25	0.976	0.149							
	5	21	0.853	0.094							

Table A2 – 2. Summary results for red emperor captured in Phase II. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
Condition Factor	1	21	26.27	0.868		3	4.410	0.006	5	23.8893		
	2	18	27.35	0.992					4	24.4206		
	4	24	24.42	0.592					1	26.2738	26.2738	
	5	23	23.89	0.590					2		27.3457	
LSI	1	21	0.997	0.059		3	5.221	0.002	5	0.7462		
	2	18	1.020	0.059					4	0.8543	0.8543	
	4	24	0.854	0.061					1		0.9971	
	5	23	0.746	0.042					2		1.0201	
GSI (male)	1	8	0.047	0.007		3	.869	0.468				
	2	4	0.042	0.011								
	4	8	0.107	0.024								
	5	10	0.244	0.167								
GSI (female)	1	12	1.432	0.208		3	3.254	0.030	1	1.4315471		
	2	12	1.571	0.199					2	1.5708651		
	4	15	2.145	0.195					5	2.1143668	2.1143668	
	5	12	2.114	0.213					4		2.1451688	
EROD (pmol/min/mg pr)	1	20	57.3	9.92	log	3	4.583	0.005	4	1.3306		
	2	17	50.9	12.91					5	1.3493		
	4	23	28.0	6.28					2	1.5286	1.5286	
	5	23	26.9	3.45					1		1.6507	
Naphthalene-type metabolites (mg/mg pr)	1	18	7912	819	log	3	4.749	0.004	5	3.5614		
	2	16	8152	2692					4	3.7063	3.7063	
	4	24	7249	2021					2		3.7798	
	5	24	3999	378					1		3.8559	
Pyrene-type metabolites (µg/mg pr)	1	19	4171	1633	log	3	14.923	<0.001	5	2.7800		
	2	16	1248	135					4		3.0108	
	4	24	1476	371					2		3.0646	
	5	21	673	69					1			3.3898
Benzo(a) Pyrene-type metabolites (µg/mg pr)	1	19	17440	6207	log	3	11.040	<0.001	2	3.5112		
	2	16	4450	1392					5	3.5251		
	4	24	6980	1877					4	3.6485		
	5	21	4721	1533					1		4.0551	

		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
SDH (mIU)	1	21	9.830	1.450		3	0.667	0.575				
	2	18	8.412	1.483								
	4	22	9.734	1.370								
	5	23	11.198	1.211								
DNA damage (ng/ml/mg pr)	1	21	0.578	0.061		3	3.844	0.013	5	1.5958		
	2	18	0.450	0.046					4	1.7918		
	4	24	0.401	0.038					2	2.1180		
	5	23	0.376	0.039					1		3.6944	

Table A2 – 3. Summary results for rainbow runner captured in Phase II. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets		
						df	F	sig	Site	1	2
Condition Factor	4	25	10.688	1.151		3	0.179	0.910			
	5	1	9.324	–							
	6	13	9.761	0.297							
	7	5	9.557	1.042							
LSI*	4	25	0.708	0.030							
	5	1	0.893	–							
	6	13	0.753	0.034							
	7	5	1.078	0.238							
GSI (male)	4	10	3.301	0.250		2	1.399	0.272			
	5	–	–	–							
	6	7	3.735	0.629							
	7	4	4.552	0.750							
GSI (female)**	4	15	2.289	0.194		3	8.564	0.001			
	5	–	–	–							
	6	6	3.288	0.337							
	7	1	3.880	–							
EROD (pmol/min/mg pr)	4	25	10.58	1.03	log	2	7.814	0.001	7	0.7484	
	5	1	9.20	–					6	0.8189	
	6	13	6.58	0.42					4		0.9840
	7	5	5.76	0.73							
Naphthalene–type metabolites (mg/mg pr)	4	25	1592	148	log	2	0.088	0.916			
	5	1	981	–							
	6	12	1415	221							
	7	5	1405	148							
Pyrene–type metabolites (µg/mg pr)	4	25	316	39.1	log	2	1.114	0.338			
	5	1	249	–							
	6	12	204	41.3							
	7	5	212	56.9							
Benzo(a) Pyrene–type metabolites (µg/mg pr)	4	25	3009	1410	log	2	0.226	0.799			
	5	1	1959	–							
	6	12	1173	158							
	7	5	2467	953							
SDH (mIU)	4	24	10.691	1.386		3	0.312	0.817			
	5	1	10.611	–							
	6	12	9.164	2.927							
	7	5	13.119	2.275							

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets		
						df	F	sig	Site	1	2
DNA damage (ng/ml/mg pr)	4	22	0.438	0.044		3	1.183	0.330			
	5	1	0.589	–							
	6	12	0.441	0.049							
	7	5	0.292	0.039							

*non-parametric no sig diff between sites, **no post-hoc as fewer than 3 groups.

Table A2 – 4. Summary results for Spanish mackerel captured in Phase II. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets		
						df	F	sig	Site	1	2
Condition Factor	4	3	8.547	1.131		1	0.088	0.786			
	6	2	8.114	0.005							
LSI	4	3	0.799	0.125		1	0.556	0.510			
	6	2	0.678	0.022							
GSI (male)	4	1	0.760	–							
	6	1	0.432	–							
GSI (female)	4	2	1.865	0.154							
	6	1	0.286	–							
EROD (pmol/min/mg pr)	4	2	12.85	1.95	log	1	7.676	0.109			
	6	2	8.25	0.25							
Naphthalene–type metab. (mg/mg pr)	4	3	8995	2181	log	1	0.008	0.935			
	6	2	10096	4795							
Pyrene–type metab. (µg/mg pr)	4	3	813	324	log	1	1.651	0.289			
	6	2	1534	535							
Benzo(a) Pyrene–type metab. (µg/mg pr)	4	3	879	620	log	1	0.111	0.771			
	6	2	1366	210							
SDH (mIU)	4	2	9.646	2.894		1	0.022	0.895			
	6	2	9.164	1.447							
DNA damage (ng/ml/mg pr)	4	3	0.459	0.222		1	0.331	0.605			
	6	2	0.629	0.010							

Appendix C – Statistical Results Phase III

Table A3 – 1. Summary results for goldband snapper captured in Phase II. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
Condition Factor	1	24	19.87237	0.223066		5	4.034	0.002	8	18.8178		
	2	35	19.60606	0.242196					2	19.6061	19.6061	
	4	23	19.76174	0.215754					4	19.7617	19.7617	
	5	31	20.157	0.17942					1		19.8723	
	8	24	18.81778	0.228236					9		19.9549	
	9	20	19.95486	0.265607					5		20.1570	
LSI	1	24	0.778184	0.041478		5	5.871	<0.001	5	0.7006		
	2	35	0.95771	0.044625					9	0.7764	0.7764	
	4	23	0.914671	0.048503					1	0.7782	0.7782	0.7782
	5	31	0.700648	0.026527					8		0.8895	0.8895
	8	24	0.889541	0.05313					4		0.9147	0.9147
	9	20	0.776361	0.04352					2			0.9577
GSI (male)	1	15	0.216295	0.025133		5	0.617	0.687				
	2	16	0.231329	0.028347								
	4	10	0.217474	0.053212								
	5	14	0.205043	0.034939								
	8	12	0.202752	0.028423								
	9	12	0.158735	0.017156								
GSI (female)	1	9	0.791479	0.179193		5	2.577	0.034	1	0.7915		
	2	19	1.034564	0.117358					5	0.8440	0.8440	
	4	13	0.980815	0.125408					4	0.9808	0.9808	
	5	17	0.844032	0.149622					9	0.9814	0.9814	
	8	12	1.524468	0.172859					2	1.0346	1.0346	
	9	7	0.981431	0.266239					8		1.5245	
EROD (pmol/min/mg pr) (one factor in two factor ANOVA – no sig interaction)	1	17	26.76471	2.347604		5	1.802	0.119				
	2	19	26.05263	2.544888								
	4	18	38.88889	2.62702								
	5	20	19.7	2.171708								
	8	22	29.62273	2.308257								
	9	20	24.9	2.307767								

		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
Naphthalene-type metabolites (mg/mg pr)	1	20	7354.35	498.1729		5	4.285	0.001	5	5353.9		
	2	20	7337.85	605.0693					4	5356.1		
	4	20	5356.05	388.6615					8	7015.5	7015.5	
	5	21	5353.905	584.8158					2	7337.9	7337.9	
	8	22	7015.5	413.5523					1	7354.4	7354.4	
	9	19	8071.684	732.2138					9		8071.7	
Pyrene-type metabolites (µg/mg pr)	1	20	1257.8	154.4212	log	5	3.003	0.013	4	2.9181		
	2	20	1111.7	111.4335					5	2.9422	2.9422	
	4	20	883.2	77.82601					2	3.0029	3.0029	
	5	21	995.5238	118.8884					1	3.0524	3.0524	
	8	22	1519.182	197.8415					8	3.1014	3.1014	
	9	19	1496.263	157.9369					9		3.1246	
Benzo(a) Pyrene-type metabolites (µg/mg pr) (post-hocs based on non-logged data – logged tukeys says no diff)	1	20	7902.25	1254.462	log	5	3.465	0.006	5	4948.0		
	2	20	5744.25	562.7884					4	5130.8	5130.8	
	4	20	4947.95	290.1948					2	5744.3	5744.3	
	5	21	5130.81	594.8676					8	6933.7	6933.7	
	8	22	6933.682	532.4972					9	7106.6	7106.6	
	9	19	7106.579	634.9854					1		7902.3	
SDH (mIU)	1	19	25.11036	7.254186		5	0.268	0.930				
	2	21	32.39631	7.513514								
	4	20	23.37097	4.77476								
	5	21	20.59908	4.057786								
	8	24	19.27419	2.934565								
	9	19	16.09508	3.902118								
DNA damage (ng/ml/mg pr)	1	16	1.379798	0.176707		5	2.408	0.041		Post-hocs don't identify differences. Closest is difference between sites 2 and 9		
	2	20	0.918831	0.090075								
	4	19	1.049659	0.089261								
	5	18	1.324223	0.151047								
	8	19	1.321676	0.137129								
	9	17	1.483769	0.194482								

Table A3 – 2. Summary results for red emperor captured in Phase II. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
Condition Factor	1	21	26.27	0.868		–	–	0.016	8	24.6035		
(Kruskal Wallis)	2	18	27.35	0.992					9		26.5190	
	4	24	24.42	0.592					4		26.6281	
	5	23	23.89	0.590					5		26.8341	
	8								2		27.3021	
	9								1		27.4421	
LSI	1	35	0.695416	0.025804		5	4.178	0.001	4	0.2125		
	2	27	0.692718	0.041936					2	0.2254	0.2254	
	4	38	0.639996	0.026737					1	0.2277	0.2277	
	5	20	0.751942	0.058157					5	0.2386	0.2386	
	8	16	0.841975	0.033926					9		0.2594	
	9	20	0.827585	0.044922					8		0.2642	
GSI (male)	1	15	0.061459	0.006254		–	–	0.027	2	0.0537		
(Kruskal–Wallis)	2	10	0.053705	0.003056					4	0.0575		
	4	16	0.057471	0.004225					1	0.0615		
	5	7	0.096787	0.017271					9	0.0756	0.0756	
	8	1	0.065156						5		0.0968	
	9	8	0.075572	0.005066								
GSI (female)	1	19	1.483892	0.221404		5	0.768	0.576				
	2	17	1.251865	0.132502								
	4	22	1.566428	0.465634								
	5	12	2.012885	0.409285								
	8	11	1.552402	0.163472								
	9	11	2.085371	0.301213								
EROD (pmol/min/mg pr) (results on one factor in two factor ANOVA. No sig interaction but males > females)	1	15	55.4	5.664257		5	2.141	0.067				
	2	23	76.48696	13.25446								
	4	17	84.35294	9.938761								
	5	20	37.145	1.889047								
	8	16	54.7875	3.33189								
	9	20	48.22	4.186115								

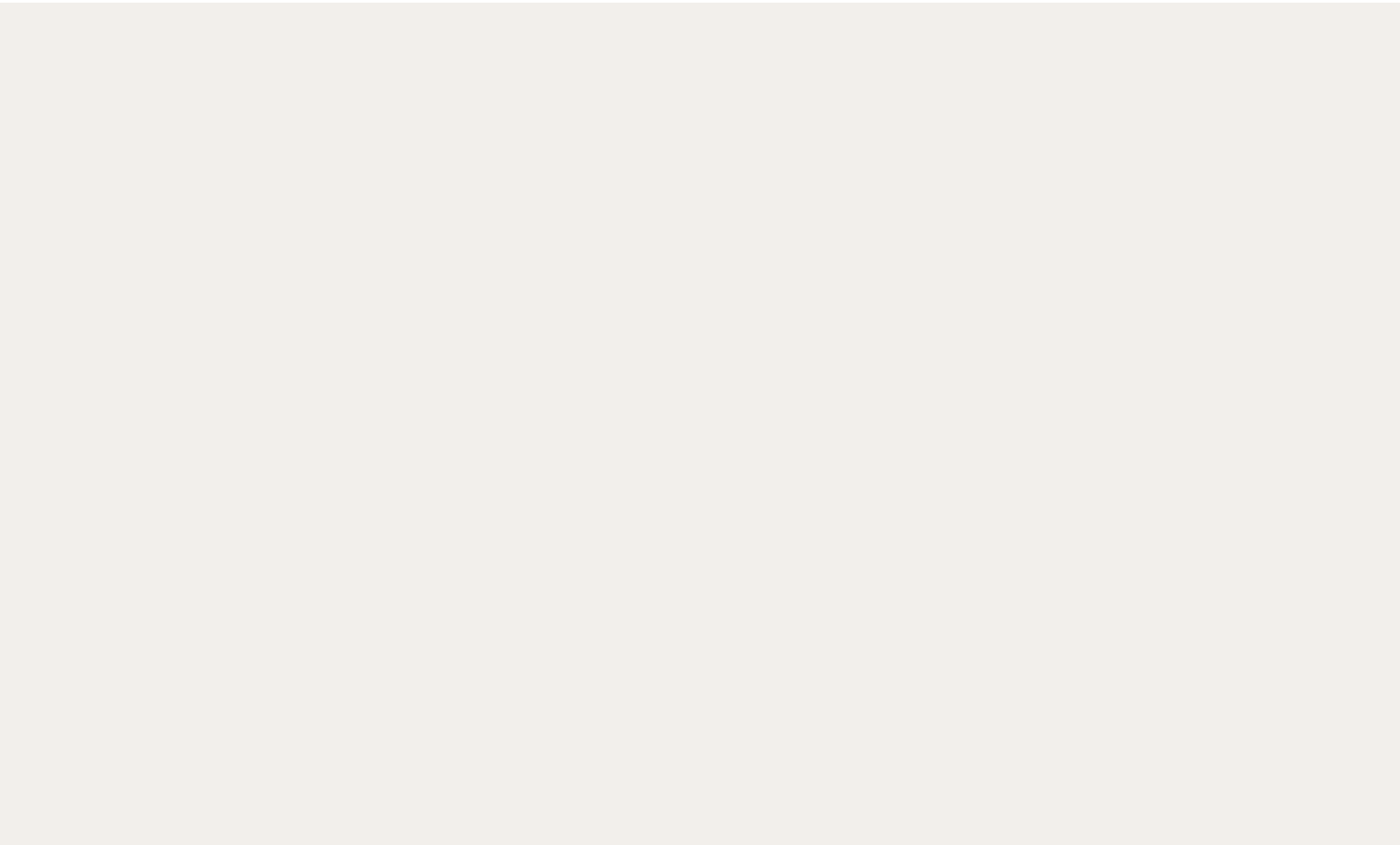
		Descriptive			Statistical Analysis							
Parameter	Site	N	Mean	SEM	Transform	ANOVA			Homogeneous subsets			
						df	F	sig	Site	1	2	3
Naphthalene-type metabolites (mg/mg pr)	1	14	6207.214	716.7889	log	5	4.887	<0.001	5	3.5560		
	2	26	7214.692	748.1767					4	3.6199	3.6199	
	4	20	4635.85	418.033					1		3.7549	
	5	17	4010.941	410.7718					8		3.7728	
	8	14	6164.643	473.8261					9		3.7771	
	9	20	6405.5	550.3534					2		3.8071	
Pyrene-type metabolites (µg/mg pr)	1	14	991.7857	145.233	log	5	3.108	0.012	5	2.7743		
	2	26	1144.885	139.7465					4	2.9315	2.9315	
	4	20	1041.75	157.4607					1	2.9344	2.9344	
	5	17	709.8824	84.61022					2	2.9738	2.9738	
	8	14	1256.643	125.8971					8		3.0749	
	9	20	1661.7	366.2651					9		3.0976	
Benzo(a) Pyrene-type metabolites (µg/mg pr)	1	14	7667.071	2333.879	log	5	3.831	0.003	4	3.5095		
	2	26	5466.808	601.0923					5	3.5221		
	4	20	4004.2	508.6545					2	3.6756	3.6756	
	5	17	3618.118	286.648					1	3.7301	3.7301	
	8	14	5894.214	507.4154					8	3.7515	3.7515	
	9	20	7016.45	951.3941					9		3.7846	
SDH (mIU) (Kruskal Wallis)	1	15	9.354839	1.024452		-	-	0.122				
	2	28	10.95622	1.198803								
	4	19	16.19694	3.126521								
	5	20	11.08065	1.072761								
	8	16	13.36694	2.690048								
	9	20	16.40323	1.672461								
DNA damage (ng/ml/mg pr)	1	14	2.270221	0.416306		5	0.487	0.785				
	2	26	2.555274	0.242771								
	4	19	2.740561	0.343252								
	5	17	2.274125	0.390754								
		13	2.484355	0.348725								
		16	3.042245	0.684518								

Table A3 – 3. Summary results for rainbow runner captured in Phase II. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	t-test			Homogeneous subsets		
						df	t	sig	Site	1	2
Condition Factor	4	3	10.48195	0.087904		4	3.869	0.018			
	7	3	9.662269	0.192753							
LSI*	4	3	0.63663	0.061385		4	-1.574	0.191			
	7	3	0.796326	0.08081							
GSI (male)	4	1	2.901235			-	-	-			
	7	0									
GSI (female)**	4	2	1.879413	0.730477		2.235	-1.361	0.294			
	7	3	3.153316	0.585139							
EROD (pmol/min/mg pr)	4	3	8.766667	1.039765		1	3.21	0.171			
Naphthalene-type metabolites (mg/mg pr) (t-test)	4	2	1569.5	326.5	log	3	-1.162	0.912			
	7	3	1515	134.0497							
Pyrene-type metabolites (µg/mg pr) (t-test)	4	2	307.5	42.5	log	3	0.12	0.962			
	7	3	385.3333	177.9928							
Benzo(a) Pyrene-type metabolites (µg/mg pr) (t-test)	4	2	1766	378	log	3	-0.052	0.273			
	7	3	780.6667	463.1761							
SDH (mIU) (t-test)	4	2	14.03226	6.290323		3	0.994	0.393			
	7	3	6.774194	0.558726							
DNA damage (ng/ml/mg pr) (t-test)	4	2	0.539965	0.077826		3	-0.667	0.552			
	7	3	1.220983	0.789299							

Table A3 – 4. Summary results for Spanish mackerel captured in Phase II. Homogeneous subsets defined by Tukeys Post-hoc tests

		Descriptive			Statistical Analysis						
Parameter	Site	N	Mean	SEM	Transform	t-test			Homogeneous subsets		
						df	t	sig	Site	1	2
Condition Factor	4	11	7.678314	0.341105		13	0.907	0.381			
(t-test)	6	4	7.117739	0.382082							
LSI	4	11	0.965583	0.306155		13	0.697	0.498			
(t-test)	6	4	0.603121	0.021965							
GSI (male)	4	2	0.176927	0.129998		-	-	-			
	6	0									
GSI (female)	4	9	0.87212	0.276765		11	-1.298	0.221			
(t-test)	6	4	1.65519	0.681011							
EROD (pmol/min/mg pr) (2way – no int or sex diff)	4	11	17.48182	1.208742		1	3.681	0.081			
	6	4	13.2	1.400595							
Naphthalene-type metab. (mg/mg pr)	4	12	13885.17	1771.186	log	15	0.952	0.356			
	6	4	7911.5	1405.847							
Pyrene-type metab. (µg/mg pr)	4	12	2212.833	346.676	log	15	0.731	0.476			
	6	4	1148.75	186.0463							
Benzo(a) Pyrene-type metab. (µg/mg pr)	4	12	2473.833	407.9336	log	15	1.808	0.091			
	6	4	2270.25	740.5843							
SDH (mIU)	4	13	6.104218	0.851482		15	0.806	0.433			
	6	4	6.048387	0.463271							
DNA damage (ng/ml/mg pr)	4	12	0.72665	0.091157		14	-1.525	0.150			
	6	4	1.038838	0.235138							



Appendix D – Results of Floating Material Analysis



**LEEDER
CONSULTING**

A.B.N. 540 864 910 09
4 - 5, 18 Redland Drive
Mitcham, Vic, 3132
Telephone: (03) 9874 1988
Fax: (03) 9874 1933

Chartered Chemists

14-Jan-2011

REPORT NUMBER: M101702A

Site/Client Ref: West Atlas

PTTEP Australasia

Level 1

162 Colins Street

West Perth

Western Australia 6005

Attention: Eleanor Stoney

CERTIFICATE OF ANALYSIS

SAMPLES: Three samples were received for analysis

DATE RECEIVED: **24-Nov-2010**

DATE COMMENCED: **23-Dec-2010**

METHODS: See Attached Results

RESULTS: Please refer to attached pages for results.

Note: Results are based on samples as received at Leeder Consulting's laboratories

Note: No petroleum hydrocarbons were detected in any of the samples.

REPORTED BY:

Adam Atkinson

Laboratory Manager



NATA Accredited Laboratory Number: 14429

**This Document is issued in accordance with
NATA's accreditation requirements**

**Accredited for compliance
with ISO/IEC 17025.**



(I) RESULTS

Report N°: M101702A

Matrix: Sheen

Method: MA-28 Product Identification

Analyte Name	Leeder ID	2010021817	2010021818	2010021819
	Client ID	Site 4 Heywood Shoal	Site 1-1	Site 1-2
	PQL			
Product Identification		nd	Some fatty acids	Some fatty acids

Matrix: Sheen

Method: MA-28 Product Identification

Analyte Name	Leeder ID	2010021821
	Client ID	Method
	PQL	Blank
Product Identification		nd



QUALIFIERS / NOTES FOR REPORTED RESULTS

<i>is</i>	Insufficient Sample to perform this analysis.
T	Tentative identification based on computer library search of mass spectra.
ND	Not Detected – The analyte was not detected above the reported PQL.
<i>nr</i>	Not Requested for analysis.
R	Rejected Result – results for this analysis failed QC checks.
SQ	Semi-Quantitative result – quantitation based on a generic response factor for this class of analyte.
IM	Inappropriate method of analysis for this compound
U	Unable to provide Quality Control data – high levels of compounds in sample interfered with analysis of QC results.
UF	Unable to provide Quality Control data- Surrogates failed QCchecks due to sample matrix effects
UI	Unable to provide Quality Control data – insufficient sample to perform QC checks.
B	This analyte also detected in analysis of the Method Blank.
D	Deviation from standard method – see notes for specific explanation.
L	Analyte detected at a level above the linear response of calibration curve.
NT	No blank sorbent tubes provided for QC analysis.
C1	These compounds co-elute.
C2	These compounds co-elute.



**LEEDER
CONSULTING**

APPENDIX ONE.

CHAIN OF CUSTODY DOCUMENT

From: Stoney, Eleanor [mailto:Eleanor.Stoney@au.pttep.com]
Sent: Wednesday, 22 December 2010 9:29 PM
To: Leif Cooper
Subject: RE: Sediment testing

Hi Leif

Please sample the sheen nets for identification.

If the water and sediment samples are out of their holding time then please dispose.

Regards,

Eleanor Stoney
Environmental Coordinator

Direct: (08) 9320 9514

PTTEP Australasia

Level 1
162 Colins Street
West Perth WA 6005
Tel: +61 (0)8 9483 9483 Fax: +61 (0)8 9483 9484



Leif
30/12/10

Chain of Custody Document

Incident No: WEST ATLAS

Sampled By: Leif Cooper

Comments:

Sample Description					Date	
Site 8	11° 53' 26 S	125° 34' 45 E	(+dup)		11/11/10	11:35
Site 9			(+dup)		12/11/10	2:00
S1	12° 43' 90 S	124° 34' 04 E	72m	(+dup)	13/11/10	9:20
S2	12° 44' 10 S	124° 32' 90 E	85m			9:53
S3	12° 44' 12 S	124° 31' 80 E	84m			10:11
S4	12° 43' 90 S	124° 30' 65 E	85m			10:30
S5	12° 43' 08 S	124° 29' 59 E	85m			10:49
S6	12° 43' 10 S	124° 30' 67 E	85m			11:10
S7	12° 43' 09 S	124° 31' 81 E	84m			11:32
S8	12° 43' 14 S	124° 32' 86 E	84m			11:52
S9	12° 43' 09 S	124° 33' 98 E	83m			12:12
S10	12° 43' 09 S	124° 35' 15 E	75m			12:31
S11	12° 41' 09 S	124° 36' 10 E	67m			12:50
S12	12° 42' 03 S	124° 36' 06 E	80m	- no dup		2:06
S13	12° 42' 07 S	124° 34' 03 E	83m	(+dup)		2:35
S14	12° 42' 09 S	124° 32' 94 E	83m			2:55
S15	12° 42' 14 S	124° 31' 92 E	84m			3:15
S16	12° 42' 03 S	124° 30' 77 E	84m			3:37
S17	12° 42' 02 S	124° 29' 64 E	86m			3:58
S18	12° 41' 93 S	124° 28' 75 E	86m			4:16

Name <u>Leif</u>	Signature <u>[Signature]</u>	Date <u>21/11/10</u>
Organisation		Time



Chain of Custody Document

Incident No: WEST ATLAS

Sampled By: Leif Cooper

Comments: _____

Sample Description					Date
S19	12° 40' 86 S	124° 28' 67 E	83 m	(+ dup)	13/11/10 4:35
S20	12° 40' 78 S	124° 29' 67 E	84 m		4:50
S21	12° 40' 84 S	124° 30' 78 E	85 m		5:06
S22	12° 40' 90 S	124° 31' 84 E	82 m		14/11/10 8:19
S23	12° 40' 87 S	124° 32' 90 E	75 m		8:39
S24	12° 40' 84 S	124° 33' 03 E	75 m		8:58
S25	12° 40' 85 S	124° 35' 10 E	63 m	- no dup	9:13
S26	12° 40' 84 S	124° 36' 20 E	59 m	(+ dup)	9:42
S27	12° 39' 66 S	124° 36' 26 E	73 m		10:14
S28	12° 39' 75 S	124° 35' 10 E	78 m		10:30
S29	12° 39' 74 S	124° 33' 97 E	70 m		10:59
S30	12° 39' 81 S	124° 32' 87 E	73 m		11:17
S31	12° 39' 88 S	124° 31' 78 E	81 m		11:45
S32	12° 39' 91 S	124° 30' 73 E	83 m		12:07
S33	12° 39' 82 S	124° 29' 61 E	82 m		12:24
S34	12° 39' 83 S	124° 28' 54 E	76 m		2:03
S35	12° 38' 71 S	124° 28' 66 E	75 m		2:23
S36	12° 38' 68 S	124° 29' 65 E	78 m		2:41
S37	12° 38' 71 S	124° 30' 76 E	84 m		2:59
S38	12° 38' 75 S	124° 31' 90 E	80 m		3:20

Name <u>Leif</u>	Signature <u>[Signature]</u>	Date <u>21/11/10</u>
Organisation		Time



Chain of Custody Document

Incident No: West Area

Sampled By: Lif Cooper

Comments:

Sample Description					Date	
S39	12° 38' 78 S	124° 32' 93 E	74m	(+dup)	14/11/10	3:35
S40	12° 38' 76 S	124° 34' 01	71m			3:51
S41	12° 38' 75 S	124° 35' 19	76m			4:08
S42	12° 38' 72 S	124° 36' 14	81m			4:21
S43	12° 37' 61 S	124° 35' 18	68m			4:46
S44	12° 37' 61 S	124° 34' 07	74m			5:10
S45	12° 37' 66 S	124° 32' 99	75m			5:28
S46	12° 37' 61 S	124° 31' 93	74m			5:45
S47	12° 37' 54 S	124° 30' 75	81m		15/11/10	8:58
S48	12° 37' 58 S	124° 29' 67	77m			9:21
S49	12° 36' 66 S	124° 30' 82	79m			9:39
S50	12° 36' 60 S	124° 31' 92	83m			9:59
S51	12° 36' 56 S	124° 32' 91	76m			10:15
S52	12° 36' 63 S	124° 33' 92	76m	↓	↓	10:31
Site 1	12° 52' 23 S	124° 40' 20 E			16/11	
Site 4	Shore sample (net)			13° 24' 33 S 124° 09' 90 E	18/11	11:30
Site 4				13° 24' 40 S 124° 08' 94 E	18/11	1:30

Name <u>Lif</u>	Signature <u>[Signature]</u>	Date <u>21/11/10</u>
Organisation		Time

Form-054



Issue Date 1-10-02

From: Chris Rawson [mailto:C.Rawson@curtin.edu.au]
Sent: Thursday, 2 December 2010 5:07 PM
To: Leif Cooper
Subject: RE: Equipment delivery

Hi Leif,

The sea-legs are still sith me (i.e., the land legs are not!), Hopefully I will come good in a day or so.

We picked up the net samples quite close to the rig but I strongly suspect they are algae. One of the Grant's other boats was in the area and reported seeing some brown substance on the surface so we picked it up.

I also should have mentioned that we could not get a sediment sample at Scott Reef (our site 7). The structure of the reef is such that it was very rough bottom and the dredge would come up empty, with a few stones, or with a bit of coral. Further off the reef the water depth drops dramatically to over 400m.

We will send the remaining nets with some fish samples, assuming they arrive here at some point.

Cheers
Chris

Dr Christopher Rawson
Post-Doctoral Fellow
Department of Environment and Agriculture
Faculty of Science and Engineering

Curtin University
Tel | +61 8 9266 2033
Fax | +61 8 9266 2495
Mobile | +61 415 291 959

Email | c.rawson@curtin.edu.au



Curtin University is a trademark of Curtin University of Technology.
CRICOS Provider Code 00301J (WA), 02637B (NSW)

From: Leif Cooper [mailto:Leif@leederconsulting.com]
Sent: Thursday, 2 December 2010 2:02 PM
To: Chris Rawson
Subject: RE: Equipment delivery

Hi Chris,

I can confirm that we received all our packages in Melbourne. Very interested to see two sheen/net samples there – what do you suspect is in them?

Hope your sea-legs have come good.

Regards,

Leif Cooper
Senior Chemist

Leeder Consulting Pty Ltd
5/18 Redland Dve, Mitcham Vic. 3123
T: +61 3 9874 1988 | M: 0414 993 888 | F: +61 3 9874 1933 | www.leederconsulting.com

You are reminded that electronic communication involves unavoidable risks. Leeder Consulting does not warrant or represent that this communication is confidential or that it is free of any computer virus.

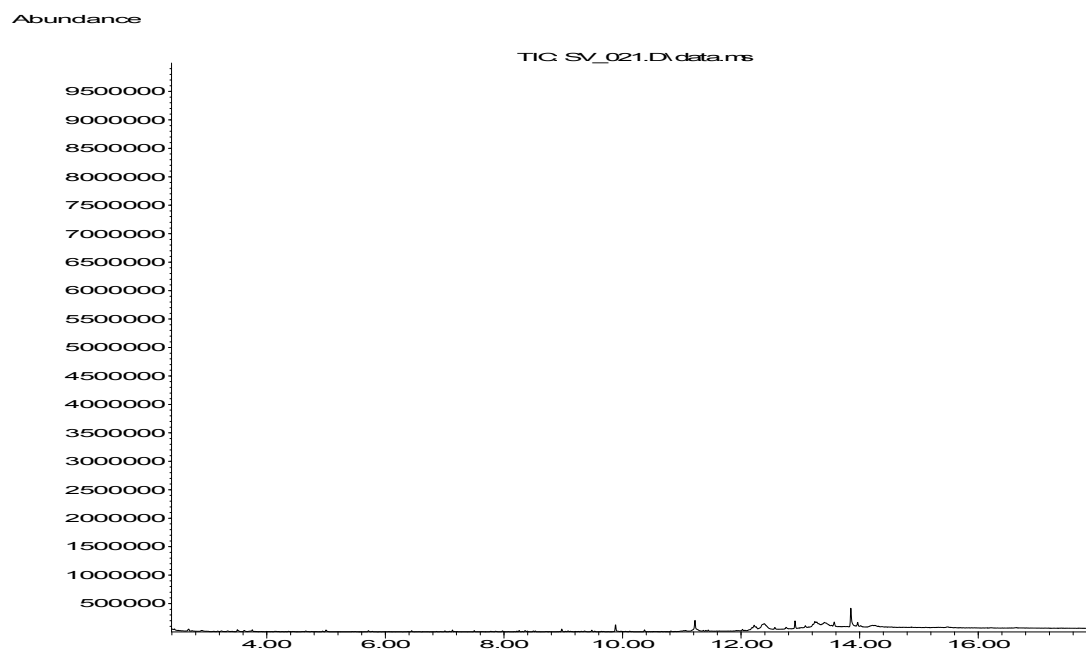
This message is intended only for the use of the individual or entity to whom it is addressed and may contain information that is privileged and/or confidential. Any unauthorised use, disclosure, copying or distribution of this message or information is prohibited. If you have received this message in error, please telephone me on +61 3 9874 1988 or contact me by return email immediately.



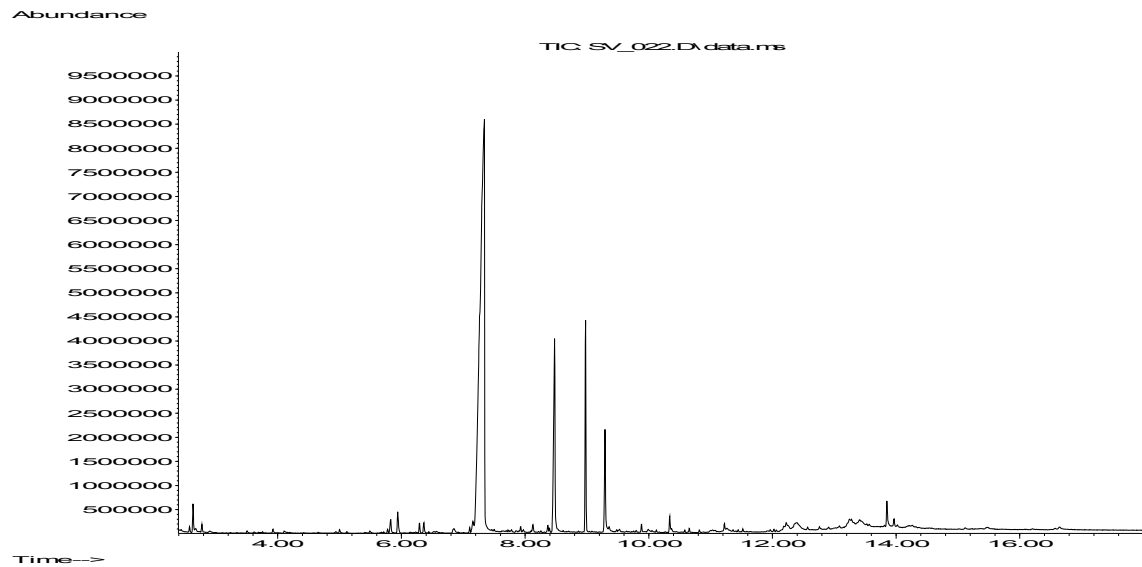


APPENDIX TWO

TOTAL ION CHROMATOGRAMS

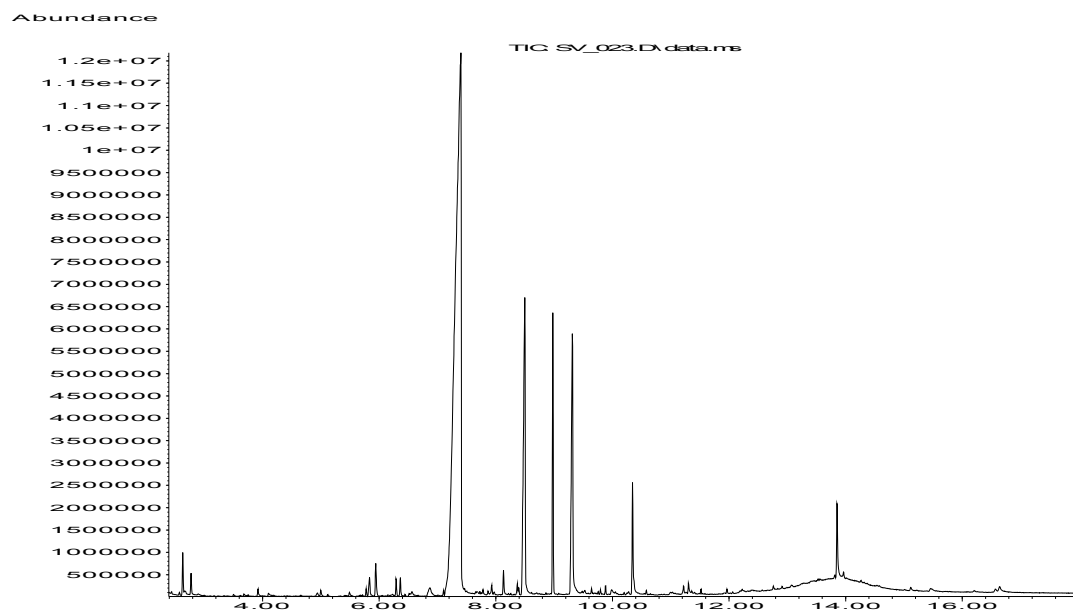


Site 4 – Heywood Shoal
Leeder ID: 2010021817



Site 1-1

Leeder ID: 2010021818



Site 1-2

Leeder ID: 2010021819

Appendix E – Results of Water Samples Analysis

(Phase I and Phase II)



LEEDER CONSULTING

Chartered Chemists & Toxicologists

4th December 2009

Australian Maritime Safety Authority
GPO Box 2181
Canberra, ACT 2601
Attn: Paul Nelson

A.B.N. 54 086 491 009
4-5, 18 Redland Drive
Mitcham, Victoria
AUSTRALIA, 3132
Telephone +61 3 9874 1988
Fax +61 3 9874 1933

REPORT No: M091766
Site/Client Ref: West Atlas

CERTIFICATE OF ANALYSIS

SAMPLES: Fifty-two samples were received for analysis

DATE RECEIVED: 20-Nov-2009

DATE COMMENCED: 20-Nov-2009

METHODS: See Attached Results

RESULTS: Please refer to attached pages for results

Note: Results are based on samples as received at Leeder Consulting's Laboratories

Report By:

Leif Cooper
Senior Chemist



This report has been prepared in accordance with the quality system of Leeder Consulting Pty. Ltd and may not be reproduced except in full.



LEEDER CONSULTING

(I) RESULTS

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2009023660	2009023662	2009023664	2009023666	2009023668	2009023671	2009023672
		Client ID	Site 1 0A 16/11/2009	Site 1 1A 16/11/2009	Site 1 3A 16/11/2009	Site 1 5A 16/11/2009	Site 1 10A 16/11/2009	Site 1 15B 16/11/2009	Site 2 0A 9/11/2009
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2009023674	2009023676	2009023678	2009023680	2009023682	2009023683	2009023984
		Client ID	Site 2 1A 9/11/2009	Site 2 3A 9/11/2009	Site 2 5A 9/11/2009	Site 2 10A 9/11/2009	Site 2 15B 9/11/2009	Site 2 20A 9/11/2009	Site 2-R 0A 9/11/2009
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd



LEEDER CONSULTING

(I) RESULTS

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2009023686	2009023688	2009023690	2009023692	2009023694	2009023696	2009023698
		Client ID	Site 2-R 1A	Site 2-R 3A 19/11/2009	Site 2-R 5A 19/11/2009	Site 2-R 10A 19/11/2009	Site 2-R 15A 19/11/2009	Site 3 0A 11/11/2009	Site 3 1A 11/11/2009
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2009023698	2009023700	2009023702	2009023704	2009023706	2009023708	2009023710
		Client ID	Site 3 1A 11/11/2009	Site 3 3A 11/11/2009	Site 3 5A 11/11/2009	Site 3 10A 11/11/2009	Site 3 15A 11/11/2009	Site 4 0A 12/11/2009	Site 4 1A 12/11/2009
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd



LEEDER CONSULTING

(I) RESULTS

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2009023712	2009023714	2009023716	2009023718	2009023720	2009023722	2009023724
		Client ID	Site 4 3A 12/11/2009	Site 4 5A 12/11/2009	Site 4 10A 12/11/2009	Site 4 15A 12/11/2009	Site 4.1 0A 12/11/2009	Site 4.1 1A 12/11/2009	Site 4.1 3A 12/11/2009
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2009023726	2009023728	2009023730	2009023732	2009023734	2009023736	2009023738
		Client ID	Site 4.1 5A 12/11/2009	Site 4.1 10A 12/11/2009	Site 4.1 15A 12/11/2009	Site 5 0A 14/11/2009	Site 5 1A 14/11/2009	Site 5 3A 14/11/2009	Site 5 5A 14/11/2009
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd



LEEDER CONSULTING

(I) RESULTS

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2009023740	2009023742	2009023744	2009023746	2009023748	2009023750	2009023752
		Client ID	Site 5 10A 14/11/2009	Site 5 15A 14/11/2009	Site 6 0A 15/11/2009	Site 6 1A 15/11/2009	Site 6 3A 15/11/2009	Site 6 5A 15/11/2009	Site 6 10A 15/11/2009
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2009023754	2009023766	2009023767	2009023768
		Client ID	Site 6 15A 15/11/2009	Method	Method	Method
		PQL		Blank	Blank	Blank
C6-C9	0.01		nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd



LEEDER CONSULTING

(I) RESULTS

Matrix Wax

Method: MA-28 Product Identification

Analyte Name	Leeder ID	2009023771	2009023772
	Client ID	Site 4.1 Wax Residue	Brown Floating Residue
	PQL		
Product Identification		Waxy Crude	Waxy Crude
Weathering loss (%w/w)		88%	90%



LEEDER CONSULTING

(II) QUALITY CONTROL

Matrix Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leader ID	2009023777	2009023778	2009023779	2009023780	2009023781	2009023782
		Client ID	Method	Method	Method	Method	Method	Method
		PQL	Spike	Spike Dup	Spike	Spike Dup	Spike	Spike Dup
Total C6-C36			97	98	97	104	119	103



**LEEDER
CONSULTING**

Report No: M091766

APPENDIX ONE

CHAIN OF CUSTODY



LEEDER CONSULTING

Lyndall Stevens

From: Stoney, Eleanor [Eleanor.Stoney@au.pttep.com]
Sent: Thursday, 19 November 2009 12:26 PM
To: Leif Cooper
Cc: Sample Reception; John Wardrop
Subject: FW: URGENT FREIGHT TO BE ORGANISED

Importance: High

Follow Up Flag: Follow up
Flag Status: Flagged

Categories: Samples Arriving

Leif

Please see below.

To clarify, there are two sets of samples being sent to Leeder:

- o Set 1 – from Curtin University (in Darwin) containing water samples. These samples will be delivered to the lab by TNT most likely on Friday (20/11) afternoon.
- o Set 2 – from Uni Queensland (in Broome) containing water and sediment samples. These samples will be delivered to Melbourne Airport by AAE. I will arrange for TNT to collect the samples and deliver to the lab on Friday (20/11) afternoon.

Our contracts person, Jeff Hunneybun, is organising a contract to be drafted and will send this through to you regarding the handling and payment of samples. This will be based on the quote you send through earlier.

Please give me a call if you wish to discuss.

Kind Regards,

Eleanor Stoney
HSE Coordinator

Direct: (08) 9320 9514

PTTEP Australasia
Level 1
162 Collins Street
West Perth WA 6005
Tel: +61 (0)8 9483 9483 Fax: +61 (0)8 9483 9484



From: Stoney, Eleanor
Sent: Thursday, 19 November 2009 8:19 AM
To: Mooney, Christine
Subject: FW: URGENT FREIGHT TO BE ORGANISED
Importance: High

Hi Chrissy

Sorry, I didn't specify in my email below, but the biological and equipment should be sent to Perth on overnight express or first class, whichever one guarantees to get to Perth by tomorrow (20/11) afternoon.

Kind Regards,

Eleanor Stoney
HSE Coordinator

Direct: (08) 9320 9514



LEEDER CONSULTING

PTTEP Australasia

Level 1
162 Colins Street
West Perth WA 6005
Tel: +61 (0)8 9483 9483 Fax: +61 (0)8 9483 9484

From: Stoney, Eleanor
Sent: Wednesday, 18 November 2009 7:42 PM
To: Mooney, Christine; Penny, Charlie
Cc: 'Ross, Barbara'
Subject: URGENT FREIGHT TO BE ORGANISED
Importance: High

Chrissy / Charlie

Here are the details, as discussed:

Monique Gagnon (Curtin Uni) – 0401 103 312

Monique and Christopher will arrive in DWN wharf (@ duckpond I believe) tomorrow (19/11) morning at approximately 0800. Could we please organise someone to collect them from the wharf? They will have a lot of equipment, approximately 16 containers. Both her and Chris are on the 'Megan M'.

There will be water samples that need to be sent to Leeder Consulting in Melbourne.

The address is:

Attention: Lelf Cooper
Leeder Consulting
Unit 5, 18 Redland Drive
Mitcham Victoria Australia 3132

There will be equipment and biological samples that need to be sent to Perth. The address is:

Curtin University, Bentley Campus,
Kent Street
Building 311, room 111
(Bldg 311 is close from the Curtin Uni main bus stop)

You may need to ask Monique who she wishes the samples to be sent to.

Monique should provide a list of the box contents, weight and dimensions. There will be some (???) liquid nitrogen dry shippers. If TNT gives you any issues, say that we confirm that they are NOT DG and are designed for domestic travel.

Please give me a call if you want to discuss.

Kind Regards,

Eleanor Stoney
HSE Coordinator

Direct: (08) 9320 9514

PTTEP Australasia

Level 1
162 Colins Street
West Perth WA 6005
Tel: +61 (0)8 9483 9483 Fax: +61 (0)8 9483 9484

DISCLAIMER

This message contains confidential information and is intended only for the individual named. If you are not the named addressee you should not disseminate, distribute or copy this e-mail. Please notify the sender immediately by e-mail if you have received this e-mail by mistake and delete this e-mail from your system. E-mail transmission cannot be guaranteed to be secure or error-free as information could be intercepted, corrupted, lost, destroyed, arrive late or incomplete,





LEEDER CONSULTING

Report No: M091763

Erynne McCrorey

From: Leif Cooper
Sent: Tuesday, 24 November 2009 10:16 AM
To: Lyndall Stevens; Erynne McCrorey
Subject: FW: Analysis

Lyndall/Erynne,

Confirmed analysis for the PTTEP jobs.

'Curtin Uni' refers to M091766
'Browse Island' is already done (M091762)
'Uni of Qld' refers to M091763

Regards,

Leif

From: John Wardrop [mailto:wardropcons@iprimus.com.au]
Sent: Tuesday, 24 November 2009 10:12 AM
To: Leif Cooper
Subject: Analysis

Leif,

Attached Master Sample List Updated.

Please do:

- Curtin Uni Oil/Wax (Red): ID and % loss
- Curtin Uni Water (Red): All non duplicates for TPH (and PAH if positive) (careful, for a couple of locations there is no A sample)
- Browse island Survey oils/sheen (Blue) ID and % loss
- Univ Qld Oils (Green) ID and % loss
- Uni Qld Waters (Green): TPH (and PAH if positive)
- Uni of Qld Biota (Green): TPH on whole sample (BTEX and PAH id positive).
- Uni of Qld Seds (Green): TPH

Please also

- Check List for completeness
- Fill out sample Quality column and
- Return

Regards

Wardrop[





**LEEDER
CONSULTING**

A.B.N. 540 864 910 09
4 - 5, 18 Redland Drive
Mitcham, Vic, 3132
Telephone: (03) 9874 1988
Fax: (03) 9874 1933

Chartered Chemists

25-May-2010

REPORT NUMBER: M100344R1

Site/Client Ref: Montara S4A Phase II

PTTEP Australasia

Level 1

162 Colins Street

West Perth

Western Australia 6005

Attention: Eleanor Stoney

CERTIFICATE OF ANALYSIS

This report replaces previous report dated 29-Mar-2010

SAMPLES: Thirty-four samples were received for analysis

DATE RECEIVED: **19-Mar-2010**

DATE COMMENCED: **22-Mar-2010**

METHODS: See Attached Results

RESULTS: Please refer to attached pages for results.

Note: Results are based on samples as received at Leeder Consulting's laboratories

REPORTED BY:

Adam Atkinson

Laboratory Manager



NATA Accredited Laboratory Number: 14429

**This Document is issued in accordance with
NATA's accreditation requirements**

**Accredited for compliance
with ISO/IEC 17025.**



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-10.WW.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004044	2010004045	2010004046	2010004047	2010004048	2010004049	2010004050
		Client ID	Site 1-0A	Site 1-1A	Site 1-3B	Site 1-5A	Site 1-10B	Site 1-15A	Site 2-0A
Benzene	0.001		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.001		nd	nd	nd	nd	nd	nd	nd
Toluene	0.001		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.001		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.001		nd	nd	nd	nd	nd	nd	nd

Matrix: Water

Method: MA-10.WW.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004051	2010004052	2010004053	2010004054	2010004055	2010004056	2010004057
		Client ID	Site 2-3A	Site 2-5A	Site 2-10A	Site 4-0A	Site 4-1A	Site 4-3A	Site 4-5A
Benzene	0.001		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.001		nd	nd	nd	nd	nd	nd	nd
Toluene	0.001		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.001		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.001		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-10.WW.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004058	2010004059	2010004060	2010004061	2010004062	2010004063	2010004064
		Client ID	Site 4-10A	Site 4-15A	Site 5-0A	Site 5-1B	Site 5-3A	Site 5-5B	Site 5-10A
		PQL							
Benzene	0.001		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.001		nd	nd	nd	nd	nd	nd	nd
Toluene	0.001		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.001		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.001		nd	nd	nd	nd	nd	nd	nd

Matrix: Water

Method: MA-10.WW.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004065	2010004066	2010004067	2010004068	2010004069	2010004070	2010004071
		Client ID	Site 5-15A	Site 6-0B	Site 6-1B	Site 6-3A	Site 6-5B	Site 6-10A	Site 6-15A
		PQL							
Benzene	0.001		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.001		nd	nd	nd	nd	nd	nd	nd
Toluene	0.001		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.001		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.001		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-10.WW.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004072	2010004073	2010004074	2010004075	2010004076	2010004077
		Client ID	Site 7-0A	Site 7-1A	Site 7-3A	Site 7-5A	Site 7-10A	Site 7-15A
Benzene	0.001		nd	nd	nd	nd	nd	nd
Ethylbenzene	0.001		nd	nd	nd	nd	nd	nd
Toluene	0.001		nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.001		nd	nd	nd	nd	nd	nd
o-Xylene	0.001		nd	nd	nd	nd	nd	nd

Matrix: Water

Method: MA-10.WW.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004124	2010004125	2010004126
		Client ID	Method	Method	Method
			Blank	Blank	Blank
Benzene	0.001		nd	nd	nd
Ethylbenzene	0.001		nd	nd	nd
Toluene	0.001		nd	nd	nd
m&p-Xylenes	0.001		nd	nd	nd
o-Xylene	0.001		nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004044	2010004045	2010004046	2010004047	2010004048	2010004049	2010004050
		Client ID	Site 1-0A	Site 1-1A	Site 1-3B	Site 1-5A	Site 1-10B	Site 1-15A	Site 2-0A
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd

Matrix: Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004051	2010004052	2010004053	2010004054	2010004055	2010004056	2010004057
		Client ID	Site 2-3A	Site 2-5A	Site 2-10A	Site 4-0A	Site 4-1A	Site 4-3A	Site 4-5A
		PQL							
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	0.39	0.13	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	0.39	0.13	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004058	2010004059	2010004060	2010004061	2010004062	2010004063	2010004064
		Client ID	Site 4-10A	Site 4-15A	Site 5-0A	Site 5-1B	Site 5-3A	Site 5-5B	Site 5-10A
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd

Matrix: Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004065	2010004066	2010004067	2010004068	2010004069	2010004070	2010004071
		Client ID	Site 5-15A	Site 6-0B	Site 6-1B	Site 6-3A	Site 6-5B	Site 6-10A	Site 6-15A
C6-C9	0.01		nd	nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004072	2010004073	2010004074	2010004075	2010004076	2010004077
		Client ID	Site 7-0A	Site 7-1A	Site 7-3A	Site 7-5A	Site 7-10A	Site 7-15A
		PQL						
C6-C9	0.01		nd	nd	nd	nd	nd	nd
C10-C14	0.01		nd	nd	nd	nd	nd	nd
C15-C28	0.05		nd	nd	nd	nd	nd	nd
C29-C36	0.05		nd	nd	nd	nd	nd	nd
Total C6-C36	0.05		nd	nd	nd	nd	nd	nd

Matrix: Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004124	2010004125
		Client ID	Method	Method
		PQL	Blank	Blank
C6-C9	0.01		nd	nd
C10-C14	0.01		nd	nd
C15-C28	0.05		nd	nd
C29-C36	0.05		nd	nd
Total C6-C36	0.05		nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-72.WW.01 Polynuclear Aromatic Hydrocarbons

Sample units are expressed in mg/L

		Leeder ID	2010004044	2010004045	2010004046	2010004047	2010004048	2010004049	2010004050
		Client ID	Site 1-0A	Site 1-1A	Site 1-3B	Site 1-5A	Site 1-10B	Site 1-15A	Site 2-0A
Analyte Name	PQL								
Acenaphthene	0.001		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.001		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.001		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.001		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.001		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.001		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.001		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.001		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.001		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-72.WW.01 Polynuclear Aromatic Hydrocarbons

Sample units are expressed in mg/L

		Leeder ID	2010004051	2010004052	2010004053	2010004054	2010004055	2010004056	2010004057
		Client ID	Site 2-3A	Site 2-5A	Site 2-10A	Site 4-0A	Site 4-1A	Site 4-3A	Site 4-5A
Analyte Name	PQL								
Acenaphthene	0.001		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.001		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.001		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.001		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.001		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.001		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.001		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.001		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.001		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-72.WW.01 Polynuclear Aromatic Hydrocarbons

Sample units are expressed in mg/L

		Leeder ID	2010004058	2010004059	2010004060	2010004061	2010004062	2010004063	2010004064
		Client ID	Site 4-10A	Site 4-15A	Site 5-0A	Site 5-1B	Site 5-3A	Site 5-5B	Site 5-10A
Analyte Name	PQL								
Acenaphthene	0.001		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.001		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.001		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.001		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.001		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.001		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.001		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.001		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.001		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-72.WW.01 Polynuclear Aromatic Hydrocarbons

Sample units are expressed in mg/L

		Leeder ID	2010004065	2010004066	2010004067	2010004068	2010004069	2010004070	2010004071
		Client ID	Site 5-15A	Site 6-0B	Site 6-1B	Site 6-3A	Site 6-5B	Site 6-10A	Site 6-15A
Analyte Name	PQL								
Acenaphthene	0.001		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.001		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.001		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.001		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.001		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.001		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.001		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.001		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.001		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.001		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.001		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.001		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-72.WW.01 Polynuclear Aromatic Hydrocarbons

Sample units are expressed in mg/L

		Leeder ID	2010004072	2010004073	2010004074	2010004075	2010004076	2010004077
		Client ID	Site 7-0A	Site 7-1A	Site 7-3A	Site 7-5A	Site 7-10A	Site 7-15A
Analyte Name	PQL							
Acenaphthene	0.001		nd	nd	nd	nd	nd	nd
Acenaphthylene	0.001		nd	nd	nd	nd	nd	nd
Anthracene	0.001		nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.001		nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.001		nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.001		nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.001		nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.001		nd	nd	nd	nd	nd	nd
Chrysene	0.001		nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.001		nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.001		nd	nd	nd	nd	nd	nd
Fluoranthene	0.001		nd	nd	nd	nd	nd	nd
Fluorene	0.001		nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.001		nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.001		nd	nd	nd	nd	nd	nd
Naphthalene	0.001		nd	nd	nd	nd	nd	nd
Phenanthrene	0.001		nd	nd	nd	nd	nd	nd
Pyrene	0.001		nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: MA-72.WW.01 Polynuclear Aromatic Hydrocarbons

Sample units are expressed in mg/L

Analyte Name	PQL	Leeder ID	2010004124	2010004125
		Client ID	Method	Method
			Blank	Blank
Acenaphthene	0.001		nd	nd
Acenaphthylene	0.001		nd	nd
Anthracene	0.001		nd	nd
Benzo(a)anthracene	0.001		nd	nd
Benzo (a) pyrene	0.001		nd	nd
Benzo (b) fluoranthene	0.001		nd	nd
Benzo (ghi) perylene	0.001		nd	nd
Benzo (k) fluoranthene	0.001		nd	nd
Chrysene	0.001		nd	nd
Dibenz (ah) anthracene	0.001		nd	nd
7,12-Dimethylbenz(a)anthracene	0.001		nd	nd
Fluoranthene	0.001		nd	nd
Fluorene	0.001		nd	nd
Indeno (1,2,3-cd) pyrene	0.001		nd	nd
3-Methylcholanthrene	0.001		nd	nd
Naphthalene	0.001		nd	nd
Phenanthrene	0.001		nd	nd
Pyrene	0.001		nd	nd



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: VOC Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004044	2010004045	2010004046	2010004047	2010004048	2010004049	2010004050
		Client ID	Site 1-0A	Site 1-1A	Site 1-3B	Site 1-5A	Site 1-10B	Site 1-15A	Site 2-0A
Dibromofluoromethane			80	129	122	119	121	120	109
12-Dichloroethane-d4			90	125	117	112	127	115	104
Toluene-d8			71	107	97	100	127	104	96
p-Bromofluorobenzene			64	93	84	92	110	90	86

Matrix: Water

Method: VOC Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004051	2010004052	2010004053	2010004054	2010004055	2010004056	2010004057
		Client ID	Site 2-3A	Site 2-5A	Site 2-10A	Site 4-0A	Site 4-1A	Site 4-3A	Site 4-5A
Dibromofluoromethane			118	112	113	120	117	115	120
12-Dichloroethane-d4			114	110	112	117	115	114	117
Toluene-d8			105	83	94	103	100	93	101
p-Bromofluorobenzene			93	74	85	92	88	81	89



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: VOC Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004058	2010004059	2010004060	2010004061	2010004062	2010004063	2010004064
		Client ID	Site 4-10A	Site 4-15A	Site 5-0A	Site 5-1B	Site 5-3A	Site 5-5B	Site 5-10A
Dibromofluoromethane			120	120	117	119	109	118	114
12-Dichloroethane-d4			116	117	115	118	109	118	114
Toluene-d8			103	100	98	99	95	97	92
p-Bromofluorobenzene			91	85	85	86	86	87	83

Matrix: Water

Method: VOC Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004065	2010004066	2010004067	2010004068	2010004069	2010004070	2010004071
		Client ID	Site 5-15A	Site 6-0B	Site 6-1B	Site 6-3A	Site 6-5B	Site 6-10A	Site 6-15A
Dibromofluoromethane			108	117	117	116	108	103	112
12-Dichloroethane-d4			107	119	117	119	113	111	119
Toluene-d8			95	89	103	92	86	72	84
p-Bromofluorobenzene			80	79	93	84	77	62	72



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: VOC Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004072	2010004073	2010004074	2010004075	2010004076	2010004077
		Client ID	Site 7-0A	Site 7-1A	Site 7-3A	Site 7-5A	Site 7-10A	Site 7-15A
Dibromofluoromethane			97	103	107	107	116	111
12-Dichloroethane-d4			117	108	114	115	126	117
Toluene-d8			64	93	82	81	91	92
p-Bromofluorobenzene			74	82	70	73	80	81

Matrix: Water

Method: VOC Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004124	2010004125	2010004126
		Client ID	Method	Method	Method
			Blank	Blank	Blank
Dibromofluoromethane			95	106	112
12-Dichloroethane-d4			98	94	100
Toluene-d8			86	80	88
p-Bromofluorobenzene			79	70	77



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004044	2010004045	2010004046	2010004047	2010004048	2010004049	2010004050
		Client ID	Site 1-0A	Site 1-1A	Site 1-3B	Site 1-5A	Site 1-10B	Site 1-15A	Site 2-0A
Fluorobiphenyl			112	113	100	102	112	110	105
Fluorophenol			74	74	70	80	76	76	80
Nitrobenzene-d5			121	116	113	114	109	105	96
Phenol-d6			60	62	70	65	61	61	76
p-Terphenyl-d14			109	108	125	124	104	102	100
2,4,6-Tribromophenol			86	68	82	81	75	72	70

Matrix: Water

Method: Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004051	2010004052	2010004053	2010004054	2010004055	2010004056	2010004057
		Client ID	Site 2-3A	Site 2-5A	Site 2-10A	Site 4-0A	Site 4-1A	Site 4-3A	Site 4-5A
Fluorobiphenyl			114	119	104	114	92	100	114
Fluorophenol			87	83	86	94	78	76	80
Nitrobenzene-d5			109	118	93	94	100	94	112
Phenol-d6			68	64	62	69	65	69	65
p-Terphenyl-d14			115	112	102	110	119	96	118
2,4,6-Tribromophenol			81	76	66	89	75	64	73



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004058	2010004059	2010004060	2010004061	2010004062	2010004063	2010004064
		Client ID	Site 4-10A	Site 4-15A	Site 5-0A	Site 5-1B	Site 5-3A	Site 5-5B	Site 5-10A
Fluorobiphenyl			121	111	114	108	114	106	112
Fluorophenol			78	70	78	77	83	76	69
Nitrobenzene-d5			116	95	120	113	118	87	101
Phenol-d6			60	62	60	61	63	61	60
p-Terphenyl-d14			107	120	116	105	122	113	108
2,4,6-Tribromophenol			80	69	84	86	76	68	70

Matrix: Water

Method: Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004065	2010004066	2010004067	2010004068	2010004069	2010004070	2010004071
		Client ID	Site 5-15A	Site 6-0B	Site 6-1B	Site 6-3A	Site 6-5B	Site 6-10A	Site 6-15A
Fluorobiphenyl			114	99	106	111	120	112	123
Fluorophenol			72	69	68	74	71	78	74
Nitrobenzene-d5			102	103	96	104	111	100	107
Phenol-d6			72	61	60	62	63	62	61
p-Terphenyl-d14			114	101	113	117	108	126	123
2,4,6-Tribromophenol			77	71	70	76	80	81	91



(I) RESULTS

Report N°: M100344R1

Matrix: Water

Method: Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004072	2010004073	2010004074	2010004075	2010004076	2010004077
		Client ID	Site 7-0A	Site 7-1A	Site 7-3A	Site 7-5A	Site 7-10A	Site 7-15A
Fluorobiphenyl			121	116	106	103	99	105
Fluorophenol			76	82	76	78	76	76
Nitrobenzene-d5			119	106	96	89	92	100
Phenol-d6			62	65	68	68	65	69
p-Terphenyl-d14			114	102	117	107	115	109
2,4,6-Tribromophenol			76	67	70	68	64	65

Matrix: Water

Method: Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010004124	2010004125
		Client ID	Method	Method
			Blank	Blank
Fluorobiphenyl			118	92
Fluorophenol			74	65
Nitrobenzene-d5			127	104
Phenol-d6			68	65
p-Terphenyl-d14			122	89
2,4,6-Tribromophenol			77	70



(II) QUALITY CONTROL

Report N°: M100344R1

Matrix: Water

Method: MA-10.WW.01 Monoaromatic Hydrocarbons

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	Leeder ID	2010004128	2010004129	2010004130	2010004131
	Client ID	Method	Method	Method	Method
	PQL	Spike	Spike Dup	Spike	Spike Dup
Benzene		105	107	72	74
Ethylbenzene		97	103	73	75
Toluene		101	105	69	72

Matrix: Water

Method: MA-30.WW.01 Total Petroleum Hydrocarbons

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	Leeder ID	2010004128	2010004129	2010004130	2010004131
	Client ID	Method	Method	Method	Method
	PQL	Spike	Spike Dup	Spike	Spike Dup
Total C6-C36		122	117	121	119



(II) QUALITY CONTROL

Report N°: M100344R1

Matrix: Water

Method: MA-72.WW.01 Polynuclear Aromatic Hydrocarbons

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leeder ID	2010004128	2010004129	2010004130	2010004131
		Client ID	Method	Method	Method	Method
			Spike	Spike Dup	Spike	Spike Dup
Acenaphthene			115	116	115	113
Pyrene			102	102	113	100

Matrix: Water

Method: VOC Surrogate Recovery

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leeder ID	2010004128	2010004129	2010004130	2010004131
		Client ID	Method	Method	Method	Method
			Spike	Spike Dup	Spike	Spike Dup
Dibromofluoromethane			103	99	98	101
12-Dichloroethane-d4			103	99	85	91
Toluene-d8			101	98	92	94
p-Bromofluorobenzene			97	93	76	77



(II) QUALITY CONTROL

Report N°: M100344R1

Matrix: Water

Method: Surrogate Recovery

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leeder ID	2010004128	2010004129	2010004130	2010004131
		Client ID	Method	Method	Method	Method
			Spike	Spike Dup	Spike	Spike Dup
Fluorobiphenyl			122	122	97	110
Fluorophenol			67	77	70	66
Nitrobenzene-d5			112	99	95	112
Phenol-d6			64	66	66	66
p-Terphenyl-d14			124	122	117	114
2,4,6-Tribromophenol			88	102	75	61



QUALIFIERS / NOTES FOR REPORTED RESULTS

PQL	Practical Quantitation Limit
<i>is</i>	Insufficient Sample to perform this analysis.
T	Tentative identification based on computer library search of mass spectra.
ND	Not Detected – The analyte was not detected above the reported PQL.
<i>nr</i>	Not Requested for analysis.
R	Rejected Result – results for this analysis failed QC checks.
SQ	Semi-Quantitative result – quantitation based on a generic response factor for this class of analyte.
IM	Inappropriate method of analysis for this compound
U	Unable to provide Quality Control data – high levels of compounds in sample interfered with analysis of QC results.
UF	Unable to provide Quality Control data- Surrogates failed QC checks due to sample matrix effects
UI	Unable to provide Quality Control data – insufficient sample to perform QC checks.
B	This analyte also detected in analysis of the Method Blank.
D	Deviation from standard method – see notes for specific explanation.
L	Analyte detected at a level above the linear response of calibration curve.
NT	No blank sorbent tubes provided for QC analysis.
C1	These compounds co-elute.
C2	These compounds co-elute.



**LEEDER
CONSULTING**

APPENDIX ONE.

CHAIN OF CUSTODY DOCUMENT

Appendix F – Results of Sediment Analysis (Phase III)



**LEEDER
CONSULTING**

A.B.N. 540 864 910 09
4 - 5, 18 Redland Drive
Mitcham, Vic, 3132
Telephone: (03) 9874 1988
Fax: (03) 9874 1933

Chartered Chemists

15-Dec-2010

REPORT NUMBER: M101702

Site/Client Ref: West Atlas

PTTEP Australasia

Level 1

162 Colins Street

West Perth

Western Australia 6005

Attention: Eleanor Stoney

CERTIFICATE OF ANALYSIS

SAMPLES: Fifty-seven samples were received for analysis

DATE RECEIVED: **24-Nov-2010**

DATE COMMENCED: **7-Dec-2010**

METHODS: See Attached Results

RESULTS: Please refer to attached pages for results.

Note: Results are based on samples as received at Leeder Consulting's laboratories

REPORTED BY:

Adam Atkinson

Laboratory Manager



NATA Accredited Laboratory Number: 14429

**This Document is issued in accordance with
NATA's accreditation requirements**

**Accredited for compliance
with ISO/IEC 17025.**



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

		Leeder ID	2010021208	2010021209	2010021210	2010021211	2010021212	2010021213	2010021214
		Client ID	S1 12° 43.9 S 124° 34.04 E	S2 12° 44.1 S 124° 32.9 E	S3 12° 44.12 S 124° 31.8 E	S4 12° 43.9 S 124° 30.65 E	S5 12° 43.08 S 124° 29.59 E	S6 12° 43.1 S 124° 30.67 E	S7 12° 43.09 S 124° 31.81 E
Analyte Name	PQL								
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

		Leeder ID	2010021215	2010021216	2010021217	2010021218	2010021219	2010021220	2010021221
		Client ID	S8 12° 43.14 S 124° 32.86 E	S9 12° 43.09 S 124° 33.98 E	S10 12° 43.09 S 124° 35.15 E	S11 12° 41.99 S 124° 36.1 E	S12 12° 42.03 S 124° 35.06 E	S13 12° 42.07 S 124° 34.03 E	S14 12° 42.09 S 124° 32.94 E
Analyte Name	PQL								
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-10.SD.01 Monoaromatic Hydrocarbons
Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021222	2010021223	2010021224	2010021225	2010021226	2010021227	2010021228
		Client ID	S15 12° 42.14 S 124° 31.92 E	S16 12° 42.03 S 124° 30.79 E	S17 12° 42.02 S 124° 29.64 E	S18 12° 41.93 S 124° 28.75 E	S19 12° 40.86 S 124° 28.67 E	S20 12° 40.78 S 124° 29.67 E	S21 12° 40.84 S 124° 30.78 E
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment
Method: MA-10.SD.01 Monoaromatic Hydrocarbons
Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021229	2010021230	2010021231	2010021232	2010021233	2010021234	2010021235
		Client ID	S22 12° 40.9 S 124° 31.84 E	S23 12° 40.87 S 124° 32.9 E	S24 12° 40.84 S 124° 34.03 E	S25 12° 40.85 S 124° 35.1 E	S26 12° 40.84 S 124° 36.2 E	S27 12° 39.66 S 124° 36.26 E	S28 12° 39.75 S 124° 35.1 E
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021236	2010021237	2010021238	2010021239	2010021240	2010021241	2010021242
		Client ID	S29 12° 39.74 S 124° 33.97 E	S30 12° 39.81 S 124° 32.87 E	S31 12° 39.88 S 124° 31.78 E	S32 12° 39.91 S 124° 30.73 E	S33 12° 39.82 S 124° 29.61 E	S34 12° 39.83 S 124° 28.54 E	S35 12° 38.71 S 124° 28.66 E
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021243	2010021244	2010021245	2010021246	2010021247	2010021248	2010021249
		Client ID	S36 12° 38.68 S 124° 29.65 E	S37 12° 38.71 S 124° 30.76 E	S38 12° 38.75 S 124° 31.9 E	S39 12° 38.78 S 124° 32.93 E	S40 12° 38.76 S 124° 34.01 E	S41 12° 38.75 S 124° 35.19 E	S42 12° 38.72 S 124° 36.14 E
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021250	2010021251	2010021252	2010021253	2010021254	2010021255	2010021256
		Client ID	S43 12° 37.61 S 124° 35.18 E	S44 12° 37.61 S 124° 34.07 E	S45 12° 37.66 S 124° 32.99 E	S46 12° 37.61 S 124° 31.93 E	S47 12° 37.54 S 124° 30.75 E	S48 12° 37.58 S 124° 29.67 E	S49 12° 36.66 S 124° 30.82 E
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021257	2010021258	2010021259	2010021260	2010021261	2010021262	2010021263
		Client ID	S50 12° 36.6 S 124° 31.92 E	S51 12° 36.56 S 124° 32.91 E	S52 12° 36.63 S 124° 33.92 E	Site 1	Site 4	Site 5	Site 8
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

		Leeder ID	2010021264
		Client ID	Site 9
Analyte Name	PQL		
Benzene	0.5		nd
Ethylbenzene	0.5		nd
Toluene	0.5		nd
m&p-Xylenes	0.5		nd
o-Xylene	0.5		nd

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

		Leeder ID	2010021322
		Client ID	S1 12° 43.9 S 124° 34.04 E
Analyte Name	PQL		Duplicate
Benzene	0.5		nd
Ethylbenzene	0.5		nd
Toluene	0.5		nd
m&p-Xylenes	0.5		nd
o-Xylene	0.5		nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021323	2010021324	2010021325	2010021805	2010021806	2010021807	2010021808
		Client ID	S11 12° 41.99 S 124° 36.1 E	S21 12° 40.84 S 124° 30.78 E	S31 12° 39.88 S 124° 31.78 E	S41 12° 38.75 S 124° 35.19 E	S51 12° 36.56 S 124° 32.91 E	Method	Method
		PQL	Duplicate	Duplicate	Duplicate	Duplicate	Duplicate	Blank	Blank
Benzene	0.5		nd	nd	nd	nd	nd	nd	nd
Ethylbenzene	0.5		nd	nd	nd	nd	nd	nd	nd
Toluene	0.5		nd	nd	nd	nd	nd	nd	nd
m&p-Xylenes	0.5		nd	nd	nd	nd	nd	nd	nd
o-Xylene	0.5		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021809
		Client ID	Method
		PQL	Blank
Benzene	0.5		nd
Ethylbenzene	0.5		nd
Toluene	0.5		nd
m&p-Xylenes	0.5		nd
o-Xylene	0.5		nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021208	2010021209	2010021210	2010021211	2010021212	2010021213	2010021214
		Client ID	S1 12° 43.9 S 124° 34.04 E	S2 12° 44.1 S 124° 32.9 E	S3 12° 44.12 S 124° 31.8 E	S4 12° 43.9 S 124° 30.65 E	S5 12° 43.08 S 124° 29.59 E	S6 12° 43.1 S 124° 30.67 E	S7 12° 43.09 S 124° 31.81 E
Analyte Name	PQL								
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021215	2010021216	2010021217	2010021218	2010021219	2010021220	2010021221
		Client ID	S8 12° 43.14 S 124° 32.86 E	S9 12° 43.09 S 124° 33.98 E	S10 12° 43.09 S 124° 35.15 E	S11 12° 41.99 S 124° 36.1 E	S12 12° 42.03 S 124° 35.06 E	S13 12° 42.07 S 124° 34.03 E	S14 12° 42.09 S 124° 32.94 E
Analyte Name	PQL								
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021222	2010021223	2010021224	2010021225	2010021226	2010021227	2010021228
		Client ID	S15 12° 42.14 S 124° 31.92 E	S16 12° 42.03 S 124° 30.79 E	S17 12° 42.02 S 124° 29.64 E	S18 12° 41.93 S 124° 28.75 E	S19 12° 40.86 S 124° 28.67 E	S20 12° 40.78 S 124° 29.67 E	S21 12° 40.84 S 124° 30.78 E
Analyte Name	PQL								
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021229	2010021230	2010021231	2010021232	2010021233	2010021234	2010021235
		Client ID	S22 12° 40.9 S 124° 31.84 E	S23 12° 40.87 S 124° 32.9 E	S24 12° 40.84 S 124° 34.03 E	S25 12° 40.85 S 124° 35.1 E	S26 12° 40.84 S 124° 36.2 E	S27 12° 39.66 S 124° 36.26 E	S28 12° 39.75 S 124° 35.1 E
Analyte Name	PQL								
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021236	2010021237	2010021238	2010021239	2010021240	2010021241	2010021242
		Client ID	S29 12° 39.74 S 124° 33.97 E	S30 12° 39.81 S 124° 32.87 E	S31 12° 39.88 S 124° 31.78 E	S32 12° 39.91 S 124° 30.73 E	S33 12° 39.82 S 124° 29.61 E	S34 12° 39.83 S 124° 28.54 E	S35 12° 38.71 S 124° 28.66 E
Analyte Name	PQL								
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021243	2010021244	2010021245	2010021246	2010021247	2010021248	2010021249
		Client ID	S36 12° 38.68 S 124° 29.65 E	S37 12° 38.71 S 124° 30.76 E	S38 12° 38.75 S 124° 31.9 E	S39 12° 38.78 S 124° 32.93 E	S40 12° 38.76 S 124° 34.01 E	S41 12° 38.75 S 124° 35.19 E	S42 12° 38.72 S 124° 36.14 E
Analyte Name	PQL								
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021250	2010021251	2010021252	2010021253	2010021254	2010021255	2010021256
		Client ID	S43 12° 37.61 S 124° 35.18 E	S44 12° 37.61 S 124° 34.07 E	S45 12° 37.66 S 124° 32.99 E	S46 12° 37.61 S 124° 31.93 E	S47 12° 37.54 S 124° 30.75 E	S48 12° 37.58 S 124° 29.67 E	S49 12° 36.66 S 124° 30.82 E
Analyte Name	PQL								
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021257	2010021258	2010021259	2010021260	2010021261	2010021262	2010021263
		Client ID	S50 12° 36.6 S 124° 31.92 E	S51 12° 36.56 S 124° 32.91 E	S52 12° 36.63 S 124° 33.92 E	Site 1	Site 4	Site 5	Site 8
Analyte Name	PQL								
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: MA-30.SD.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/kg

		Leeder ID	2010021264
		Client ID	Site 9
Analyte Name	PQL		
C6-C9	10		nd
C10-C14	10		nd
C15-C28	20		nd
C29-C36	20		nd
Total C6-C36	20		nd

Matrix: Sediment

Method: MA-30.SD.01 Total Petroleum Hydrocarbons

Sample units are expressed in mg/kg

		Leeder ID	2010021322
		Client ID	S1 12° 43.9 S 124° 34.04 E
Analyte Name	PQL		Duplicate
C6-C9	10		nd
C10-C14	10		nd
C15-C28	20		nd
C29-C36	20		nd
Total C6-C36	20		nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021323	2010021324	2010021325	2010021805	2010021806	2010021807	2010021808
		Client ID	S11 12° 41.99 S 124° 36.1 E	S21 12° 40.84 S 124° 30.78 E	S31 12° 39.88 S 124° 31.78 E	S41 12° 38.75 S 124° 35.19 E	S51 12° 36.56 S 124° 32.91 E	Method	Method
			Duplicate	Duplicate	Duplicate	Duplicate	Duplicate	Blank	Blank
C6-C9	10		nd	nd	nd	nd	nd	nd	nd
C10-C14	10		nd	nd	nd	nd	nd	nd	nd
C15-C28	20		nd	nd	nd	nd	nd	nd	nd
C29-C36	20		nd	nd	nd	nd	nd	nd	nd
Total C6-C36	20		nd	nd	nd	nd	nd	nd	nd

Matrix: Sediment
Method: MA-30.SD.01 Total Petroleum Hydrocarbons
Sample units are expressed in mg/kg

Analyte Name	PQL	Leeder ID	2010021809
		Client ID	Method
			Blank
C6-C9	10		nd
C10-C14	10		nd
C15-C28	20		nd
C29-C36	20		nd
Total C6-C36	20		nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021208	2010021209	2010021210	2010021211	2010021212	2010021213	2010021214
		Client ID	S1 12° 43.9 S 124° 34.04 E	S2 12° 44.1 S 124° 32.9 E	S3 12° 44.12 S 124° 31.8 E	S4 12° 43.9 S 124° 30.65 E	S5 12° 43.08 S 124° 29.59 E	S6 12° 43.1 S 124° 30.67 E	S7 12° 43.09 S 124° 31.81 E
Analyte Name	PQL								
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021215	2010021216	2010021217	2010021218	2010021219	2010021220	2010021221
		Client ID	S8 12° 43.14 S 124° 32.86 E	S9 12° 43.09 S 124° 33.98 E	S10 12° 43.09 S 124° 35.15 E	S11 12° 41.99 S 124° 36.1 E	S12 12° 42.03 S 124° 35.06 E	S13 12° 42.07 S 124° 34.03 E	S14 12° 42.09 S 124° 32.94 E
Analyte Name	PQL								
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021222	2010021223	2010021224	2010021225	2010021226	2010021227	2010021228
		Client ID	S15 12° 42.14 S 124° 31.92 E	S16 12° 42.03 S 124° 30.79 E	S17 12° 42.02 S 124° 29.64 E	S18 12° 41.93 S 124° 28.75 E	S19 12° 40.86 S 124° 28.67 E	S20 12° 40.78 S 124° 29.67 E	S21 12° 40.84 S 124° 30.78 E
Analyte Name	PQL								
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021229	2010021230	2010021231	2010021232	2010021233	2010021234	2010021235
		Client ID	S22 12° 40.9 S 124° 31.84 E	S23 12° 40.87 S 124° 32.9 E	S24 12° 40.84 S 124° 34.03 E	S25 12° 40.85 S 124° 35.1 E	S26 12° 40.84 S 124° 36.2 E	S27 12° 39.66 S 124° 36.26 E	S28 12° 39.75 S 124° 35.1 E
Analyte Name	PQL								
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021236	2010021237	2010021238	2010021239	2010021240	2010021241	2010021242
		Client ID	S29 12° 39.74 S 124° 33.97 E	S30 12° 39.81 S 124° 32.87 E	S31 12° 39.88 S 124° 31.78 E	S32 12° 39.91 S 124° 30.73 E	S33 12° 39.82 S 124° 29.61 E	S34 12° 39.83 S 124° 28.54 E	S35 12° 38.71 S 124° 28.66 E
Analyte Name	PQL								
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021243	2010021244	2010021245	2010021246	2010021247	2010021248	2010021249
		Client ID	S36 12° 38.68 S 124° 29.65 E	S37 12° 38.71 S 124° 30.76 E	S38 12° 38.75 S 124° 31.9 E	S39 12° 38.78 S 124° 32.93 E	S40 12° 38.76 S 124° 34.01 E	S41 12° 38.75 S 124° 35.19 E	S42 12° 38.72 S 124° 36.14 E
Analyte Name	PQL								
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021250	2010021251	2010021252	2010021253	2010021254	2010021255	2010021256
		Client ID	S43 12° 37.61 S 124° 35.18 E	S44 12° 37.61 S 124° 34.07 E	S45 12° 37.66 S 124° 32.99 E	S46 12° 37.61 S 124° 31.93 E	S47 12° 37.54 S 124° 30.75 E	S48 12° 37.58 S 124° 29.67 E	S49 12° 36.66 S 124° 30.82 E
Analyte Name	PQL								
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021257	2010021258	2010021259	2010021260	2010021261	2010021262	2010021263
		Client ID	S50 12° 36.6 S 124° 31.92 E	S51 12° 36.56 S 124° 32.91 E	S52 12° 36.63 S 124° 33.92 E	Site 1	Site 4	Site 5	Site 8
Analyte Name		PQL							
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons

Sample units are expressed in mg/kg

Leeder ID	2010021264
Client ID	Site 9
Analyte Name	PQL

Acenaphthene	0.5	nd
Acenaphthylene	0.5	nd
Anthracene	0.5	nd
Benzo(a)anthracene	0.5	nd
Benzo (a) pyrene	0.5	nd
Benzo (b) fluoranthene	0.5	nd
Benzo (ghi) perylene	0.5	nd
Benzo (k) fluoranthene	0.5	nd
Chrysene	0.5	nd
Dibenz (ah) anthracene	0.5	nd
7,12-Dimethylbenz(a)anthracene	0.5	nd
Fluoranthene	0.5	nd
Fluorene	0.5	nd
Indeno (1,2,3-cd) pyrene	0.5	nd
3-Methylcholanthrene	0.5	nd
Naphthalene	0.5	nd
Phenanthrene	0.5	nd
Pyrene	0.5	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons

Sample units are expressed in mg/kg

Leeder ID	2010021322
Client ID	S1 12° 43.9 S 124° 34.04 E
Analyte Name	PQL Duplicate

Acenaphthene	0.5	nd
Acenaphthylene	0.5	nd
Anthracene	0.5	nd
Benzo(a)anthracene	0.5	nd
Benzo (a) pyrene	0.5	nd
Benzo (b) fluoranthene	0.5	nd
Benzo (ghi) perylene	0.5	nd
Benzo (k) fluoranthene	0.5	nd
Chrysene	0.5	nd
Dibenz (ah) anthracene	0.5	nd
7,12-Dimethylbenz(a)anthracene	0.5	nd
Fluoranthene	0.5	nd
Fluorene	0.5	nd
Indeno (1,2,3-cd) pyrene	0.5	nd
3-Methylcholanthrene	0.5	nd
Naphthalene	0.5	nd
Phenanthrene	0.5	nd
Pyrene	0.5	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID	2010021323	2010021324	2010021325	2010021805	2010021806	2010021807	2010021808
		Client ID	S11 12° 41.99 S 124° 36.1 E	S21 12° 40.84 S 124° 30.78 E	S31 12° 39.88 S 124° 31.78 E	S41 12° 38.75 S 124° 35.19 E	S51 12° 36.56 S 124° 32.91 E	Method	Method
Analyte Name		PQL	Duplicate	Duplicate	Duplicate	Duplicate	Duplicate	Blank	Blank
Acenaphthene	0.5		nd	nd	nd	nd	nd	nd	nd
Acenaphthylene	0.5		nd	nd	nd	nd	nd	nd	nd
Anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (a) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (b) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (ghi) perylene	0.5		nd	nd	nd	nd	nd	nd	nd
Benzo (k) fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Chrysene	0.5		nd	nd	nd	nd	nd	nd	nd
Dibenz (ah) anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
7,12-Dimethylbenz(a)anthracene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluoranthene	0.5		nd	nd	nd	nd	nd	nd	nd
Fluorene	0.5		nd	nd	nd	nd	nd	nd	nd
Indeno (1,2,3-cd) pyrene	0.5		nd	nd	nd	nd	nd	nd	nd
3-Methylcholanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Naphthalene	0.5		nd	nd	nd	nd	nd	nd	nd
Phenanthrene	0.5		nd	nd	nd	nd	nd	nd	nd
Pyrene	0.5		nd	nd	nd	nd	nd	nd	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons
Sample units are expressed in mg/kg

		Leeder ID
		Client ID
		Method
		Blank
Analyte Name	PQL	
Acenaphthene	0.5	nd
Acenaphthylene	0.5	nd
Anthracene	0.5	nd
Benzo(a)anthracene	0.5	nd
Benzo (a) pyrene	0.5	nd
Benzo (b) fluoranthene	0.5	nd
Benzo (ghi) perylene	0.5	nd
Benzo (k) fluoranthene	0.5	nd
Chrysene	0.5	nd
Dibenz (ah) anthracene	0.5	nd
7,12-Dimethylbenz(a)anthracene	0.5	nd
Fluoranthene	0.5	nd
Fluorene	0.5	nd
Indeno (1,2,3-cd) pyrene	0.5	nd
3-Methylcholanthrene	0.5	nd
Naphthalene	0.5	nd
Phenanthrene	0.5	nd
Pyrene	0.5	nd



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: VOC Surrogate Recovery

Sample units are expressed in %

		Leeder ID	2010021208	2010021209	2010021210	2010021211	2010021212	2010021213	2010021214
		Client ID	S1 12° 43.9 S 124° 34.04 E	S2 12° 44.1 S 124° 32.9 E	S3 12° 44.12 S 124° 31.8 E	S4 12° 43.9 S 124° 30.65 E	S5 12° 43.08 S 124° 29.59 E	S6 12° 43.1 S 124° 30.67 E	S7 12° 43.09 S 124° 31.81 E
Analyte Name		PQL							
Dibromofluoromethane			75	69	75	78	87	80	82
12-Dichloroethane-d4			77	86	86	94	97	93	102
Toluene-d8			75	73	74	64	72	71	76
p-Bromofluorobenzene			73	71	67	70	66	62	68

Matrix: Sediment

Method: VOC Surrogate Recovery

Sample units are expressed in %

		Leeder ID	2010021215	2010021216	2010021217	2010021218	2010021219	2010021220	2010021221
		Client ID	S8 12° 43.14 S 124° 32.86 E	S9 12° 43.09 S 124° 33.98 E	S10 12° 43.09 S 124° 35.15 E	S11 12° 41.99 S 124° 36.1 E	S12 12° 42.03 S 124° 35.06 E	S13 12° 42.07 S 124° 34.03 E	S14 12° 42.09 S 124° 32.94 E
Analyte Name		PQL							
Dibromofluoromethane			69	81	73	74	68	81	71
12-Dichloroethane-d4			87	97	87	94	98	94	98
Toluene-d8			62	68	64	76	71	87	64
p-Bromofluorobenzene			65	74	66	75	69	66	67



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: VOC Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021222	2010021223	2010021224	2010021225	2010021226	2010021227	2010021228
		Client ID	S15 12° 42.14 S 124° 31.92 E	S16 12° 42.03 S 124° 30.79 E	S17 12° 42.02 S 124° 29.64 E	S18 12° 41.93 S 124° 28.75 E	S19 12° 40.86 S 124° 28.67 E	S20 12° 40.78 S 124° 29.67 E	S21 12° 40.84 S 124° 30.78 E
Analyte Name		PQL							
Dibromofluoromethane			75	78	79	80	81	75	84
12-Dichloroethane-d4			88	88	90	98	100	87	105
Toluene-d8			64	62	68	74	66	74	70
p-Bromofluorobenzene			72	62	69	72	70	67	72

Matrix: Sediment
Method: VOC Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021229	2010021230	2010021231	2010021232	2010021233	2010021234	2010021235
		Client ID	S22 12° 40.9 S 124° 31.84 E	S23 12° 40.87 S 124° 32.9 E	S24 12° 40.84 S 124° 34.03 E	S25 12° 40.85 S 124° 35.1 E	S26 12° 40.84 S 124° 36.2 E	S27 12° 39.66 S 124° 36.26 E	S28 12° 39.75 S 124° 35.1 E
Analyte Name		PQL							
Dibromofluoromethane			68	63	75	72	69	71	71
12-Dichloroethane-d4			95	74	95	85	91	92	88
Toluene-d8			73	76	69	67	70	69	70
p-Bromofluorobenzene			66	74	74	65	66	71	67



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: VOC Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021236	2010021237	2010021238	2010021239	2010021240	2010021241	2010021242
		Client ID	S29 12° 39.74 S 124° 33.97 E	S30 12° 39.81 S 124° 32.87 E	S31 12° 39.88 S 124° 31.78 E	S32 12° 39.91 S 124° 30.73 E	S33 12° 39.82 S 124° 29.61 E	S34 12° 39.83 S 124° 28.54 E	S35 12° 38.71 S 124° 28.66 E
Analyte Name		PQL							
Dibromofluoromethane			75	81	66	80	82	79	72
12-Dichloroethane-d4			93	108	90	106	104	101	100
Toluene-d8			73	76	64	82	68	70	66
p-Bromofluorobenzene			70	70	65	72	63	63	65

Matrix: Sediment
Method: VOC Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021243	2010021244	2010021245	2010021246	2010021247	2010021248	2010021249
		Client ID	S36 12° 38.68 S 124° 29.65 E	S37 12° 38.71 S 124° 30.76 E	S38 12° 38.75 S 124° 31.9 E	S39 12° 38.78 S 124° 32.93 E	S40 12° 38.76 S 124° 34.01 E	S41 12° 38.75 S 124° 35.19 E	S42 12° 38.72 S 124° 36.14 E
Analyte Name		PQL							
Dibromofluoromethane			87	72	81	80	66	79	77
12-Dichloroethane-d4			118	101	105	106	78	108	99
Toluene-d8			69	70	65	66	66	77	73
p-Bromofluorobenzene			66	64	66	73	67	78	69



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: VOC Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010021250	2010021251	2010021252	2010021253	2010021254	2010021255	2010021256
		Client ID	S43 12° 37.61 S 124° 35.18 E	S44 12° 37.61 S 124° 34.07 E	S45 12° 37.66 S 124° 32.99 E	S46 12° 37.61 S 124° 31.93 E	S47 12° 37.54 S 124° 30.75 E	S48 12° 37.58 S 124° 29.67 E	S49 12° 36.66 S 124° 30.82 E
Dibromofluoromethane			93	75	80	77	93	107	85
12-Dichloroethane-d4			107	92	98	115	120	126	99
Toluene-d8			74	65	71	72	85	91	76
p-Bromofluorobenzene			71	64	71	63	66	65	76

Matrix: Sediment

Method: VOC Surrogate Recovery

Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010021257	2010021258	2010021259	2010021260	2010021261	2010021262	2010021263
		Client ID	S50 12° 36.6 S 124° 31.92 E	S51 12° 36.56 S 124° 32.91 E	S52 12° 36.63 S 124° 33.92 E	Site 1	Site 4	Site 5	Site 8
Dibromofluoromethane			74	72	89	93	85	74	73
12-Dichloroethane-d4			104	99	116	118	117	108	100
Toluene-d8			62	70	87	72	69	62	70
p-Bromofluorobenzene			65	76	70	69	65	69	69



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: VOC Surrogate Recovery

Sample units are expressed in %

		Leeder ID	2010021264
		Client ID	Site 9
Analyte Name	PQL		
Dibromofluoromethane			99
12-Dichloroethane-d4			93
Toluene-d8			72
p-Bromofluorobenzene			64

Matrix: Sediment

Method: VOC Surrogate Recovery

Sample units are expressed in %

		Leeder ID	2010021322
		Client ID	S1 12° 43.9 S 124° 34.04 E
Analyte Name	PQL		Duplicate
Dibromofluoromethane			79
12-Dichloroethane-d4			88
Toluene-d8			71
p-Bromofluorobenzene			79



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: VOC Surrogate Recovery
Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010021323	2010021324	2010021325	2010021805	2010021806	2010021807	2010021808
		Client ID	S11 12° 41.99 S 124° 36.1 E	S21 12° 40.84 S 124° 30.78 E	S31 12° 39.88 S 124° 31.78 E	S41 12° 38.75 S 124° 35.19 E	S51 12° 36.56 S 124° 32.91 E	Method	Method
		PQL	Duplicate	Duplicate	Duplicate	Duplicate	Duplicate	Blank	Blank
Dibromofluoromethane			77	71	67	74	93	76	103
12-Dichloroethane-d4			101	92	90	96	124	77	107
Toluene-d8			66	65	66	69	85	77	83
p-Bromofluorobenzene			65	67	68	67	85	73	74

Matrix: Sediment
Method: VOC Surrogate Recovery
Sample units are expressed in %

Analyte Name	PQL	Leeder ID	2010021809
		Client ID	Method
		PQL	Blank
Dibromofluoromethane			78
12-Dichloroethane-d4			90
Toluene-d8			69
p-Bromofluorobenzene			77



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021208	2010021209	2010021210	2010021211	2010021212	2010021213	2010021214
		Client ID	S1 12° 43.9 S 124° 34.04 E	S2 12° 44.1 S 124° 32.9 E	S3 12° 44.12 S 124° 31.8 E	S4 12° 43.9 S 124° 30.65 E	S5 12° 43.08 S 124° 29.59 E	S6 12° 43.1 S 124° 30.67 E	S7 12° 43.09 S 124° 31.81 E
Analyte Name	PQL								
Fluorobiphenyl			110	91	90	96	79	99	88
Fluorophenol			125	92	100	97	83	107	92
Nitrobenzene-d5			104	83	81	87	71	88	76
Phenol-d6			106	87	85	90	75	94	83
p-Terphenyl-d14			114	95	91	97	79	104	94

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021215	2010021216	2010021217	2010021218	2010021219	2010021220	2010021221
		Client ID	S8 12° 43.14 S 124° 32.86 E	S9 12° 43.09 S 124° 33.98 E	S10 12° 43.09 S 124° 35.15 E	S11 12° 41.99 S 124° 36.1 E	S12 12° 42.03 S 124° 35.06 E	S13 12° 42.07 S 124° 34.03 E	S14 12° 42.09 S 124° 32.94 E
Analyte Name	PQL								
Fluorobiphenyl			76	77	85	71	67	75	70
Fluorophenol			78	85	97	79	74	79	79
Nitrobenzene-d5			68	66	75	60	65	66	63
Phenol-d6			71	72	81	65	62	71	66
p-Terphenyl-d14			78	79	90	72	70	78	73



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021222	2010021223	2010021224	2010021225	2010021226	2010021227	2010021228
		Client ID	S15 12° 42.14 S 124° 31.92 E	S16 12° 42.03 S 124° 30.79 E	S17 12° 42.02 S 124° 29.64 E	S18 12° 41.93 S 124° 28.75 E	S19 12° 40.86 S 124° 28.67 E	S20 12° 40.78 S 124° 29.67 E	S21 12° 40.84 S 124° 30.78 E
Analyte Name		PQL							
Fluorobiphenyl			77	77	71	77	75	78	66
Fluorophenol			79	78	72	85	83	89	79
Nitrobenzene-d5			66	65	61	65	63	68	63
Phenol-d6			70	71	64	71	70	74	67
p-Terphenyl-d14			77	77	69	77	76	88	61

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021229	2010021230	2010021231	2010021232	2010021233	2010021234	2010021235
		Client ID	S22 12° 40.9 S 124° 31.84 E	S23 12° 40.87 S 124° 32.9 E	S24 12° 40.84 S 124° 34.03 E	S25 12° 40.85 S 124° 35.1 E	S26 12° 40.84 S 124° 36.2 E	S27 12° 39.66 S 124° 36.26 E	S28 12° 39.75 S 124° 35.1 E
Analyte Name		PQL							
Fluorobiphenyl			63	79	77	101	63	66	67
Fluorophenol			76	94	96	107	67	89	68
Nitrobenzene-d5			62	67	67	92	66	62	68
Phenol-d6			66	79	83	93	62	67	63
p-Terphenyl-d14			71	81	82	103	76	67	72



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021236	2010021237	2010021238	2010021239	2010021240	2010021241	2010021242
		Client ID	S29 12° 39.74 S 124° 33.97 E	S30 12° 39.81 S 124° 32.87 E	S31 12° 39.88 S 124° 31.78 E	S32 12° 39.91 S 124° 30.73 E	S33 12° 39.82 S 124° 29.61 E	S34 12° 39.83 S 124° 28.54 E	S35 12° 38.71 S 124° 28.66 E
Analyte Name		PQL							
Fluorobiphenyl			62	65	80	62	82	85	85
Fluorophenol			71	78	98	71	92	92	90
Nitrobenzene-d5			73	67	68	64	72	70	75
Phenol-d6			65	67	84	63	82	78	81
p-Terphenyl-d14			64	69	85	66	89	92	89

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021243	2010021244	2010021245	2010021246	2010021247	2010021248	2010021249
		Client ID	S36 12° 38.68 S 124° 29.65 E	S37 12° 38.71 S 124° 30.76 E	S38 12° 38.75 S 124° 31.9 E	S39 12° 38.78 S 124° 32.93 E	S40 12° 38.76 S 124° 34.01 E	S41 12° 38.75 S 124° 35.19 E	S42 12° 38.72 S 124° 36.14 E
Analyte Name		PQL							
Fluorobiphenyl			77	75	65	87	88	77	67
Fluorophenol			87	76	72	97	98	77	66
Nitrobenzene-d5			68	62	76	73	74	71	60
Phenol-d6			74	68	61	88	82	76	65
p-Terphenyl-d14			81	79	82	90	91	113	78



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021250	2010021251	2010021252	2010021253	2010021254	2010021255	2010021256
		Client ID	S43 12° 37.61 S 124° 35.18 E	S44 12° 37.61 S 124° 34.07 E	S45 12° 37.66 S 124° 32.99 E	S46 12° 37.61 S 124° 31.93 E	S47 12° 37.54 S 124° 30.75 E	S48 12° 37.58 S 124° 29.67 E	S49 12° 36.66 S 124° 30.82 E
Analyte Name		PQL							
Fluorobiphenyl			82	78	79	73	75	70	83
Fluorophenol			78	76	76	67	72	61	77
Nitrobenzene-d5			74	71	72	65	70	63	74
Phenol-d6			78	75	76	69	73	60	77
p-Terphenyl-d14			89	85	86	81	76	97	90

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021257	2010021258	2010021259	2010021260	2010021261	2010021262	2010021263
		Client ID	S50 12° 36.6 S 124° 31.92 E	S51 12° 36.56 S 124° 32.91 E	S52 12° 36.63 S 124° 33.92 E	Site 1	Site 4	Site 5	Site 8
Analyte Name		PQL							
Fluorobiphenyl			86	69	74	76	67	71	91
Fluorophenol			76	64	70	72	71	61	83
Nitrobenzene-d5			71	61	66	66	60	62	79
Phenol-d6			77	65	69	70	70	60	82
p-Terphenyl-d14			109	77	91	93	77	74	109



(I) RESULTS

Report N°: M101702

Matrix: Sediment

Method: Surrogate Recovery

Sample units are expressed in %

		Leeder ID	2010021264
		Client ID	Site 9
Analyte Name	PQL		
Fluorobiphenyl			70
Fluorophenol			65
Nitrobenzene-d5			62
Phenol-d6			64
p-Terphenyl-d14			106

Matrix: Sediment

Method: Surrogate Recovery

Sample units are expressed in %

		Leeder ID	2010021322
		Client ID	S1 12° 43.9 S 124° 34.04 E
Analyte Name	PQL		Duplicate
Fluorobiphenyl			92
Fluorophenol			95
Nitrobenzene-d5			86
Phenol-d6			87
p-Terphenyl-d14			91



(I) RESULTS

Report N°: M101702

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021323	2010021324	2010021325	2010021805	2010021806	2010021807	2010021808
		Client ID	S11 12° 41.99 S 124° 36.1 E	S21 12° 40.84 S 124° 30.78 E	S31 12° 39.88 S 124° 31.78 E	S41 12° 38.75 S 124° 35.19 E	S51 12° 36.56 S 124° 32.91 E	Method	Method
		PQL	Duplicate	Duplicate	Duplicate	Duplicate	Duplicate	Blank	Blank
Analyte Name									
Fluorobiphenyl			80	62	64	88	92	66	83
Fluorophenol			91	76	69	84	86	74	87
Nitrobenzene-d5			71	70	72	81	82	62	76
Phenol-d6			76	67	60	84	84	63	77
p-Terphenyl-d14			84	62	70	85	103	68	85

Matrix: Sediment
Method: Surrogate Recovery
Sample units are expressed in %

		Leeder ID	2010021809
		Client ID	Method
		PQL	Blank
Analyte Name			
Fluorobiphenyl			80
Fluorophenol			79
Nitrobenzene-d5			72
Phenol-d6			78
p-Terphenyl-d14			97



(II) QUALITY CONTROL

Report N°: M101702

Matrix: Sediment

Method: MA-10.SD.01 Monoaromatic Hydrocarbons

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leeder ID	2010021810	2010021811	2010023115	2010023116	2010023117	2010023118
		Client ID	S1 12° 43.9 S 124° 34.04 E	S1 12° 43.9 S 124° 34.04 E	S21 12° 40.84 S 124° 30.78 E	S21 12° 40.84 S 124° 30.78 E	S41 12° 38.75 S 124° 35.19 E	S41 12° 38.75 S 124° 35.19 E
		PQL	Spike	Spike Dup	Spike	Spike Dup	Spike	Spike Dup
Benzene			79	82	75	83	90	83
Ethylbenzene			86	79	80	76	96	101
Toluene			83	98	109	96	74	78

Matrix: Sediment

Method: MA-30.SD.01 Total Petroleum Hydrocarbons

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leeder ID	2010021810	2010021811	2010023115	2010023116	2010023117	2010023118
		Client ID	S1 12° 43.9 S 124° 34.04 E	S1 12° 43.9 S 124° 34.04 E	S21 12° 40.84 S 124° 30.78 E	S21 12° 40.84 S 124° 30.78 E	S41 12° 38.75 S 124° 35.19 E	S41 12° 38.75 S 124° 35.19 E
		PQL	Spike	Spike Dup	Spike	Spike Dup	Spike	Spike Dup
Total C6-C36			109	90	95	111	92	110



(II) QUALITY CONTROL

Report N°: M101702

Matrix: Sediment

Method: MA-72.SD.01 Polynuclear Aromatic Hydrocarbons

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leeder ID	2010021810	2010021811	2010023115	2010023116	2010023117	2010023118
		Client ID	S1 12° 43.9 S 124° 34.04 E	S1 12° 43.9 S 124° 34.04 E	S21 12° 40.84 S 124° 30.78 E	S21 12° 40.84 S 124° 30.78 E	S41 12° 38.75 S 124° 35.19 E	S41 12° 38.75 S 124° 35.19 E
		PQL	Spike	Spike Dup	Spike	Spike Dup	Spike	Spike Dup
Acenaphthene			68	82	69	74	68	67
Pyrene			66	72	67	65	87	79

Matrix: Sediment

Method: VOC Surrogate Recovery

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leeder ID	2010021810	2010021811	2010023115	2010023116	2010023117	2010023118
		Client ID	S1 12° 43.9 S 124° 34.04 E	S1 12° 43.9 S 124° 34.04 E	S21 12° 40.84 S 124° 30.78 E	S21 12° 40.84 S 124° 30.78 E	S41 12° 38.75 S 124° 35.19 E	S41 12° 38.75 S 124° 35.19 E
		PQL	Spike	Spike Dup	Spike	Spike Dup	Spike	Spike Dup
Dibromofluoromethane			78	87	75	76	84	82
12-Dichloroethane-d4			85	93	95	91	101	103
Toluene-d8			78	87	101	72	72	86
p-Bromofluorobenzene			65	73	70	62	73	80



(II) QUALITY CONTROL

Report N°: M101702

Matrix: Sediment

Method: Surrogate Recovery

Quality Control Results are expressed in Percent Recovery of expected result

Analyte Name	PQL	Leeder ID	2010021810	2010021811	2010023115	2010023116	2010023117	2010023118
		Client ID	S1 12° 43.9 S 124° 34.04 E	S1 12° 43.9 S 124° 34.04 E	S21 12° 40.84 S 124° 30.78 E	S21 12° 40.84 S 124° 30.78 E	S41 12° 38.75 S 124° 35.19 E	S41 12° 38.75 S 124° 35.19 E
			Spike	Spike Dup	Spike	Spike Dup	Spike	Spike Dup
Fluorobiphenyl			107	94	87	65	80	108
Fluorophenol			114	95	98	73	79	105
Nitrobenzene-d5			100	86	81	66	73	77
Phenol-d6			104	88	85	62	81	90
p-Terphenyl-d14			109	97	94	70	111	126



QUALIFIERS / NOTES FOR REPORTED RESULTS

PQL Practical Quantitation Limit

is Insufficient Sample to perform this analysis.

T Tentative identification based on computer library search of mass spectra.

ND Not Detected – The analyte was not detected above the reported PQL.

nr Not Requested for analysis.

R Rejected Result – results for this analysis failed QC checks.

SQ Semi-Quantitative result – quantitation based on a generic response factor for this class of analyte.

IM Inappropriate method of analysis for this compound

U Unable to provide Quality Control data – high levels of compounds in sample interfered with analysis of QC results.

UF Unable to provide Quality Control data- Surrogates failed QC checks due to sample matrix effects

UI Unable to provide Quality Control data – insufficient sample to perform QC checks.

B This analyte also detected in analysis of the Method Blank.

D Deviation from standard method – see notes for specific explanation.

L Analyte detected at a level above the linear response of calibration curve.

NT No blank sorbent tubes provided for QC analysis.

C1 These compounds co-elute.

C2 These compounds co-elute.



**LEEDER
CONSULTING**

APPENDIX ONE.

CHAIN OF CUSTODY DOCUMENT

Chain of Custody Document

Incident No: WEST ATLAS

Sampled By: Leif Cooper

Comments: _____

Sample Description				Date
SITE 8	11° 53' 26 S	125° 34' 45 E	(+dup)	11/11/10 11:35
SITE 9			(+dup)	12/11/10 2:00
S1	12° 43' 90 S	124° 34' 04 E	72m (+dup)	13/11/10 9:20
S2	12° 44' 10 S	124° 32' 90 E	85m	9:53
S3	12° 44' 12 S	124° 31' 80 E	84m	10:11
S4	12° 43' 90 S	124° 30' 65 E	85m	10:30
S5	12° 43' 08 S	124° 29' 59 E	85m	10:49
S6	12° 43' 10 S	124° 30' 67 E	85m	11:10
S7	12° 43' 09 S	124° 31' 81 E	84m	11:32
S8	12° 43' 14 S	124° 32' 86 E	84m	11:52
S9	12° 43' 09 S	124° 33' 98 E	83m	12:12
S10	12° 43' 09 S	124° 35' 15 E	75m	12:31
S11	12° 41' 99 S	124° 36' 10 E	67m	12:50
S12	12° 42' 03 S	124° 35' 06 E	80m - no dup	2:06
S13	12° 42' 07 S	124° 34' 03 E	83m (+dup)	2:35
S14	12° 42' 09 S	124° 32' 94 E	83m	2:55
S15	12° 42' 14 S	124° 31' 92 E	84m	3:15
S16	12° 42' 03 S	124° 30' 71 E	84m	3:37
S17	12° 42' 02 S	124° 29' 64 E	86m	3:58
S18	12° 41' 93 S	124° 28' 75 E	86m	4:16

Name <u>Leif</u>	Signature <u>[Signature]</u>	Date <u>21/11/10</u>
Organisation		Time



Chain of Custody Document

Incident No: WEST ATLAS

Sampled By: Leif Cooper

Comments:

Sample Description					Date
S19	12° 40' 86 S	124° 28' 67 E	83m	(+ dup)	13/11/10 4:35
S20	12° 40' 78 S	124° 29' 67 E	84m		↓ 4:50
S21	12° 40' 84 S	124° 30' 78 E	85m		↓ 5:06
S22	12° 40' 90 S	124° 31' 84 E	82m		14/11/10 8:19
S23	12° 40' 87 S	124° 32' 90 E	75m		8:39
S24	12° 40' 84 S	124° 33' 03 E	75m		8:58
S25	12° 40' 85 S	124° 35' 10 E	63m	- no dup	9:13
S26	12° 40' 84 S	124° 36' 20 E	59m	(+dup)	9:42
S27	12° 39' 66 S	124° 36' 26 E	73m		10:14
S28	12° 39' 75 S	124° 35' 10 E	78m		10:30
S29	12° 39' 74 S	124° 33' 97 E	70m		10:59
S30	12° 39' 81 S	124° 32' 87 E	73m		11:17
S31	12° 39' 88 S	124° 31' 78 E	81m		11:45
S32	12° 39' 91 S	124° 30' 73 E	83m		12:07
S33	12° 39' 82 S	124° 29' 61 E	82m		12:24
S34	12° 39' 83 S	124° 28' 54 E	76m		2:03
S35	12° 38' 71 S	124° 28' 66 E	75m		2:23
S36	12° 38' 68 S	124° 29' 65 E	78m		2:41
S37	12° 38' 71 S	124° 30' 76 E	84m		2:59
S38	12° 38' 75 S	124° 31' 90 E	80m		3:20

Name <u>Leif</u>	Signature <u>[Signature]</u>	Date <u>21/11/10</u>
Organisation		Time

Form-054



Issue Date 1-10-02

Chain of Custody Document

Incident No: West Atlas

Sampled By: Leif Cooper

Comments:

Sample Description						Date	
S39	12° 38' 78 S	124° 32' 93 E	74m	(+ dup)		14/11/10	3:35
S40	12° 38' 76 S	124° 34' 01	71m				3:51
S41	12° 38' 75 S	124° 35' 19	76m				4:08
S42	12° 38' 72 S	124° 36' 14	81m				4:21
S43	12° 37' 61 S	124° 35' 18	68m				4:46
S44	12° 37' 61 S	124° 34' 07	74m				5:10
S45	12° 37' 66 S	124° 32' 99	75m				5:28
S46	12° 37' 61 S	124° 31' 93	74m			↓	5:45
S47	12° 37' 54 S	124° 30' 75	81m			15/11/10	8:58
S48	12° 37' 58 S	124° 29' 67	77m				9:21
S49	12° 36' 66 S	124° 30' 82	79m				9:39
S50	12° 36' 60 S	124° 31' 92	83m				9:59
S51	12° 36' 56 S	124° 32' 91	76m				10:15
S52	12° 36' 63 S	124° 33' 92	76m	↓		↓	10:31
SITE 1	12° 52' 23 S	124° 40' 20 E				16/11	
SITE 4	shewn sample (net)				13° 24' 33 S 124° 09' 90 E	18/11	11:30
SITE 4					13° 24' 40 S 124° 08' 94 E	18/11	1:30

Name	<u>Leif</u>	Signature	<u>[Signature]</u>	Date	<u>21/11/10</u>
Organisation				Time	

Form-054

Issue Date 1-10-02



Erynne McCrorey

From: Leif Cooper
Sent: Monday, 6 December 2010 4:17 PM
To: Lyndall Stevens; Erynne McCrorey
Subject: FW: Sediment testing

Analysis for M101702

Regards,

Leif

From: Stoney, Eleanor [<mailto:Eleanor.Stoney@au.pttep.com>]
Sent: Monday, 6 December 2010 4:15 PM
To: Leif Cooper
Subject: Re: Sediment testing

Hi Leif,

Please analyze for TPH, PAH, and BTEX. To what resolution can we analyze for PAH?

Eleanor Stoney
Environmental Coordinator

On 06/12/2010, at 2:16 PM, "Leif Cooper" <Leif@leederconsulting.com> wrote:

Hi Eleanor,

The samples from the recent sediment sampling program have now arrived at our laboratory. Could we have written confirmation that these sediments are to be analysed for TPH, and PAH if any positives are found?

Many thanks

Regards,

Leif Cooper

Senior Chemist

Leeder Consulting Pty Ltd
5/18 Redland Dve, Mitcham Vic. 3123

T: +61 3 **9874 1988** | M: 0414 993 888 | F: +61 3 **9874 1933** |
www.leederconsulting.com



